# RAINBO FISIE Their











#### Rainbowfishes ~ Their Care & Keeping in Captivity

Adrian R. Tappin



Melanotaenia trifasciata (Cato River, Northern Territory) photo: Dave Wilson

# Rainbowfishes

Their Care & Keeping in Captivity

Adrian R. Tappin

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The aim of this publication is to provide a comprehensive and illustrated guide to the remarkable rainbowfishes found in Australia and New Guinea. The information provided covers topics such as descriptions, habitats, biology, ecology, distribution and their care and keeping in captivity.

This project would not have been possible without the enthusiastic assistance and support of many friends and fellow hobbyists all over the world that gave me direct and indirect help in providing their experience, knowledge and assistance. I owe a great deal of gratitude to the contributing photographers, who graciously shared their photographs and expertise on the pages of this book.

Adrian R. Tappin

"Familiarity will save ecosystems, as the better an ecosystem is known, the less likely it will be destroyed. In the end, we will conserve only what we love, we will love only what we understand, and we will understand only what we are taught." ~ Baba Dioum (1968)

Millions of people all round the world keep fish in ponds and aquaria to enjoy their beauty and their habits as they try to replicate a little piece of nature in their backyard or their living room. For some it is a chance to keep species from the four corners of the globe while for others the lure is for natives of ones own country. While Australia may have fewer species of native freshwater fishes suitable for aquarium life than the traditional tropical aquarium fish suppliers of Asia, Africa and the Americas, what we lack in numbers we make up for in quality and interest.

Currently there is world-wide enthusiasm for aquarium fish from our region – especially for the rainbowfishes and blue-eyes from Australia and New Guinea. Consequently there is a matching desire for information on these fishes from enthusiasts both here and abroad. The information sought concerns not only how to best care for them but also anything and everything about where they come from. To many the ultimate challenge is to research the species, travel to the habitat and capture the specimens, transport them home, look after them well, breed them and make them available to others.

If you want to know anything about these fishes this is the source for you – as comprehensive and authoritative as is currently possible. Adrian Tappin is an acknowledged paramount expert on the Aquariology of this group based on a lifetime of aquarium hobby experience and decades of personal involvement with all aspects of native fish interest. Not only is this the best reference I know of for these fishes, but in my opinion it sets the benchmark for the future when it comes to hobby guides. Adrian knows his stuff and is happy to share his expertise so that the hobbyist can get the most enjoyment and the fish will be cared for optimally.

~ Bruce Hansen

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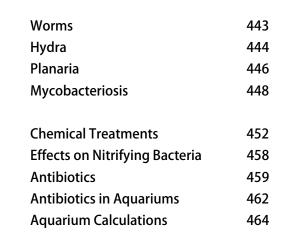
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Source of Information



"Australian fishes still are very little known, but I have reason to believe that their number is very considerable. In fact any person collecting even the most common kinds, particularly the small ones, in any river, lake or stream, is almost certain of rendering good service to science." ~ Count F.L. de Castelnau, 1878.

Melanotaeniidae and Pseudomugilidae are two closely related families of freshwater fishes restricted in distribution to Australia and New Guinea. They are commonly known as rainbowfishes and blue-eyes respectively. Ten genera are currently recognised: *Cairnsichthys, Chilatherina, Glossolepis, Iriatherina, Kiunga, Melanotaenia, Pelangia, Pseudomugil, Rhadinocentrus,* and *Scaturiginichthys. Melanotaenia* is by far the largest genus. The Pseudomugilidae family has been well studied in recent years but controversy still exists as to whether they should be regarded as a separate family or in a subfamily of Melanotaeniidae. No less controversial in recent years has been the position of their relationship to other atheriniformes, most notably to the Telmatherinidae and Bedotiidae families.

Australia and New Guinea are much more than just adjacent land masses as they have been connected throughout most of their history. The Sahul Shelf, beneath the shallow Arafura Sea and Torres Strait that now separate the two countries, was above sea level until as recently as about 6–8,000 years ago during the latest glacial lowering of the sea level, and southern New Guinea streams were confluent with those of the adjacent Australian coast. Indeed, about 50 species of freshwater fishes from southern New Guinea also occur in northern Australia, emphasising the historical link between the regions, and many of these are endemic to the two regions. The Olive and Jardine Rivers of Cape York Peninsula show some of the strongest relationship, with 81% and 63% of the fish species found in these rivers being common between the two countries.

Australia and New Guinea are perhaps better known for their marine fish fauna rather than for their freshwater fauna. Nevertheless, the freshwater fishes of Australia and New Guinea are distinctive and have been the subject of significant discovery over the last 30 years. The amount of recent ichthyological activity in Australia and New Guinea can be gauged conservatively from the number of recently described species or subspecies. Since 1970 about 70 Australian and about 130 New Guinea freshwater fish species or subspecies were described or are awaiting description. New species have been found and described at rates as high as any in the history of Australian and New Guinea ichthyological exploration.

The Australian fauna has been reasonably well studied, in terms of their systematics, but new species continue to be described based mainly on genetic taxonomic research of known species. Australian freshwater fishes were reviewed by Allen (1989), who recorded 187 species and subspecies; subsequent collecting and research have taken this count to around 302 (Allen *et al.* 2002).

In contrast to the Australian species, very little indeed is known about the New Guinea fauna and collecting has mainly focused on the major river systems. Freshwater fishes of New Guinea were reviewed by Allen (1991), who listed 320 species, which including some estuarine forms; subsequent collecting and research have taken this count to about 360.

Increased understanding of freshwater fish systematics and distribution in the region has largely stemmed from the application of collecting and systematic techniques by professional ichthyologists. However, no less significant has been the input from amateur ichthyologists and aquarists. In particularly, during the last three decades a profound increase in interest in keeping freshwater fishes has resulted in the formation of various specialty societies e.g., Australia New Guinea Fishes Association (ANGFA) and the Internationale Gesellschaft für Regenbogenfische (IRG), and in numerous amateur collecting expeditions. Aquarist interest has particularly concentrated on the endemic rainbowfishes, and this, in combination with taxonomic and field studies, mainly by Gerald R. Allen, has resulted in a dramatic increase in the number of species recognised in this family; of the approximately 75 species and subspecies currently recognised, more than 60% have been described since 1978.

Other notable discoveries include *Scaturiginichthys vermeilipinnis*, a new genus and species of pseudomugilid from an artesian spring system in central Queensland (Ivantsoff *et al.*, 1991); and numerous rainbowfish species endemic to various river and lake systems in New Guinea. There have also been significant advances in our understanding of the phylogenetic relationships and biogeography of melanotaeniid fishes in Australian and New Guinea (Zhu *et al.* 1994; McGuigan *et al.* 2000; Unmack 2001).

The first rainbowfish (*Melanotaenia nigrans*) was scientifically described in 1843 from a collection of freshwater fishes acquired in the Northern Territory. They were collected by John Gilbert in 1840, from the King River, near Victoria Settlement in the Northern Territory, Australia. A single specimen ended up in the British Museum in London where John Richardson described it as a new species of hardyhead named *Atherina nigrans*. The differences between *A. nigrans* and the real hardyheads were enough for the American, Thomas Gill, to create the genus *Melanotaenia* for this lone species in 1862, still within the family Atherinidae. The next step was the creation of a subfamily Melanotaeniinae by Gill in 1894 to stress the differences with the hardyheads even more.

Another 70 years were to pass before Ian Munro treated the rainbowfishes as an independent family for the first time in 1964. Although a greater variety of Australian rainbowfishes were by then scientifically identified, many New Guinea rainbowfishes were still undiscovered. Gerald Allen, whilst employed with the Western Australian Museum, undertook a full generic classification of the rainbowfish family in 1980.





Rainbowfishes form the most speciose group of fishes inhabiting freshwaters within the Australia-New Guinea region. Despite this, relatively little is known about the biology and ecology of the majority of rainbowfish species in their natural habitat. A review of the literature currently available does highlight a number of gaps in our knowledge of many species. There are some species where there is a considerable amount of information available while there are other species where there is little or no information available. In addition, there are specific gaps in the information available in otherwise well documented species. As well as a number of species that are in need of additional research, information such as reproduction and natural habitat conditions is limited. These include water quality requirements, spawning frequency and habitat preferences. Spawning information in the wild is particularly lacking for almost all species, as is general information on egg and larvae development, habitat preference and water quality tolerances.

Despite such a variety of species, research into their basic natural biology and ecology is lacking and most information that is available is mainly based on aquarium observations. Obviously, there is urgent need for such studies in order that species can be properly conserved and managed.

Clearly, there is also need for much more survey work to be done in Australia and New Guinea, as some areas remain poorly collected. There is also a need for more careful study of the many widespread species, as it is highly likely that such study will lead to a significant increase in the number of recognised species. For example, variation in morphology within the Melanotaenia genus is high, with species differing from one another though small variations in colour, morphology and meristics. Indeed, one species, Melanotaenia trifasciata, has been divided into many geographic forms, each with highly restricted, allopatric distributions. Populations of almost every river system they occupy have their own distinctive body colour and pattern. At the same time, body form within species is relatively pliant and appears to be dependent upon streamflow and correlated habitat characteristics, which can sometimes make identification in the field difficult. Much could be gained from careful analysis of the many morphological characters already at hand, such as the colouration characters noted for many of the rainbowfish "varieties". Colouration characters, however, when not supported by other characters, have generally been dismissed by ichthyologists working on rainbowfishes from Australia.

The recognition of taxonomic diversity is a key issue underlying the problems associated with assigning species status to this group of fishes. Can we be sure that a species is truly defined, or is it a species complex, or multiple species with distinct characteristics - sufficiently isolated to be recognised as a species. Where a single species might be seen as common, in reality there might be numerous species.





Distinct geographic clades within species are regarded alternatively as 'Evolutionarily Significant Units' (Moritz 1994) and are not named. An evolutionarily significant unit is a population of organisms that is considered distinct for purposes of conservation. This term can apply to any species, subspecies or geographic population. Subspecies are morphological variants distinguished at the level of the population — 75% or more of the individuals of the populations of one subspecies can be distinguished from those of other subspecies.

Existing data suggest that New Guinea is worthy of the highest conservation priority due to its extraordinary species diversity, significant endemism, and high degree of threat. It is not surprising that there are still many new rainbowfish species in New Guinea that await discovery. More than half the known species of rainbowfishes are endemic to New Guinea. There are several areas that have particular potential as reservoirs of undiscovered species. New Guinea is today less known than any other habitable area of equal size on the globe.

These are also times of serious concern for the present and future health of rainbowfish populations, and other aquatic organisms. New Guinea, particularly the Indonesian province of West Papua, is one of the most threatened biological hotspots, with its plants and animals facing possible extinction due to slash-and-burn subsistence farming, transmigration, rampant logging, illegal poaching, unregulated mining and other practices. Many human activities are increasingly disturbing and, in some cases, destroying freshwater habitats. Wherever human populations are expanding, so too are the harmful waste products of mining, industry, agriculture, and population growth. These impacts have negative and sometimes devastating effects on aquatic life and habitats. Freshwater fish species and aquatic communities have also been placed in harm's way by the introduction of non-native species.

Specific rainbowfishes that are considered threatened are: Chilatherina axelrodi, C. bleheri, C. bulolo, C. sentaniensis, Glossolepis incisus, G. maculosus, G. pseudoincisus, G, ramuensis, G. wanamensis, Kiunga ballochi, K. bleheri, Melanotaenia ajamaruensis, M. angfa, M. arfakensis, M. boesemani, M. catherinae, M. corona, M. eachamensis, M. exquisita, M. gracilis, M. herbertaxelrodi, M. iris, M. lacustris, M. maylandi, M. misoolensis, M. monticola, M. ogilbyi, M. oktediensis, M. papuae, M. parva, M. pimaensis, M. praecox, M. pygmaea, M. sexlineata, M. vanheurni, Pseudomugil connieae, P. furcatus, P. majusculus, P. mellis, P. paskai, and Scaturiginichthys vermeilipinnis (Conservation International 2002; IUCN 2009).



#### History of Rainbowfishes in Captivity

Australian rainbowfishes have been maintained in home aquaria at least since the beginning of the last century. As early as 1915, Albert Gale in his book *Aquarian Nature Studies & Economic Fish Farming* made known the hobby of keeping Australian freshwater fishes. This book covered many subjects on the captive maintenance and care of a number of species. A section of the book also explored the possibilities of commercially breeding Australian freshwater fishes for the aquarium hobby. Albert Gale was a member of the Royal Zoological Society of New South Wales and regularly wrote articles about Australian freshwater fishes for *Aquatic Life*. This magazine was edited by W. A. Poyser and published by Joseph E. Bausman in the USA during the early part of the last century.

During the late 1920s and early 1930s large Aquarium Societies were established in major cities all around the world. Fish shipments at the time were in old-fashioned flat metal "German" cans, with a small neck and very wide body to give maximum air surface. As sea voyages were long and no oxygen was used, the fish generally arrived in poor condition. However, some survived the journey and were bred by experienced hobbyists.

During this early period rainbowfishes were known as sunfish. When exactly this group was called rainbowfish nobody really knows. In January 1934, National Geographic Magazine published an article written by Walter H. Chute, then director of the Shedd Aquarium in Chicago USA, called "Tropical Fish Immigrants Reveal New Nature Wonders" in which appeared a reference to the Australian rainbowfish. However, the earliest record that has so far been found is in the German aquarium magazine "Wochenschrift für Aquarien und Terrarienkunde" in September 1931 by Erich Henzelmann, who wrote an article about the 'Regenbogenfisch' Melanotaenia nigrans (which was actually Melanotaenia duboulavi). The earliest reference to the name rainbowfish that I have been able to find in Australia is in an excursion report of the Aquarium and Terrarium Society of Queensland. It was written by the then secretary, Amandus Rudel, and referred to a collecting trip on March 6, 1932. After this date, all collecting reports generally referred to the name rainbowfish. The common name of 'Sunfish' was designated to Rhadinocentrus ornatus, e.g., Moreton Island Sunfish.

Amandus Rudel was a founding member of the Aquarium & Terrarium Society of Queensland, and in 1927 he introduced the Australian rainbowfish to the international aquarium hobby when he sent specimens of *Melanotaenia duboulayi* by steamship to Germany, and which were later bred by the Berlin Aquarium. Speaking of *Melanotaenia duboulayi*, Amandus said, "I was astonished at the beauty of this fish the first time I saw it. Like a living rainbow, there is no other fish which can compare with its beauty. Naturally it has been my favourite ever since." It is believed that from this initial shipment *Melanotaenia duboulayi* were introduced to the organised aquarium hobby throughout Europe, and then to North America.



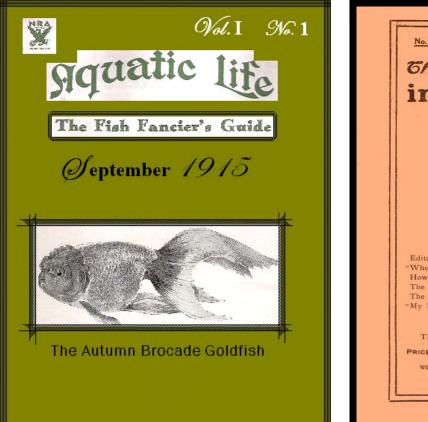
Fish shipments at the time were in old-fashioned flat "German" cans, with a small neck and very wide body to give maximum air surface.

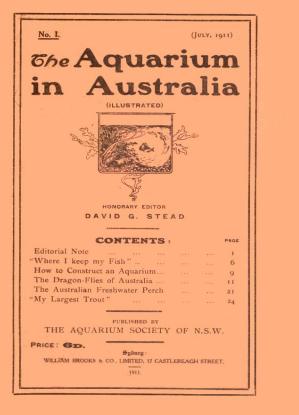
They are probably the species upon which today's common name "*Rainbowfish*" is based. In 1930, three specimens of *Melanotaenia duboulayi* were collected in the Mississippi River. This was one of the earliest accounts of an introduced ornamental fish found in the USA (O'Donnell 1935).

Amandus Rudel was also responsible for introducing another rainbowfish to the international aquarium hobby. In 1934, he sent 12 specimens of *Melanotaenia maccullochi*, collected by him near Cairns, in northern Australia, to Fritz Mayer in Hamburg, Germany. Four arrived alive and developed into two pairs. They were one of the most popular aquarium fish from Australia. In the German aquarium magazine "*Wochenschrift für Aquarien und Terrarienkunde*" in May 1935, Fritz Mayer gave the first account of their breeding, which was translated by F. H. Stoye in Innes' "*The Aquarium*" in December 1936.

The hobby went into recess during the Great Depression and following war years, and very few aquarium specimens survived that period. The aquarium hobby didn't really recover until the late 1950s when shipment with plastic bags and oxygen, in insulated containers was developed. This, together with faster air travel, enabled the aquarium hobby to flourish and was considered one of the most popular hobbies of the period.







Other shipping containers in the 1920-30s were merely a straight-sided metal pail with a tight fitting lid that were packed in a wooden shipping box. The wooden shipping box was lined with cane fibre-board for insulation and the metal container was packed inside it in sawdust.





During this growth period however, due to limited availability, rainbowfishes were never readily available to the general aquarium hobby. The only rainbowfishes that were available in the retail trade were a colourless assortment bred in Southeast Asia fish farms that looked nothing like the wild species, and were never very popular with fishkeepers.

Rainbowfishes from New Guinea started to arrive in Australia around the mid 1950's. They were being maintained by only a handful of enthusiasts and were virtually unknown to the international hobby. There are some reports that specimens of *Melanotaenia affinis, M. goldiei, M. rubrostriata* and *M. sexlineata* (although probably *M. papuae*) were being maintained in the Australian hobby as early as 1959. During the 1960's and 70's a small trickle continued to arrive in Australia from New Guinea. However, the publication in 1982 of *Rainbowfishes of Australia and Papua New Guinea* by Gerald Allen and Norbert Cross, greatly increased the popularity of keeping rainbowfishes, and the desire for the newly discovered New Guinea species, turned that trickle into a flood. ANGFA, the Australia New Guinea Fishes Association was also formed in 1982; further promoting the keeping of rainbowfishes.

The importation of New Guinea rainbowfishes into Australia during this period did not have any significant restrictions and a number of different species were brought into the country by private collectors, which were subsequently distributed in the hobby. However, the increasing importation of New Guinea rainbowfishes eventually attracted the attention of the Advisory Committee on Live Fish (ACOLF), the then Federal Government body responsible for controlling the importation of live fishes. In late 1983, ACOLF for some obscure reason decided to ban the importation into Australia of all species of freshwater fishes from New Guinea. Despite the ban, however, new rainbowfish species from New Guinea continue to be imported, bred and distributed widely in the general aquarium hobby around Australia.

During the mid 1980s, Heiko Bleher, an intrepid aquarium fish collector started collecting the newly discovered New Guinea species, breeding and distributing them into the International aquarium hobby. This trend has continued over the past two decades, developing side by side with the increase in the discovery of numerous new species. More than 68 species have been discovered and the possibility of this number increasing in the next decade is almost certain.

Most rainbowfish species that have been introduced to the aquarium hobby has been by various private collectors. Rainbowfish enthusiasts travel to far-off places in Australia and New Guinea to collect new and different coloured forms. Many of the rainbowfishes that are available in the hobby today have resulted from the activities of these enthusiasts. These same hobbyists generate a substantial amount of data on habitat conditions, collecting locations, colour varieties and reproductive biology.

Keeping rainbowfishes in an aquarium can be a fascinating activity for the whole family with a number of advantages over the keeping of more conventional pets. Relatively little space is required; an aquarium can be aesthetically pleasing, and the fish themselves are clean, quiet and non-demanding, given a few simple requirements. Rainbowfishes have very similar breeding habits, their food requirements are similar, and water that suits one particular species will suit all. All are of good-natured temperament and will live harmoniously, more or less, with one another. They also possess all the attributes we look for in aquarium fishes; ease of breeding, hardy, beautiful colours, peaceful disposition, and they won't destroy your plants or move the gravel around in your aquarium. They also come in a variety of sizes to suit almost any aquarium.

Most rainbowfishes available in the retail hobby today are bred in captivity. This is due in the main to the difficulties involved in collecting and transporting live rainbowfishes from their natural habitats in remote areas of Australia and New Guinea. This is even more relevant in New Guinea where dense rainforests, virtually no infrastructure like roads and airports has meant that organised collecting for the aquarium trade is just simply not possible.

Rainbowfishes spawn readily in captivity and there is now a large captive breeding pool that generally satisfies most of the commercial demand, thus negating the need for wild-caught fish. However, there is probably some limited collecting of wild fish in New Guinea, although reliable data is lacking.

One of the major problems affecting the popularity of rainbowfishes is that they generally don't display their best colouration when kept under normal retail store conditions. Most retail stores maintain their fishes in bare, over-crowded aquariums. This is done mainly for economic reasons and to facilitate their ease of capture and sale. However, rainbowfishes maintained under these conditions will feel stressed and lose the beautiful colours that they are renowned for. In addition to this, too many species are offered for sale at a small size before they have developed their full adult colouration. Unless the general fishkeeper knows what they will look like when taken home they will more than likely be passed over for some other species.

Blue-eyes (Pseudomugilidae) are small colourful fishes rarely exceeding 5-cm in length. Their natural habitats are similar to that of rainbowfishes, although they are usually found in shallower water. Some species are found in brackish and marine waters. Blue-eyes are so called because of the striking blue colour of the iris. They are close relatives of the rainbowfishes and were previously classified with them but were given family rank by Saeed *et al.* (1989). Although, Dyer and Chernoff (1996) consider pseudomugilids as a subfamily of the Melanotaeniidae.

In general, male blue-eyes are brightly coloured, and their fins have elongate filaments that are utilised in elaborate courtship displays. They are valued as aquarium fishes due to their beauty, small size, and peaceful disposition and are easy to maintain and breed in captivity. They are hardy aquarium fishes despite their small size, and adapt well to captivity showing their colouration at all times. Ideally, they should share their aquarium with similar sized tankmates and be kept in small groups. Unfortunately, they are not as prolific as rainbowfishes and it is for this reason that most commercial breeders have ignored them and therefore are not generally available in the retail aquarium trade.



## **Rainbowfishes** Distribution & Habitat



Photo: Alan Travers

Rainbowfishes are one of the most speciose groups of freshwater fishes inhabiting the Australia-New Guinea region. Australia lies between latitudes 10°41'S (Cape York) and 43°39'S (South East Cape, Tasmania) and between longitudes 113°09'E (Steep Point) and 153°39'E (Cape Byron). The latitudinal distance between Cape York and South East Cape, Tasmania is 3,680 km. The longitudinal distance between Steep Point and Cape Byron is about 4,000 km. With a total land area of 7,682,000 km<sup>2</sup>, it is the lowest, the flattest and, with the exception of Antarctica, the driest of the continents. The continent has a wide range of climatic zones, from the tropical regions of the north, through the arid expanses of the interior, to the temperate regions of the south. Seasonal fluctuations can be great, with the temperatures ranging from above 50°C to well below zero. The continent often experiences natural disasters, particularly droughts, floods, tropical cyclones, severe storms and bushfires.

Australia is an isolated continent, with the Indian Ocean to the west, South Pacific Ocean to the east, and Southern Ocean to the south, but with New Guinea and Southeast Asia just to the north. The birth of Australia began soon after the dinosaurs disappeared, 65 million years ago. It was the last landmass to split away from the ancient southern super-continent Gondwana. The world's continents were once all joined in a single landmass called Pangaea. In the Jurassic period (about 160 million years ago) a northern continent, Laurasia, and a southern continent, Gondwana, split apart. The exact nature of the break-up of Gondwana is not understood with precision, but it gradually fragmented over geological time, with India and then New Zealand moving away from the Australia-Antarctica-South America group during the Cretaceous period (140 million years ago). The latter group of continents separated from each other during the Tertiary period (from about 70 million years ago).

It took many millions of years for Australia and Antarctica to fully separate, with Tasmania caught in the middle. But finally, about 40 million years ago, they parted and commenced a northward drift. Australia dragged Tasmania north, leaving Antarctica alone at the bottom of the world. With Australia out of the way, ocean currents were free to circle the South Pole, as they still do today, greatly influencing the world's climate.

During this time, Australia experienced numerous changes in climate, but the overall trend was towards greater aridity. The great inland seas and lakes dried out. Much of the longestablished broad-leaf deciduous forest began to give way to the distinctive hard-leaved sclerophyllous plants that characterise the modern Australian landscape. For many species, the primary refuge was the relatively cool and wellwatered Great Dividing Range. Even today, pockets of remnant vegetation remain in the cool uplands, some species not much changed from the Gondwanan forms of 60 or 90 million years ago. New Guinea began to form then, along the northern edge of the Australian continental plate, developing in two parts. One part was the northern rim of the Australian plate itself and the other a string of islands off the north-east coast, away from Laurasia. The islands and mainland only came together towards the end of the Tertiary, forming the high and rugged mountains and giving New Guinea its present form. That process is on-going, with some mountains having now reached 4,884 metres ASL in little more than 3 million years since the beginning of the accelerated uplift. This had a number of important consequences, including the formation of the New Guinea highlands, and providing an opportunity for the dispersal of Asian taxa to the now relatively close Australia, and similarly for Australia taxa to emigrate.

New Guinea itself should be considered part of greater Australia, because, for a great deal of its history, it has been part of the Australian mainland (as has Tasmania). New Guinea's proximity to the Sunda Shelf and many islands may well have provided a stepping-stone for taxa to move between the two worlds. The Sahul Shelf, constituting Australia, Tasmania, New Guinea and adjacent islands, possibly including Halmahera Island, has continued on a northward path ever since.

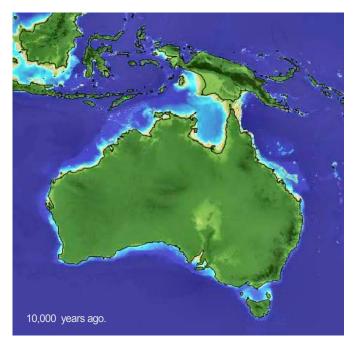
The Sahul Shelf is a structural platform of the ocean floor and is bounded to the northeast by a series of deep-sea troughs and to the northwest by troughs, a chain of coral reefs, and a series of submarine ridges. The Sahul Shelf was once above sea level, and its surface still bears erosional features formed when streams crossed it to the oceans. The shelf was slowly warped downward by crustal forces. This subsidence is evidenced in coral atolls along its edge, composed of coral that grew as the land sank. The shelf's main divisions are the shallow Arafura Shelf, covered by the Arafura Sea and Gulf of Carpentaria; the Sahul Shelf under the Timor Sea; and the Rowley Shelf underlying a part of the northwest Indian Ocean extending to North West Cape, Western Australia. To the north lay the deeper Timor tough and the volcanic Lesser Sunda Islands, separating the Sahul from the Sunda Shelf.

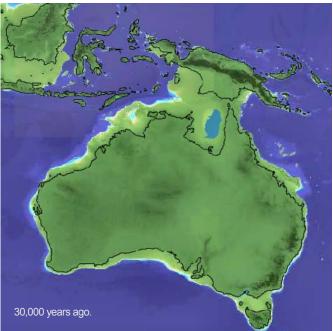
Australia and New Guinea have been alternately land-linked and separated by water on a number of occasions over millions of years. The alternating global warming and cooling episodes of the last three million years repeatedly isolated then reconnected New Guinea and Australia, as the Arafura Plain successively submerged and emerged with changing sea levels. Global sea levels are currently higher than at anytime during the last 120,000 years, separating Australia and New Guinea by sea. However, Torres Strait has been acting almost consistently as a land-bridge since the last interglacial about 118,000 years ago up until 6~8,000 years ago, when marine transgression closed the bridge. About 12,000 years ago, sea levels were low enough that the Arafura Shelf was exposed, and 20,000 years ago, sea levels were 120 metres below present levels. The water barrier, which is now the Arafura Sea, Gulf of Carpentaria, and Torres Strait, which separates Australia and New Guinea are extremely shallow, with average depths ranging from about 15 to 60 metres.



Between 12,000 and 55,000 years ago, the Gulf of Carpentaria was a large (~30,000 km<sup>2</sup>) inland lake. Not only did Cape York Peninsula provide a land link between New Guinea and north-east Australia, but also Lake Carpentaria would have provided a freshwater aquatic link. The lake would have been fresh or brackish for much of its existence. Evidence from deep core drilling reveals a pattern of establishment and marine inundation of Lake Carpentaria that appears to have been repeated. It was a freshwater lake in the Jurassic then inundated by a marine transgression (in limestone deposits), and there was a further freshwater episode in the Miocene, followed by another marine transgression. As the sea levels rose, this lake disappeared.

One reminder of this ancient lake is the current fragmented distribution of rainbowfishes such as the *Iriatherina werneri* and *Melanotaenia maccullochi* in rivers of the Northern Territory, northern Queensland and southern New Guinea. The fish species of Cape York Peninsula also have a strong affinity with New Guinea. The Olive and Jardine Rivers show some of the strongest relationship, with 81% and 63% of the fish species found in these rivers being common between the two countries.

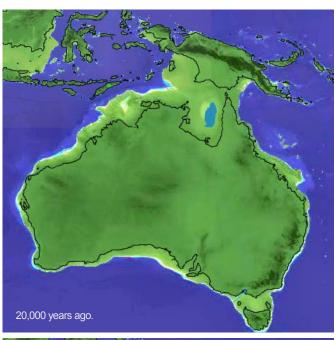




There is convincing geological evidence for the historical existence of Lake Carpentaria. Moreover, it has been suggested that the outflow of Papua New Guinea's Fly River was diverted westward into Lake Carpentaria during this period, although this hypothesis is still controversial. Harris *et al.* (1996) found no evidence for a past westward diversion of the Fly River, and suggested that the outflow of the river in 'recent' geological time has always remained on an easterly course into the Coral Sea. However, the hypothesis that Lake Carpentaria provided habitat for, and facilitated gene flow among freshwater *Macrobrachium* populations during the late Pleistocene is supported by recent analyses (De Bruyn *et. al.* 2004).

These maps show the changing shape of Australia and New Guinea that mimics the rise and fall of sea levels over the past 10,000~50,000 years. The green sections of the map indicate dry land. It was during such periods that rainbowfishes were dispersed between Australia and New Guinea.

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Cape York Peninsula provided the main land link, but a second land link between Arnhem Land and New Guinea formed at much lower sea levels. This made possible the movement of terrestrial plants and animals so that a potential biological 'bridge' existed between the continent and sub-continent with a wide plain across what is now the Arafura Sea. The only high ground on the plain were low hills that are now islands fringing the Kimberley coast and Arnhem Land, the islands in Torres Strait and the low hills that fronted the north-western coastline of the Arafura plain (now the Aru Islands). Major river systems flowed across this plain, arising from both the south and the north. The plain had vast shallow lakes, and embayments fringed with mangroves and salt-marsh. Rainforests were largely confined to the mountains and slopes to the north and to riparian zones and protected gorges in the south, with much of the Arafura plain a savannah, similar to parts of northern Australia today.

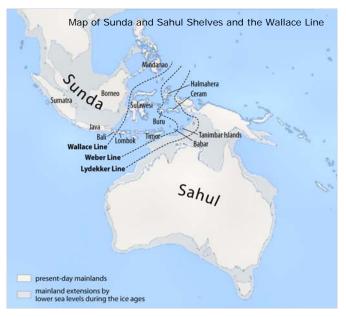
The connections were especially strong, close and more enduring between Cape York Peninsula and New Guinea. There are several plant and animal species, which only occur on Cape York Peninsula and in New Guinea. Plants, birds, reptiles, and mammals with this distribution are largely found in the northern half of the Peninsula and reach their greatest diversity in the mid-Peninsula rainforests. The rising sea also fragmented the range of many other plants and animals. Comparable environments and species assemblages persist in the Fly River region, Port Moresby and Popondetta areas of southern New Guinea, and across northern Australia.

The current distribution of a number of northern Australian and southern New Guinea rainbowfish species can be explained by the opportunities the lake and the exposed Arafura shelf provided. The Arafura Shelf that defined the western boundary of Lake Carpentaria would also have provided a land-bridge to New Guinea presumably with drainages flowing west to the Timor Sea. This would have allowed potential interchange of forms between West Papua, Arnhem Land and the Kimberley via coastal rivers and associated habitat quite different from that provided by Lake Carpentaria. It would also have isolated the rainbowfish fauna from these western and west-central rivers from those flowing into the eastern seaboard of Australia and south-eastern New Guinea. This may explain the different species found in the Kimberley and western Arnhem Land.

Unfortunately, no rainbowfish fossils exist so their evolutionary history will probably remain obscure. However, there is some belief that rainbowfishes probably originated in the north of Australia, or in southern New Guinea and then spread eastward, north into New Guinea and southward down the northeast coast of Australia, differentiating into the various species we know today. In south-eastern Australia, the primary driving force behind current rainbowfish distributions appears to be climatic.

Many factors affect the distribution of rainbowfishes but one of the most important is biogeographical boundaries. As far as rainbowfishes are concerned, the most important biogeographical features are the drainage division boundaries. It is important to note that biogeographical boundaries do not necessarily correspond with governmental boundaries. The western half of New Guinea is the Indonesian province of West Papua. However, Indonesia is part of the Asian continental plate and was, until 20 million years ago, well separated from Australia and New Guinea. The Indonesian archipelago spans two major biogeographical regions divided by Wallace's line. West of this line lies the Indo-Malayan region, which includes the islands of Java, Borneo and Sumatra on the Sunda Shelf; to the east lies the Australasian region. Wallacea is a biogeographical designation for a group of Indonesian islands separated by deep water straits from the Asian and Australian continental shelves. Wallacea, comprising the Lesser Sunda Islands, the Moluccas and Sulawesi, has had no recent land connection to either continent.

The islands of the Sahul Shelf which include Waigeo, Batanta, Salawati and Misool to the west; Aru Islands to the south; and Japen to the north in Cenderawasih Bay all had recent intermittent land connections with mainland New Guinea. Those which lie off the Sahul Shelf had no connections with New Guinea in the recent past. The island of Bougainville is part of Papua New Guinea, but is biogeographically most similar to the Solomon Islands. Similar biogeographical and governmental boundaries exist across the Torres Strait between the southern part of Papua New Guinea and the northern tip of Queensland, Australia.



Although the region today includes two very different nations and part of a third, and although the two main landmasses are currently separated by Torres Strait, from a biological and geological point of view, it is a single unit. Most of the fauna and flora of New Guinea are shared, at least in their origin, with the continent of Australia. While much of the rest of the world underwent significant cooling and thus loss of species diversity, Australia-New Guinea was drifting north at a pace such that the overall global cooling effect was roughly equalled by its gradual movement toward the equator. Temperatures in Australia-New Guinea remained reasonably constant for a very long time, and a vast number of different plant and animal species were able to evolve to fit particular ecological niches. Because the continent was more isolated than any other, very few outside species arrived to colonise, and unique native forms developed unimpeded.



### Natural Habitat (Australia)

"Habitat can be defined as the specific type of place where a plant or animal lives. For plants and animals which live in water, habitat is available only when it is submerged."

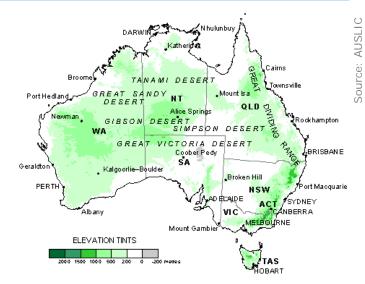
In Australia, rainbowfishes generally occupy three broad climatic zones. Above the Tropic of Capricorn has a tropical climate characterised by a generally hot, humid summer with strongly seasonal rainfall, and a mild to warm, dry winter. Below the Tropic of Capricorn is sub-tropical with a similarly hot, humid summer and seasonal rainfall, but with some significant rainfall occurring during the mild winter. The interior portion of Australia occupied by rainbowfishes experiences an arid sub-tropical climate. Summers can be extremely hot and dry, with variable rainfall; winters are cool to warm and dry, with irregular light rain.

The majority of rainbowfishes found in Australia are distributed throughout the northern and eastern coastal strips. The presence of substantial rainfall and the range of habitats found in these regions accounts for the relatively greater number of species found in this part of the continent. In southern Australia, their distribution is almost certainly controlled by winter minimum water temperatures. The family's distribution is exceptionally broad, ranging from the tributaries of the Murray-Darling system in Victoria, northwards up the east coast to Cape York, and west to the north-west coast of Western Australia. They also occur throughout the inland rivers of the arid Lake Eyre drainage in central Australia.

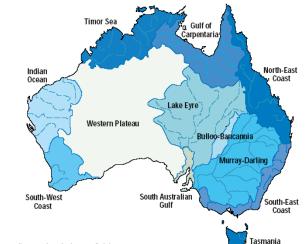
There are three major landforms on the mainland of Australia; the Great Dividing Range and its associated smaller ranges, the Central Eastern Lowlands (west of the Great Dividing Range) and the Great Western Plateau. These landforms influence the major drainage patterns of the mainland. The Australian Water Resources Council has defined twelve major drainage divisions - eleven on the mainland and Tasmania's drainage system is the twelfth, which are sub-divided into water regions which are in-turn sub-divided into river basins. Within major drainage basins, there are always minor drainage basins. Minor basins sometimes have two or three rivers in a system. Other minor basins have only one river, rising in a mountain range and draining away into lonely desert land. The waters that soak away into the sands from these rivers are not always lost. They may drain into an artesian or sub-artesian basin. The Great Artesian Basin is the world's largest and deepest artesian basin. It covers an area of 1,082,400 km<sup>2</sup>.

The Australian 1:250,000 scale map series shows about three million kilometres of rivers and streams. However only around 1400 are named rivers (approx. 166,018 km) many of which are intermittent or ephemeral. Some only flow after heavy rains, which may be years apart. Of these rivers and streams, only about four percent are dam-free. Australia stores more water per head of population than any other country, so as to provide security of supply for agricultural and urban use.

Rainbowfishes—Their Care & Keeping in Captivity



There are three major landforms on the mainland of Australia; the Great Dividing Range and its associated smaller ranges, the Central Eastern Lowlands (west of the Great Dividing Range) and the Great Western Plateau.



Australia's major drainage divisions





11



Inland waters include all water inland of estuaries, both in surface features like streams, lakes, wetlands and reservoirs, and in the subsurface as groundwater. The chemistry of our surface inland waters differs from most waters elsewhere, often being dominated by sodium chloride rather than calcium and magnesium bicarbonates. Groundwater is often very old; for example, in the Great Artesian Basin water travels across Queensland, to emerge in central Australia in bores one to two million years after it entered the ground. The generally arid climate means that mainland Australia has relatively few permanent and freshwater lakes. Lakes on the mainland are often shallow, dry and salty. Only on the Central Plateau of Tasmania do a number of larger permanent fresh-water lakes occur.

Australia, by virtue of its size, contains a large variety of different freshwater ecosystems. Broadly, the north of the continent has a monsoonal rainfall pattern, while the south generally has a temperate, winter-rainfall pattern. The eastern seaboard and the extreme south west of the continent are reasonably well-watered, while the arid interior is characterised by rainfall which is extremely variable.

Most rivers, even the larger ones, are ephemeral in most years having highly seasonal and variable flow. Many cease to flow during the dry season and tidal influences can extend some 80–100 km upstream. Some are little more than a chain of elongated waterholes for much of the year.

Extended periods of low flows during the dry season not only separate main-channel habitats from off-channel floodplain lagoons, but can also reduce contiguous mainchannel habitats to a string of shallow, isolated pools. In the drier areas, these habitat changes can dramatically influence fish community composition by increasing density dependent interactions and causing extreme water quality conditions.

Aquifer-fed streams such as the Daly River in the Northern Territory, the Gregory River in the Gulf, and the Jardine River on Cape York Peninsula, continue to have significant flows even at the end of the dry season. Such aquifer fed perennial rivers are especially important for many terrestrial and aquatic species. As a consequence of the water releases from Lake Argyle and Lake Kununurra the downstream portion of the Ord River in Western Australia is also now perennial.

Rivers never flow in a straight line. Even when there are no obstacles, flowing water has an inherent tendency to meander and, over time, the meanders themselves also move. Within the boundaries of their floodplains, river channels shift from side to side, shuffling old sediment and pushing it downstream toward the sea. A river pushes sediment along a floodplain a bit like you might use a running hose to squirt dirt from a concrete path - sweeping from one side to the other, progressively moving the dirt along. Similarly, rivers sweep from side to side within their floodplains. Each meander moves the sediment along a little then leaves it behind for the next.





▲ Daly River Billabong (Northern Territory) ▼ Gregory River (Queensland)





As a floodplain river moves, it leaves tracks in its wake: repeated, curved rills called 'scrolls', which fill with water to become billabongs. Changing flows also frequently cut off whole loops of river channel, called meanders, to create flood-filled 'oxbow' billabongs.

Fast-flowing rivers with steep gradients have small meanders, which can sweep across their narrow floodplains in just a few decades. But slow-moving lowland rivers - such as the Murray, the Ovens and the Murrumbidgee - have far larger, looping meanders that sweep much more slowly across wide floodplains. River meanders are irregular, but not random. Their looping paths have quite measurable wavelengths, which depend on the downhill gradient and the volume of water the river carries. Lower volume rivers have smaller loops, which in the underfit rivers of the Murray Basin are often superimposed on the wider loops of older, greater rivers.

By world standards, Australia has only one large river system, the Murray-Darling, whose catchment drains the western slopes of the Great Dividing Range and the arid interior. The Murray-Darling Basin covers an area in excess of a million square kilometres (14% of the entire continent) and occupies large areas of southern Queensland, inland New South Wales (NSW), and northern Victoria, as well as South Australia's south-east. The Murray-Darling is also one of Australian's most degraded river basins, an issue of special concern to South Australia - the State at the "bottom end" of the catchment. The lower Murray now experiences drought level flows three years out of every four, compared to one in twenty years under natural circumstances. The loss of biodiversity in the region and degradation of its rivers is well documented. In particular, the native fish species of the Murray-Darling Basin have suffered serious declines in both distribution and abundance resulting in the threatened status of one-quarter of the thirty-five species present.

The Murray River and its tributary, the Darling River, are the main rivers in the Murray-Darling River Basin. The Darling River flows south from the junction of the Culgoa and Barwon rivers. Although the Culgoa is longer than the Barwon, the source of the Darling is generally agreed to be the Barwon River as it has the greater volume of water.

The headwaters of the Darling can be traced to the MacIntyre River, which starts in the Great Dividing Range, and forms part of the border between NSW and Queensland. It eventually flows south into the Barwon. The Barwon-MacIntyre section is sometimes referred to as the Upper Darling. When measured from its source in Queensland to its mouth on the coast south-east of Adelaide, the Murray-Darling river system is 3,370 kilometres long, about half the length of the world's longest river, the Nile.





▲ Jardine River (Queensland) ▼ Gilbert River [Dry Season] (Queensland)



#### **Tropical Rivers**

Tropical freshwater ecosystems in northern Australia are considered to be the most biologically diverse and healthy aquatic ecosystems in Australia today. These systems are dominated by monsoonal rainfall patterns and consequently have the most seasonally-restricted discharges in the country. More than half the annual flow occurs within just a three-month period, followed by a relatively long period of little or no flow. The marked seasonality of rainfall and subsequent discharge drives massive changes in the extent of river and wetland habitats; often leaving floodplains inundated for several months each year.

The majority of Australia's tropical river systems are characterised by large catchments, with expansive, seasonally-inundated floodplains. Covering an area of more than 1.3 million km<sup>2</sup>, the tropical rivers region includes more than 60 major rivers and hundreds of smaller streams flowing directly into the sea. These extend across all catchments from the Fitzroy River near Broome in Western Australia to the Fitzroy River near Rockhampton in Queensland. It includes some of Australia's largest river systems, which are (by area size) the Flinders, Roper, Victoria and Fitzroy Rivers and (by volume) the Nicholson and Mitchell Rivers. Combined, these rivers and their tributaries extend over one million kilometres and the discharge from these rivers represents ~70% of the freshwater run-off in Australia, which is highly seasonal in almost all catchments.

In tropical rivers, one of the most important temporal phenomena affecting fish is seasonality of the flow regime. The duration and magnitude of elevated flows determine the availability of various habitat types, by regulating lateral and longitudinal connectivity, influencing local hydrology and geomorphology, removing instream vegetation, and affecting water quality. After the first flush of water in the wet season, surface waters in the region generally have very low levels of dissolved solids reflecting the highly leached land surface of the region (Conductivity range  $5-20 \mu$ S/cm). The waters are slightly acidic (pH 5.2) with a very low buffering capacity and generally very clear with low levels of suspended solids (5-60 mg/L). The soft, acidic water probably contributes to a low diversity of molluscs in the region. With each flood event, there is a further general decline in the concentration of solutes. Most of the surface water at this time is derived from surface runoff (or direct precipitation on parts of the floodplain) rather than ground water. Consequently, the proportions of major ions of surface waters closely resemble that of local rainwater. Plants and soil remove over 90% of P, NH<sub>4</sub> and NO<sub>3</sub> from rainwater.

During the dry season the water chemistry changes and the pattern of change varies with different kinds of waterbodies. The spring-fed permanent headwaters and the deep channel billabongs change very little over the year. On the other hand, the standing waters of the shallower floodplain billabongs and backflow billabongs of the lowlands evaporate to some extent and concentrate their dissolved salts steadily during the season. In some billabongs the addition of ground water from seepage may cause the solutes to increase ten-fold or more. As the waters concentrate there is a steady progression towards the composition of seawater. In some billabongs there is a sudden marked rise in conductivity at the end of the dry season; pH also rises slightly over the dry.

When flow begins early in the wet season the composition of the first flush water depends on the manner in which it arises. When the downstream progression is at a steady pace the advancing water may develop a front with high solute concentrations leached from the soils over which it passes and the *p*H may also be quite low (3.5-4.5). Consequently, when this mixes with the water in the billabongs, the water quality for the biota may be very unfavourable for a time until it is diluted by following, more dilute, waters. In some floodplain areas with jarosite soils, oxidation of sulphide to sulphate occurs after the soil becomes wet again after drying out during the dry season and allowing aeration of the soil. This causes very acidic conditions in the soil water and this allows aluminium to dissolve.

High levels of aluminium and sulphate can then be leached from the soil by the slowly advancing water and transported to billabongs. When this happens the water is potentially toxic to fish and mass fish kills may occur. These kills are invariably associated with very low oxygen levels in the water which is probably also caused by the influx of organic matter with the new water. Fish kills can also occur at this time solely from oxygen depletion resulting from influx of organic matter with storm events. When, as often happens, the first flush occurs as a large flood with rapid progression across the floodplain, there is less potential for these harmful conditions to arise.

In seasonal water bodies growth and production of submerged and emergent aquatic macrophytes begins in the early wet season each year when dry ground becomes saturated by rain or floodwater. Maximum biomass of the dominant grasses occurs in the late wet–early dry season. With the senescence of these plants there is a large increase in decomposing detritus. In some billabongs this decomposition results in the water becoming anoxic for a period and this can also be a cause of fish kills.



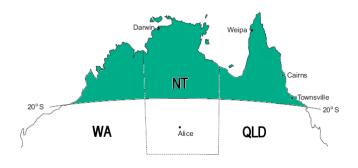


Surface water temperature averages around 30°C but may range from 25°C to 38°C depending on location and time of year. Highest temperatures are recorded late in the dry season. Thermal depth gradients are typically absent during the wet season but frequently develop during the dry. There is some diurnal variation in this gradient as surface waters cool at night. However, even small temperature differences of  $1-2^{\circ}$ C may be sufficient for stratification to occur and this can cause deoxygenation of deeper waters. This happens in many billabongs.

Dissolved oxygen levels are generally at their lowest levels at dawn after a night of steady oxygen consumption by respiration by the aquatic community and before any photosynthesis have occurred to produce more oxygen in the water. Oxygen levels typically then begins to rise soon after sunrise and reach maximum levels around mid afternoon.

There is not much data on the frequency with which total oxygen depletion occurs by this process, but it has been observed on a number of occasions. Whenever this occurs many fish species can be seen gulping at the water surface flushing their gills with the oxygenated surface film of water. The effect of these short periods of anoxia on fish has not been examined in detail. Fish can recover from short periods of this stress but more frequent and prolonged periods may have more harmful effects. Fish have been observed to jump out of the water and strand themselves on fringing vegetation in response to this oxygen depletion. Rivers in the wet tropics region of Queensland are the exception to this general trend. Here, 'dry' season base flows are maintained at relatively high levels by orographic rainfall on high peaks in catchment headwaters. There are a greater number of river systems in Queensland than in any other Australian State and run-off from these rivers accounts for more than 45 percent of the total discharge from all Australian rivers.

Freshwater fish diversity is high in the region: at least 103 native species, representing 37 families, are known to occur in the area. These species account for approximately 45% of the continent's freshwater fish species, 70% of the genera and 70% of the families. Species assemblages are quite consistent across catchments, with each river featuring upstream reductions in species richness caused by the presence of natural barriers to upstream fish movements.



Australian tropical rivers region





▲ O'Shannassy River (Queensland) ▼ Flinders River (Queensland)



Quality of habitat depends on how well the available habitat meets organisms' requirements for survival and reproduction. Different organisms have different requirements and tolerances for such habitat characteristics as flow, substrate, temperature, water chemistry, food availability, shelter etc. Overall there is a scarcity of aquatic habitats in Australia, particularly in the arid and semi-arid areas.

In contrast with most other countries, the area of saline wetlands in Australia far exceeds that occupied by fresh water. Inland waters are often broadly referred to as 'freshwater' due to a traditional distinction between marine aquatic environments and non-marine ones. However, in Australia, the relative abundance of saline inland waters, up to and exceeding the salinity of sea water, makes the term 'freshwater' inappropriate as a general descriptor of inland wetlands. Despite this, Australia has a variety of wetland habitats provided by areas such as the Kakadu wetlands area in northern Australia and the lower Cooper wetlands, including the Coongie Lakes, in the centre.

Many rivers in Australia are floodplain rivers, and during the wet season they break their banks to cover large areas of flat country. Floodplains are important areas for rainbowfishes moving upstream or downstream and for feeding and spawning. The floods covering floodplains in most areas are short lived. Flow in the river channel may persist for a longer period, but the period of extensive flooding rarely exceeds two to three weeks. However, this may occur several times in the course of a good wet season, but is dependent upon cyclones, rain depressions and monsoonal development. The floodplain surrounding lowland rivers contains a mosaic of habitats. All are replenished, regularly or irregularly, by flooding. Such waterbodies include intermittent lakes, billabongs (lagoons) and various types of flood runners (deep channels that only have water in them during high floods), as well as backwaters, anabranches and creeks. Equally importantly, river floodplains also contain swamps, marshes and other intermittently wetted areas, all of which play crucial roles in conserving river health. Indeed, the whole of a river floodplain can be considered a single, but extremely diverse, wetland.

Extensive floodplains in the near-coastal lowlands adjacent to some of the largest northern rivers form some of Australia's largest and most diverse wetlands. Examples include the Kakadu wetlands; in the wet season, 2,700 km<sup>2</sup> of Kakadu may be inundated with floodwaters. Floodplains on large rivers are large; when several rivers in the Gulf of Carpentaria merge in major floods they create a single, vast wetland of around 20,000 km<sup>2</sup>.

While perhaps not important as habitat for some species, floodplain and temporary wetland habitats may be important to rainbowfishes through the entrainment of terrestrial organic matter into organic food webs thereby increasing the abundance of food for larval and juvenile fish. Riverine habitats include open water areas over sandy or muddy bottom substrates. Aquatic vegetation fringes the margins and consists mainly of aquatic beds of floating leaved species and reeds. Mostly the vegetation is in a thin band along the margins, with leaf litter accumulating below. Water flow varies from permanent to seasonal and may dry back to non-flowing deeper holes during dry conditions. Some only flow after heavy rains, which may be years apart. Oxygen levels decline during the dry season; pH is mostly neutral and specific conductivity is low. Water flow is relatively slow except for short periods following wet season rainfall. Saltwater enters the lower reaches of coastal riverine habitats and they generally have some tidal movement.

Tributary streams are mostly slow-flowing, and seasonal in nature. They can be clear or turbid with fringing water plants such as waterlilies, emergent grasses, and sedges. Flowing water in the river channels provide few niches for rainbowfishes to live in, while lagoons have a diverse range of areas for breeding and feeding. Substrates are mud or silt, and there is an abundance of water plants growing to the surface around the margins. Sometimes they may have water plants growing in the deeper water in the middle. Rainforest streams are characterised by their clear water, usually high current, sparse aquatic plants, and almost complete shade of the water by riparian forest.

Lagoon (billabong) habitats differ significantly from riverine habitats. Billabongs are pools or lagoons left behind in a river or in a branch of a river when the water flow ceases. Billabongs are often formed when floodwaters recede, replenished only when the stream floods again. Flowing water in the river channels provide few niches for rainbowfishes to live in, while lagoons have a diverse range of areas for breeding and feeding. Substrates are mud or silt, and there is an abundance of water plants growing to the surface around the margins. Sometimes they may have water plants growing in the deeper water in the middle. Lagoons are habitats for decomposition of organic matter from terrestrial sources, often having a thick layer of leaf litter around the margins. In the wet season they often turn green, due to influx of nutrients in runoff water. A number of the smaller species of rainbowfishes and blue-eyes appear to be dependent upon these specialised habitats for their survival. Iriatherina werneri and Pseudomugil gertrudae are almost exclusively found in vegetated lagoons.

Swamps can be broadly defined as areas featuring permanent or temporary shallow, open water. This includes virtually any land, which is regularly or intermittently inundated. Swamps near river mouths are mostly slightly saline. Upstream swamps tend to be shallow and support mainly emergent water plants. There may be standing water in these swamps for most of the year. The ground storey may contain insectivorous plants (*Byblis* and *Utricularia* spp.), ferns, grasses, and a variety of sedges.



▲ South Alligator River Floodplain (Northern Territory) ▼ Keatings Lagoon (Cooktown, Queensland)



Large stretches of dune field and coastal heathland swamps; lakes and streams are found dotted along the eastern Australian coast. Within southeast Queensland and northern NSW, these coastal lowlands are known as the "wallum". The word 'wallum' is an aboriginal word which was used to describe the small woody tree, Banksia aemula. Over time, the use of the term has been extended to describe other plant communities, which tend to be dominated by Banksia aemula and other similar Banksia species. The coastal lowlands are distributed across low lying undulating alluvial plains (approximately 1 to 10 metres above sea level) found in behind coastal dune systems. Wallum habitat associated with the perched lake systems of the Fraser Island-Cooloola sand masses and those of Moreton and North Stradbroke Islands generally consists of extensive, dense reed beds in shallow areas of the lakes and the fringing areas of the lake support stands of Melaleuca quiquenervia.

One of the most distinctive features of wallum is the tea-like colour and low pH of the water bodies associated with this habitat. These habitats are generally acidic, have low conductivity (dissolved ions), but vary in their pH levels, dissolved organic matter, ionic composition, and colour. Factors contributing to these variations are age, formation, layers of low permeability and peats, proximity to the sea, surrounding vegetation, and the extent to which leaf litter accumulates and decays in the water. The creeks and swamps contain dissolved organic acids (humic acids) which give the waters their dark brown colour and low pH (2.8 to 6.8). The sandmass water bodies are usually well oxygenated but highly oligotrophic (low nutrient levels due to the surrounding infertile sands) and of low biological productivity. Rainbowfishes often found in these habitats include Iriatherina werneri, Melanotaenia maccullochi, Pseudomugil gertrudae, Pseudomugil mellis and Rhadinocentrus ornatus.

Major sedimentary basins where groundwater can be found extend under 60% of the Australian mainland. The Great Artesian Basin is the world's largest and deepest artesian basin. It covers an area of 1,082,400 km<sup>2</sup>. Artesian springs are found in large numbers on the fringes of the Great Artesian Basin and these springs are refuges for a variety of fish, invertebrates and plants, some of which are dependent on the springs for their survival. The spring water emerges as seepages, as flowing springs, or form pools of standing water. Depending on the rate of water flow, moderately large pools may form over the spring site, some feeding streams or tails several kilometres long. Many species of fish appear to be restricted to particular groups of springs such as *Scaturiginichthys vermeilipinnis*, the redfin blue eye, which is only found at Edgbaston Springs.

Extensive areas of intertidal mangrove forests occur at the lower reaches of coastal topical rivers in Australia. These mangrove forests are comparable in diversity to those of Southeast Asia, which are acclaimed as being among the richest mangrove areas in the world. During the wet season, freshwater flowing into these habitats dilutes the waters to nearly fresh. Water thus varies from saline through brackish to fresh. Rainbowfishes are seldom, if ever; found in these habitats, all live exclusively in fresh waters. However, they are the preferred environments for a number of *Pseudomugil* species.

Since riparian (stream-side) vegetation is out of the water for most of the year its relationship to and importance for rainbowfishes is not immediately obvious. However, this vegetation plays a critical role in the health and vitality of stream organisms by contributing to food supply, shade, protection from predators, shelter (from fast flowing water) and water quality. Terrestrial insects have been found to form a large proportion (20–50%) of the diets of rainbowfishes. Riparian vegetation also plays an important role in the maintenance of daily and seasonal water temperatures. Research in tropical Australia streams has found that cleared stream sites were  $3-5^{\circ}$ C warmer than nearby forested stream sites and the daily fluctuation in temperature was three times greater.

As well as providing food and habitat for rainbowfishes, riparian vegetation significantly influences the quality of water in a stream or river system. It does this by filtering and/or absorbing nutrients, chemicals and sediments derived from terrestrial sources. Vegetation on the banks of streams and rivers also helps to reduce or prevent bank erosion and hence sedimentation, which smothers habitat, food sources and spawning sites.

The health of Australia's fish communities has declined as the rivers in which they live have deteriorated. In a government commissioned report to identify the key threats to Australia's freshwater fisheries, Kearney *et al.*, (1999) list the following: habitat degradation; pollution/water quality/water temperature; reduced environmental flows; barriers to migration and introduced species.

Rainbowfishes have a number of basic habitat requirements in order to survive from day to day, to breed and to maintain long term populations. A change in any one of these factors can reduce the ability of the population to survive in the longer term. A reduction in two or more factors will seriously threaten the population. A feature of any pristine environment is the huge variety of habitats that are available. Rainbowfishes will not survive in bare, barren habitats. Cover is required to provide protection from predators and to provide flow refuges to prevent downstream displacement. In some cases, deep water provides adequate shelter, though more generally, rocks, logs, fallen branches, and aquatic and riparian vegetation provide the necessary cover. Without sites to deposit eggs and allow their fertilisation and development until hatching, rainbowfish populations will not survive. Important breeding sites include submerged aquatic and riparian vegetation.

Almost all rainbowfish species need to move within their habitat to access food resources, escape predation or competition, access breeding sites, escape unfavourable environmental conditions such as low or high water temperatures, and recolonise dry season habitats. Their habitat requirements are complex, and it is unlikely that they will ever be fully understood. An adequate supply of suitable quality water is the most fundamental requirement. Without this, rainbowfishes will not survive for more than a few minutes. Important water quality parameters include dissolved oxygen and temperature. Extreme levels of these parameters can cause massive fish kills.





▲ Searys Creek (Queensland) ▼ Edgbaston Springs (Queensland)





▲ "pristine" Kimberley habitat (Western Australia) ▼ "modified" habitat (Queensland)



#### **New Guinea**

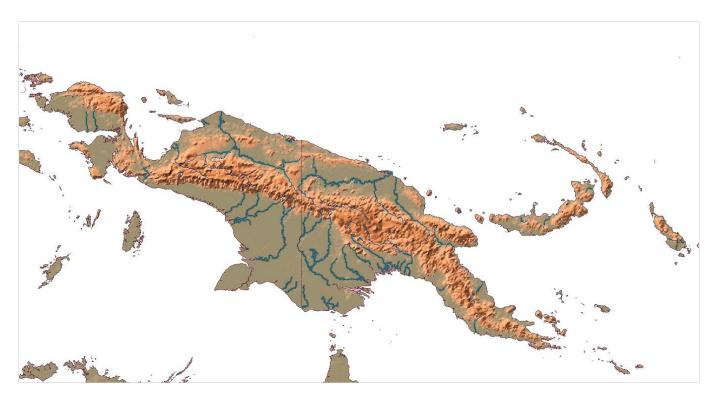
New Guinea, with a land area of approximately 876,800 km<sup>2</sup>, is located south of the equator in the south-western Pacific, just north-east of Australia. The term New Guinea refers to the entire island, consisting of both the Indonesian province of (West) Papua and the independent nation of Papuan New Guinea (PNG). The political boundary follows the 141st meridian east of Greenwich (141°E). The border does not actually run through 141°E all the way. From the northern coast near Wutung, the border follows 141°E until it reaches the Fly River, and then follows the river southward until it reaches 141°01'E, and then along 141° 01'E until it reaches the southern coast near the mouth of Bensbach River. To the north, a small section of the Sepik River also lies to the west of 141°E between 4°S and 5°S. Nevertheless, no such compromise has been made to the borderline at the two points where the Sepik River crosses 141°E.

The mainland of New Guinea and its associated archipelagos stretch across a distance of almost 3,000 km between the equator and  $12^{\circ}$  south on the south-eastern rim of the Pacific Ocean. The climate is basically equatorial and it is hot, wet and humid throughout most of the year. Rainfall is heavy throughout the island but sharply seasonal in character, with a relatively dry period between June and August. New Guinea has a mountainous cordillera which runs along its centre and here ridges rise to 4509 metres at Mount Wilhelm the highest point in Papua New Guinea and to 4884 metres at Puncak Jaya (formerly Carstensz Peak) the highest point in West Papua. These highland regions are cooler and less humid but generally equally wet. Temperatures vary along an altitudinal gradient, the hot  $(25–30^{\circ}C)$  wet tropical climate of the coastal plains giving way to much



cooler (12°C at 3000 metres) conditions in the highlands. On the south coast of West Papua, the mountains rise up in abrupt steep slopes from the alluvial plain. Here it takes less than half an hour's drive along the Freeport road to pass from the tropical rainforest to the montane moss forest. Not much further along the road, Alpine vegetation takes over, followed by glaciers. The distance from the glaciers to the Arafura Sea is but one hundred kilometres. No other region on earth offers such fast ecological transition.

New Guinea offers several major land forms, each with many different ecological zones for its biota. The southern plains and lowlands stretch in various widths from the Arafura Sea to the central mountain chain. Toward the centre of the island, this can be as much as 400 kilometres, narrowing to the east and west. Most of this lowland area is alluvial, made up or





▲ Fly River (Middle Floodplain) ▼ Lake Murray (PNG)



eroded material from the mountains. Three quarters of the country is covered by tropical rainforests and the remainder consists of flat grassland, lowland floodplains and the world's largest and most diverse mangrove area. While a relatively small portion of south-central New Guinea can be classified as savannah, most of the areas close to Australia are covered with tropical rainforest. Today, we find similar tropical rainforest in only two very small areas in the northeast corner of Australia.

Biographically and geologically, New Guinea is physically divided by an extensive mountain range that extends from the Vogelkop Peninsula to the Owen Stanley Ranges in the south-east. The mountain range varies in width from some 50 km to around 200 km. This mountain chain forms the backbone of the island and divides the island into a northern and a southern zone. The mountains, rivers and valleys all act as biological barriers to the movement or migration of plants and animals around the island. Indeed, geologically, the island is extremely complex, comprised of many terrains that have accreted. The biogeography of the island often reflects the independent evolutionary history of these different terrains. The complexity of the province's biogeography contributes to its rich biodiversity. There are several areas that have particular potential as reservoirs of undiscovered species.

The mountainous topography, in combination with high rainfall, results in numerous drainage systems, and a large array of freshwater habitats that include short coastal streams, large lowland rivers, coastal swamps and floodplain lakes, alpine streams and lakes, and large highland rivers. Both the northern coastal plain and the interior highlands feature numerous lakes. The southern costal plains have extensive and inaccessible swamps and mangrove forests. The central cordillera serves as the major divide causing all river systems to drain either northward or southward.

New Guinea is drained by 6 major and numerous minor river systems. Three of these, Mamberamo, Sepik, and Markham, flow northward. The remainder, Digoel, Fly, and Purari flow south into the Arafura Sea, Gulf of Carpentaria and Torres Strait. As a result of the high rainfall and rugged topography, most rivers in New Guinea have large flow volumes and high sediment loads, and are generally fastflowing and turbulent. However, water levels in the main rivers fluctuate dramatically through the year, creating a variety of aquatic habitats including swampy and flooded forest, swampy grasslands, oxbows, and small lakes.

Mangroves, brackish swamps, freshwater swamps and alluvial plains account for 7.5 percent of the total land area of the country. Lowland freshwater wetlands are a mosaic of open water, herbaceous swamp, swamp savannah, and woodland. Soft, slightly alkaline water chemistries characterise the larger rivers and most lakes. Acidic conditions are found in many small creeks flowing through swamps or intact forest along the coast.





Gerald Allen divides New Guinea into several major ecosystems to better delineate the freshwater fish species in each. Blackwater streams (so called because of their colouration due to the tannins leached from decomposing vegetation) commonly hold rainbowfishes, gudgeons and gobies. These rivers are generally richer in fish fauna than the large muddy rivers. In the lowland rivers, with turbid waters and silty or muddy bottoms, the aquatic vegetation is poor, and thus less fish life. The floodplain lakes, swamps and backwaters cover huge areas with good quality water rich in aquatic plants providing ample hiding places for juveniles. Common here are rainbowfishes, gudgeons and gobies and the ubiquitous catfishes. In the upland tributaries, with very clear water rapidly changing level and a general lack of aquatic plants, we find rainbowfishes, hardyheads, gudgeons (especially the genera Oxyeleotris and Mogurnda), and gobies.

The Wallace Line (or Wallace's Line) is a boundary that separates the zoogeographical regions of Asia and Australasia. West of the line are found organisms related to Asiatic species; to the east, mostly organisms related to Australian species. The line is named after Alfred Russel Wallace, who noticed the apparent dividing line during his travels through the East Indies in the 19th century. West of this line lies the Indo-Malayan region, which includes the Greater Sunda Islands of Java, Borneo and Sumatra on the Sunda shelf; to the east lies the Australasian region. During the Pleistocene period some 10,000 years ago, sea-levels were much lower than at present. The Greater Sunda Islands were connected by dry land to the Asian mainland, while New Guinea and the Aru Islands were joined to Australia. Wallacea, comprising the Lesser Sunda Islands (Nusa Tenggara), the Moluccas and Sulawesi, has had no recent land connection to either continent. The islands of the Sahul shelf which include Waigeo, Batanta, Salawati and Misool to the west; and Japen to the north in Cenderawasih Bay all had recent intermittent land connections with mainland New Guinea. Those which lie off the Sahul shelf had no connections with New Guinea in the recent past.

Because of its mountainous terrain and consequent abundance of isolated freshwater drainage systems, New Guinea represents a particularly rich area for rainbowfishes. More than 80% of the known species of rainbowfishes are found in New Guinea, and no doubt more will be discovered as a result of future systematic surveys.

The freshwater ichthyofauna can be clearly divided into two biogeographical regions. Freshwater bodies to the south of the central cordillera have an ichthyofauna closely allied with that of northern Australia, reflecting a former land connection. While several of those species with diadromous habits can be found in both southern and northern rivers, the



fish permanently inhabiting freshwater in the north are invariably different species from those in southern water bodies. Apart from the land barrier formed by the central cordillera, northern rivers are much younger than southern rivers. Of those fish families common to both northern and southern rivers, species diversity is invariably lower in the north. Only two species, *Chilatherina campsi* and *Oxyeleotris fimbriata* have managed to 'cross' the central mountains as they are found both in northern as well as southern drainages.

Existing knowledge of the fishes of New Guinea have been published in field guides and checklists, but more research remains to be done. Freshwater fishes of New Guinea were reviewed by Allen (1991), who listed 320 species, including some estuarine forms; subsequent collecting and research have taken this count to around 360 of which approximately 46% are endemic. Of great concern is the observation that this endemicity is matched by the numbers of exotic fish (17%), introduced since the 1970s. The Snakehead (*Channa striata*) and Climbing Perch (*Anabas testudineus*) are particularly threatening to the native fish.

Since 1970 about 130 New Guinea freshwater fish species or subspecies have been described or are awaiting description. New species have been found and described at rates as high as any in the history of New Guinea ichthyological exploration. In general, the fish fauna of New Guinea is closely related to that of northern Australia. Nearly all the families, most genera, and numerous species are shared between these two areas. As in Australia, the most diverse taxa in New Guinea are also Eleotrididae and Gobiidae, with about 115 species, followed by Melanotaeniidae with about 60 species. About 50 species from southern New Guinea also occur in northern Australia and are restricted to these two areas (Lundberg *et al.* 2000).

There has been a pronounced renewal of interest in the freshwater fauna during the past 30 years, in part due to the development of an efficient air transport network, as well as road construction in previously inaccessible districts. The number of fish species found in New Guinea seems to increase each time a qualified collector enters a new area.

Gerald Allen has added at least 80 new fish species to the scientific literature. This includes about 40 species of rainbowfishes (including one new genus), 9 species of blueeyes, 14 species of gudgeons, 9 species of gobies, plus various species in other families.

Most recent research has involved Papua New Guinea, the island's eastern half. Comprehensive surveys have been conducted for the Fly, Purari, Laloki, Kikori, Sepik, Ramu, and Gogol rivers, as well as many other regions. As a result of these investigations there now exists a fairly comprehensive knowledge of the fishes inhabiting the eastern half of the island. Unfortunately, the western half, the Indonesian province of West Papua, remains poorly studied. Our knowledge of the fishes of this vast area is still largely based on the now out-dated work of the early Dutch explorers. The Timika region and sections of the Mamberamo basin have been sampled, but most regions still remain unsurveyed.

Since the 1950's, more than thirty species of freshwater fish have been introduced into New Guinea waters. Not all of the introductions were successful however, but more can be expected, especially in the Indonesian province of (West) Papua. Most of the introductions have had a negative impact, either by competing for space and limited food resources, or by feeding on natives species, including their eggs and fry.

Several species including *Oreochromis mossambica*, *Clarias batrachus*, *Cyprinus carpio*, *Channa striata*, *Tor putitora* and *Anabas testudineus* appear to be undergoing rapid population increases and therefore pose a serious threat to native fishes. *Oreochromis mossambica* and *Cyprinus carpio* have established self-recruiting populations in almost all the lakes and have subsequently become the dominant species in their respective fisheries. Although the invasive fish species already present in New Guinea appear to be undergoing population expansions the specific impacts of such species on aquatic organisms endemic to New Guinea have for the most part not been determined.





# Rainbowfishes Collecting & Shipping



Photo: Jennifer Palmer

While aquarium shops or fellow enthusiasts can offer a wide selection of rainbowfishes suitable for stocking your aquarium, if you live in the appropriate area, you may wish to collect your own. Field collecting is an interesting, educational, and enjoyable activity for aquarists so long as a few simple but important rules are followed. Collecting rainbowfishes from their natural habitat is only the first step, the second, and perhaps the most rewarding, is keeping and breeding them once they have been established in your own aquarium.

Australia still has many pristine freshwater habitats where one can find rainbowfishes. Visiting one of these unpolluted streams, collecting a new species for your aquarium, or just studying the aquatic flora and fauna, I'm sure, is the dream of every rainbowfish enthusiast. As well as major and minor rivers, the coastal plains are threaded by innumerable smaller streams, lagoons and swamps. These areas contain different species of fish, crustaceans, plants and many other forms of aquatic life. Within each of these environments there are different habitats that influence the fish species composition. There can also be large seasonal changes in the composition and abundance of fish communities at different times of year.

This raises the issue of deciding on when is the most appropriate time to collect. It is advisable to schedule your collecting when fish abundance and diversity is known to be highest. Avoid collecting at times when conditions are out of the ordinary, like during drought or flood periods. Safety is also important when collecting fishes from secluded areas, always have a good quality first aid kit and wear suitable footwear when collecting in water.

Rainbowfishes have strong associations with certain habitats. The major habitat features influencing rainbowfish distribution are temperature, water quality, depth, current and density of aquatic vegetation. Therefore, the collecting should include a range of habitats present at a site. In deep pools of streams this would include the deep open water zone and the margins, both shallow and deep, with associated vegetation, woody debris and rocky substrates. In flowing streams shallow riffles and runs should be sampled as some species aggregate in the faster currents present in these areas. In lagoons both the open water zone and densely vegetated littoral zone should be sampled. These habitat features also affect the performance of different collecting equipment and methods. Consequently, different habitats may require collecting by different methods.

Many northern rivers and wetlands in Australia are dry for a large part of the year and need to be visited at an appropriate stage of the wet season when (a) water and fish are present and (b) it is possible to obtain access. Access to riverine sites is very difficult during the wet season due to high flow rates and risk of flooding that can make collecting difficult. Consequently, the end of the wet season is when access becomes possible and declining water levels make collecting easier. Fortunately this is when fish diversity is highest in lowland river habitats making the late-wet or early-dry season the optimal time for

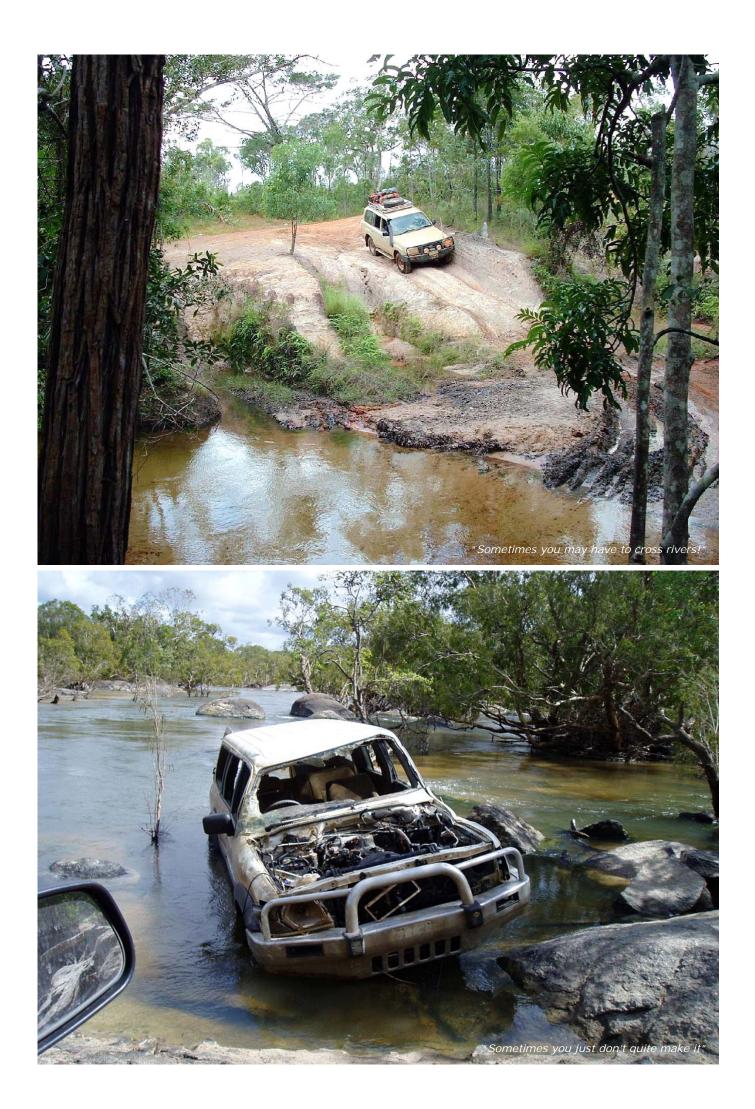
collecting from riverine locations. Later in the dry season fish diversity and abundance declines. There is also a peak in fish activity in the hour before sunrise and after sunset and some species are likely to be missed if these times are not included.

Government regulations control the collection of certain aquatic animals and plant species with regulations varying considerably from State to State. Be sure to check regulations with the local wildlife, fisheries, and/or natural resources departments. In general, a permit will be required to undertake the collection of rainbowfishes from their natural habitat. The collecting permit will probably state the maximum number of fish of any species per location that can be taken as well as the methods used to collect the fishes. In Queensland, most rainbowfish species have a take and possession limit of 20.

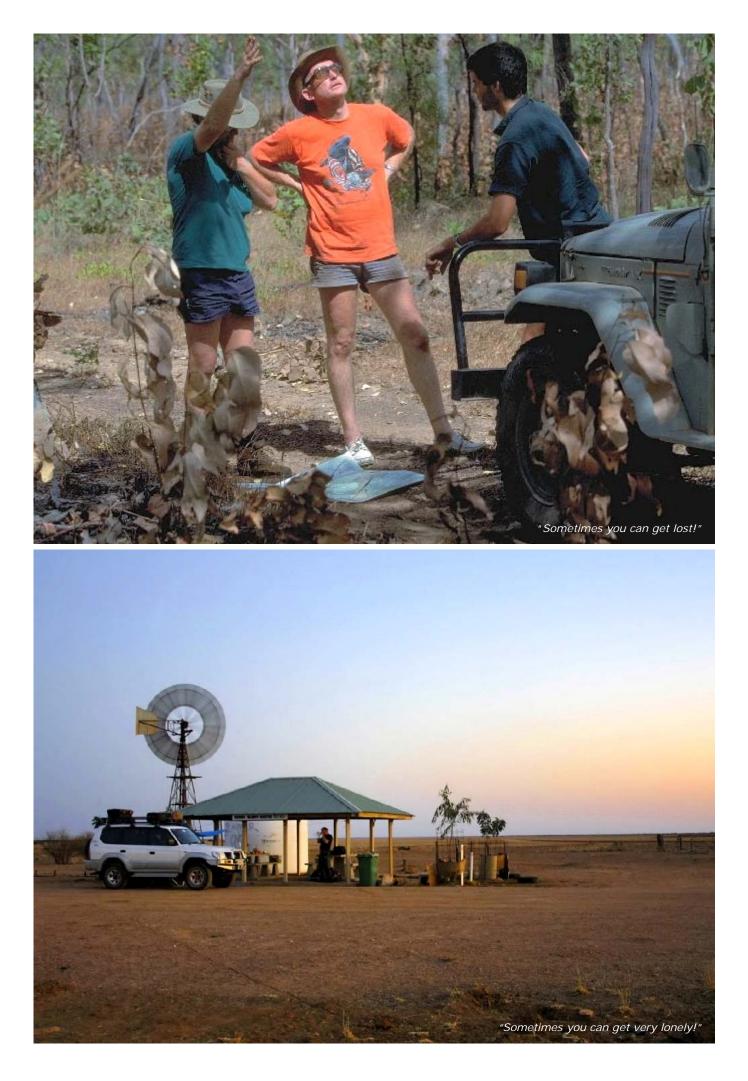
Collecting should always conform to an accepted standard of behaviour and guidelines:

- Know State and Federal Government regulations. Federal and (or) State laws govern the collection of endangered or threatened species.
- Before removing animals or plants from their native habitat, prepare a suitable environment for them and be aware of their needs for survival.
- Do not collect any restricted, vulnerable, or endangered species. Special care should be taken in areas where a threatened species of fish has previously been recorded or are predicted to occur.
- Obtain the owner's written permission to collect on private property.
- Collecting should always be conducted so as to leave the habitat as undisturbed as possible. Put rocks, driftwood, etc. back in the position in which they were found.
- Never take more than you need and do not take all specimens at a site; always leave the majority of the fauna and flora undisturbed.
- Try to keep the handling of the fish to a minimum to reduce the potential for stress and to increase the survival chances of unwanted specimens released back into the water.
- Release unwanted organisms at the collection site (only release organisms into an area from which they originated).
- Treat all animals, collected in a humane and ethical manner.











#### Equipment

The type of collecting equipment you decide to use will depend largely on the type of habitats you are surveying and your target species. Different equipment is effective under different conditions. For example, when collecting in vegetated or non-vegetated areas, you may need to use different types of equipment. I have a homemade net that consists of a rectangular stainless steel frame about 75 x 45 cm to which some soft plastic insect screen has been affixed. To complete the set-up, a long broom handle is attached to the frame. It can be used either as a long-handled dip net or without the broom handle in the same manner as a hand-held scoop net. However, the following collecting equipment can be used individually or in combination to ensure that a wide range of species can be collected, even though conditions may prevent you from using some types of equipment.

#### Seine Nets

Seine nets are lengths of netting weighted at the bottom and supported by floats at the top which is set to enclose an area and then dragged to the shore. Scientific researches rely heavily on this type of seine in certain freshwater and coastal environments, and have learned that disturbing the bottom in the path of the seine (by shuffling their feet) or adding a weight to the lead line may increase the size and diversity of the catch, especially in streams with rocky bottoms. On larger nets there is often a deep pocket built into the centre for fish to collect in and minimise their escape. Catch is dependent on mesh size but is also biased towards slower fish that are less able to avoid the net. Seine nets are good in habitats that are relatively shallow yet open water about one to two metres deep and along the edge of emergent vegetation. Small mesh seine nets can be used to collect rainbowfishes and other small bodied fish that are more abundant in shallow and bank edge habitats. Seine nets of varying lengths can also be pulled through the water with two or more people or smaller pole seines can be pushed through the water by one person. This technique is ineffective in areas densely covered in submerged plants and logs and can be difficult if used in habitats with extremely soft sediments.

#### **Dip/Scoop Nets**

The simplest of all nets, the dip net is also the most versatile, with shapes and sizes that allow use in capturing a wide variety of species. Dip or scoop netting is a useful technique in areas where water is shallow and or contains vegetation, thus making it too difficult to apply other trapping methods. These can be used to net through submerged vegetation, under and near the banks of rivers and streams. Using a mesh size of 5 to 10 mm allows the net to be dipped into and dragged through the water rapidly, increasing the chances of catching fish. Long-handled dip nets may be used from the bank, eliminating the need to enter the water.







#### Bait (Shrimp) Traps

Small bait traps (40 cm x 20 cm x 20 cm, 3 mm mesh) can be highly selective and variable in effectiveness. These work well for many rainbowfish species. They are very good in shallow littoral habitats. Bait traps may be set in areas difficult to sample by other methods and can be left while other tasks are performed or overnight. Bait the traps with dry dog or cat food and then situate them among overhanging vegetation or amongst submerged logs and vegetation. Baited traps can be set individually, or in a group attached to a set line. Ensure that you flag their location adequately so they can be found easily later. Nearly any container can be baited and used as a trap. Low numbers of fish are caught in bait traps compared to other techniques. Traps need not be baited to successfully capture fish; sometimes fish enter unbaited traps out of curiosity, for companionship (schooling behaviour), or to eat smaller organisms already in the trap. Often unbaited traps catch just as many fish.

#### Cast Nets

Cast nets are circular nets that are heavily weighted along the perimeter and thrown over fish from shore or boats. The retrieval mechanism pulls the weights into the centre of the net, trapping the enclosed fish. Cast nets are usually fished by a single individual, and typically require practice by the user to make the net take on a flat or umbrella shape when cast. Floating patches of vegetation may harbour fishes that can be collected by covering the vegetation with a circular net, and then sorting through the plant matter or shaking it.

## **Collecting Procedure**

I have found the best way to catch rainbowfishes is with the scoop net technique and bait traps. Choose a spot and go downstream a few metres. Open, reasonably shallow water is best for collecting. Manipulating a net through areas with rocky substrate, abundant aquatic plants or sunken tree branches etc. requires practise and patience. These areas are more suitable for the placement of the bait traps. Wade out and position the scoop net with the bottom edge slightly ahead of you. As you move forward the net will trap whatever is in its path. Move forward walking upstream from deeper to shallower water. When you can see some fish caught in the net, carefully lift the scoop net while keeping the fish submerged. Move to where you have stored the holding containers, and then transfer the fish from the net. Use a small plastic bowl for this purpose and avoid handling the fish by hand if possible.

Try to keep the handling of the fish to a minimum to reduce the potential for stress and to increase their chances of survival. The use of surgical gloves may enable better handling of active fish, help prevent injury to the fish during collection, and reduce loss of protective mucus, thereby minimising the likelihood of bacterial or other infections in fish following their capture. If heat and/or accumulated wastes diminish the water quality in the holding containers, partial or complete water changes can be made by using clean water from the habitat, or if necessary, water from a different habitat. Common sense should be used such as shading the holding containers from the sun. If you are collecting in the summer time and its very hot weather, the temperature in the holding containers can rise significantly. The higher the water temperature the faster the metabolic rate and oxygen requirements of the fish contained. At the same time the water's oxygen content will decrease as the temperature rises causing increased stress levels. Therefore, holding them at a cooler temperature will often mean the difference between partial losses and the total loss of specimens. Fish can be held temporarily in insulated coolers or similar containers with aeration and (or) frequently changing the water.

Accidental death of fish can occur during the collecting process. If this occurs, you could preserve some of the specimens to increase museum records on fish distribution. The preferred way of preserving fish involves storing them in 70% ethanol or 50% isopropanol for processing later. If you have difficulty in obtaining ethanol, preservation in an 80:20 methylated spirits to water mix preserves the fish adequately. If you find new species it is extremely important to get confirmation on their identification. It is also important to check your identifications for accuracy by preserving specimens or keeping photographs of the fish you can provide a well preserved sample.

Good collecting practice always involves the use of detailed field notes. Specific information about seemingly unimportant facts such as the time of day or weather conditions is often important when interpreting data. Take along some test kits to test the water parameters and a camera to record the collecting site and those wonderful memories. Recording of habitat parameters such as temperature, *p*H, etc., and collecting method is also advisable for later analysis, especially where comparison of different sites is involved. Record this information on a habitat survey datasheet.

A small glass tank comes in handy also, to be used as an observation or photographic tank. Specimens can be placed in the tank to check for skin damage or disease, and to make identification and photography easier. Colour photographs of each species collected should be taken and documented. Any fish that is unable to be identified, or of any diseased or parasitised fish should also be photographed. Photographic documentation of fish specimens can be an important means of validating field identifications. Each photograph should include an object of scale, such as a ruler, to indicate the relative size of the fish. Collectors also are encouraged to take photographs of the collecting sites. Photographs of habitats have the benefit of documenting localised conditions.

Record the dominant emergent and submergent aquatic plant species, and an approximate percentage of their coverage of the habitat's surface area. Identify plants to the genus level or to species level if possible. If required, a representative specimen of all unidentifiable plant species may be collected and submitted to a herbarium. Do not leave them out in the sun even briefly as they will wither very quickly and become useless as specimens. Generally, the whole plant should be collected. Some groups cannot be identified to species without mature fruits or flowers. Emergent plants should not be submerged, but kept in a bag with a little water in the bottom to maintain high humidity. It is best to keep each species in its own bag.





# **Transporting Home**

Netting rainbowfishes from their natural habitat and transporting them home often causes them considerable stress. The effects will vary from species to species and from fish to fish. The intensity of the stress created will determine whether the fish lives, dies immediately, or dies later. The most common cause of death among newly caught specimens is over-crowding in the holding or transport containers. Collect only a small number for your own requirements and you will have a much better chance of success.

Fish are coated with a protective layer of slimy mucous that protects them from infection, certain parasites and the effects of water. Any handling of the fish removes this mucilaginous layer and when the animal is returned to water it suffers "waterburn", which is similar to sunburn that humans experience. Without their protective mucilaginous layer, the fish are prone to infection and disease. Further, minor scratches and wounds as a result of the fish thrashing about when collected may take longer to heal and may be predisposed to infection.

The effects of handling can be reduced by the addition of sodium chloride (salt) to the transport water. Salt will often reduce the effects of stress, osmotic imbalance, and surface damage. Bacterial and parasitic infection can also be reduced by the addition of ordinary salt. The higher the water temperature, the more salt is required, and a salt concentration of 7 grams/litre has been used with success at temperatures of 25°C. A salt concentration of 3 grams/litre was found to be sufficient at lower temperatures of 10°C while 5 grams/litre can be used at intermediate temperature levels. Commercial salt (about 98% NaCl), cooking salt or ordinary table salt can be used for this purpose.

Methylene blue and salt can also be used to prevent the proliferation of bacteria during fish transport. Research has shown that both methylene blue and sodium chloride were effective in reducing bacterial load during transport of fish. Methylene blue is a redox dye which raises the oxygen consumption of cells. This means that the hydrogen to be oxidised is passed on to the oxygen. Each molecule of the dye is oxidised and reduced about 100 times per seconds. Thus, while disinfection results from this, methylene blue is also excellent against methaemoglobin intoxication (methemoglobinemia) during transport. Dosages rates of 2 g/L sodium chloride and 1 mg/L methylene blue are generally recommended, while 1 g/L sodium chloride and 3 mg/L methylene blue have also been used.

While out collecting, try to collect some natural water from the site to take home. 20 litre buckets with tight fitting lids are perfect for transporting water. The reason for doing this is to provide similar water conditions for the fish when you return home. Try to collect about two-thirds the capacity of the aquarium in which they will be housed on arrival at home.

By far the best method for transporting rainbowfishes home is polyethylene bags packed in styrofoam boxes. With the lid in place, there will be little temperature change to worry about, and the fish will remain restful in the darkness. Clear, clean water for the bags should be collected in buckets before anyone enters the stream and stirs up any sediment. The buckets are placed aside in a shady position until the fishes are ready to be bagged. If you have a battery operated air pump to provide aeration, then all the better, although it is not usually necessary.

The arrival of the fish at home can be the most critical stage of the collecting process. The rainbowfishes will be under some degree of stress and sudden exposure to water of differing characteristics can further stress the fish, often beyond what they can stand. Draining two thirds of the water from the aquarium and refilling it with the water that you bought home can ease the harmful effects of differing water conditions. Carefully place the fish into their new aquarium, which incidentally, should be a quarantine tank. All wild-caught rainbowfishes should be quarantined for at least three weeks. As you do your normal water change routine on the quarantine tank, you will slowly replace the stream water with your local tap supply. This makes it easier for the newly captured fish to adapt.

The vast majority of problems with wild-caught rainbowfishes having settled into an aquarium can be traced directly or indirectly to poor aquarium management. Good aquarium management is keeping the tank conditions relatively constant. Many well-meaning hobbyists in their quest to duplicate the water conditions of the fish's natural habitat fiddle about with various chemicals and compounds trying to maintain pH and hardness levels. Duplicating the water conditions under which the specimens were collected may seem ideal, but this often causes more problems than it solves.

#### **Consistent Biological Nomenclature**

Biological nomenclature used to document fish species collected should follow the "Australian Fish Names Standards" (AS SSA 5300-2007), or the "Codes for Australian Aquatic Biota" of common and scientific names published by the CSIRO Marine and Atmospheric Research Committee. Use of the standardised list of common and scientific names is critical to maintaining consistency and uniformity across Australia. New fish species are occasionally discovered, and systematic studies frequently lead to changes in fish taxonomy and nomenclature. Thus, the most recent edition should be used. Codes for Australian Aquatic Biota - is a continuously maintained and expanding 8digit coding system for aquatic organisms in the Australian region maintained by CSIRO Division of Marine and Atmospheric Research. Initially developed to cover fishes and selected other organisms of research or commercial interest, it has more recently been expanded to provide more comprehensive coverage of a number of aquatic groups, as information is available.



Shipping live fishes is another feature of the rainbowfish hobby, with enthusiasts exchanging fishes with each other. The system used for packaging rainbowfishes for air transport is a closed one in which all factors to meet the requirements of the fish for survival are self-sustained. Rainbowfishes are usually transported in sealed plastic bags containing small quantities of water and pure oxygen. Excess air is removed from the bag and replaced with pure oxygen. The bag is sealed, placed in an insulated container and finally into a cardboard shipping box and shipped.

During the 1920-30s fish shipments were in old-fashioned flat "German" cans, with a small neck and very wide body to give maximum air surface. Other shipping containers used in the 1920-30s were merely a straight-sided metal pail with a tight fitting lid that was packed into a wooden shipping box. The wooden shipping box was lined with cane fibre-board for insulation and the metal container was packed inside it in sawdust. As sea voyages were long and no oxygen was used, the fish generally arrived in poor condition. However, some survived the journey and were bred by experienced hobbyists.

The aquarium hobby didn't really recover until the 1950's when shipment with plastic polyethylene bags and oxygen, in insulated containers was developed. This, together with faster air travel, which allowed a much wider variety of fish to be successfully imported from distant regions of the world, consequently attracted new hobbyists and enabled the aquarium hobby to flourish. The polyethylene-bag transport system had greatly reduced the shipping weight of aquarium fish consignments, and made them a feasible option for air transport. Nevertheless, the freight cost of fish consignments is still a major cost of the aquarium hobby. For consignments from Asia to Europe and the USA, shipping cost is often more than the fish in the consignment.

The key limiting factor for the live-fish transport system is the deterioration of the water quality due to accumulation of metabolic wastes. However, a variety of techniques have been developed to manage the quality of water during transport. These include starving fish before packaging, lowering the temperature of transport water, addition of anaesthetics, ion exchange resin, buffers or drugs in the transport water.

Airfreight shipments have to comply with the IATA Live Animal Regulations. The outer container should be made of sturdy expanded polystyrene or Styrofoam with an inner plastic liner. Shipping boxes used must be an approved type. Care must be taken to ensure no sharp edge punctures the inner plastic bags that can expand from changes in altitude. The inner bags should be plastic (polyethylene) and fastened by twisting the top and folding the twisted part so that it can be sealed with elastic bands. Each bag is then doubled bagged with a similar size bag to prevent leakage of water. Bags may be double or triple layered, with newspaper between the layers to prevent punctures and leakage from fish spines. The fish can be bagged singly or in small groups, depending on the species and the relative sizes of the specimens. The bags can contain trapped atmospheric air or you can use bottled oxygen - with just enough water to completely cover the fish. If properly packaged, rainbowfishes can be expected to survive for at least 48 hours in the shipping box. Common packing densities used in the commercial shipping of rainbowfishes and based on a single seven-litre capacity size bag is 150 individuals at 25 mm size, 100 at 50 mm, and 50 at 65 mm. After packing, the bags with fish are placed in an expanded polystyrene foam box, usually four to eight bags to a box, to provide thermal insulation to prevent sudden changes in temperature of the transport water, especially when the consignments are in the cargo hold of aircraft during air transport.

The health of the fish can be affected by changes in water quality parameters while in the plastic bags during the shipping process. The parameters to be considered are temperature, dissolved oxygen, pH, carbon dioxide, ammonia, and the salt balance of the fishes' blood. The rate of change of each parameter is affected by the weight and size of fish to be transported and the duration of transport. In order to implement a successful shipment you must first have an understanding of what changes will take place, chemically and physically, inside the shipping bag during the transport period.

The most important single factor in transporting rainbowfishes is the provision of adequate concentrations of dissolved oxygen. The importance of supplying adequate levels of dissolved oxygen cannot be over emphasised. Failure to do so results in severe stress, which may cause the fish to die two or three days after transport. The amount of oxygen that can be dissolved in fresh water is based primarily on water temperature. The water is referred to as 100% saturated when the upper saturation level is reached.

The volume of pure oxygen supplied to the transport bag by commercial operators used to be up to six times the volume of transport water, but it has now been reduced to three to four times the volume of water. Even at these reduced volumes, dissolved oxygen content is never a limiting factor. Several fish packaging experiments have recorded over-saturated oxygen content of above 10 mg/L (@ 25°C, 100% saturation = 8.26 mg/L) after 24–48 hours shipment, even then there were high fish mortalities of up to 20%.

Dissolved oxygen saturation is higher for cool water than for warm water. If pure oxygen is used during bag transport, then low oxygen levels usually should not be a problem unless the bag is improperly sealed or develops holes caused by the spines of large fish. Nevertheless, it is important to have a 75 percent volume of oxygen in the bag to insure adequate diffusion of oxygen at the surface of the water.

Once a bag has water, fish and oxygen sealed inside it, certain chemical changes take place due to the metabolism of the fish. When fish breathe, they absorb oxygen and excrete other gases



and metabolites, primarily carbon dioxide (CO<sub>2</sub>) and nitrogen in the form of ammonia. Total ammonia nitrogen consists of two forms that exist in a pH and temperature dependent equilibrium of unionised ammonia (NH<sub>3</sub>) and the ammonium ion (NH<sub>4</sub><sup>+</sup>). The unionised form (NH<sub>3</sub>) is toxic to fish while the ammonium ion (NH<sub>4</sub><sup>+</sup>) is relatively non-toxic to fish. It can, however, in high concentrations, produce external burns that are identical to acid burns. This is often seen when fish are crowded in shipping bags.

The proportion of  $NH_4^+$  to  $NH_3$  increases with decreasing pH and decreases with increasing pH. The percentage of  $NH_3$  also rises with increasing temperatures - so conditions with both relatively high pH and elevated temperature are especially dangerous. Since  $NH_3$  cannot be measured directly, several tables have been created based on an equilibrium formula that predicts the relative percentages of unionised ammonia at different temperatures and pH. Total ammonia concentrations may reach more than 14 ppm during transport. However, the easiest way to reduce toxic ammonia build-up in transport water is to lower the temperature of the transport water and to stop feeding 48 to 96 hours prior to shipment.

As the fish respire they produce carbon dioxide as a byproduct. Carbon dioxide reacts with water reducing the *p*H. If the alkalinity of the transport water is less than 100 ppm, some type of buffering compound should be added to the water. Properly buffered water will help remove free carbon dioxide, which causes drops in *p*H. High levels of carbon dioxide (greater than 20 ppm) will interfere with the oxygen uptake in the fishes' blood. Cation exchange resins such as clinoptilolite, a natural zeolite, is commonly used to remove ammonia from the transport water. The resins have the ability to absorb ammonia by selective ion exchange. They are either wrapped in a net bag or added directly into the transport water at 15–20 g/L of water. They do not however, control the accumulation of carbon dioxide in the transport water.

Osmoregulatory dysfunction is common in fish that are exposed to transport stress, and the addition of salt to transport water is effective in reducing the osmoregulatory dysfunction and other physiological responses to stress, resulting in reduced fish mortality. Freshwater fish have a blood salt concentration higher than the salts of the transport water. Therefore, the fish are continually losing salts to the surrounding water. Commercial exporters commonly use coarse salt containing 95–98% sodium chloride to reduce the effects of stress on the fish. It is added directly to transport water at 0.5–3.0%, and the concentration used varies with species and exporters.

Bacterial growth is another major source of metabolic wastes. Bacteria not only increase the ammonia load and compete with fish for oxygen in the transport water, but also weaken or cause diseases. Drugs such as antibiotics, methylene blue and acriflavine are commonly added to the transport water to control bacterial growth. Methylene blue and salt can also be used to prevent the proliferation of bacteria during fish transport. Research has shown that both methylene blue and sodium chloride were effective in reducing bacterial load during transport of fish. Methylene blue is a redox dye which raises the oxygen consumption of cells. This means that the hydrogen to be oxidised is passed on to the oxygen. Each molecule of the dye is oxidised and reduced about 100 times per seconds. Thus, while disinfection results from this, methylene blue is also excellent against methaemoglobin intoxication (methemoglobinemia) during transport. Dosages rates of 2 g/L sodium chloride and 1 mg/L methylene blue are generally recommended, while 1 g/L sodium chloride and 3 mg/L methylene blue have also been used.

On receipt of the fish the plastic bags containing the fish should not be floated in the aquarium despite popular belief. Polyethylene bags do not allow the transfer of liquid-to-liquid or air-to-air. Apart from temperature, a plastic bag floating in the aquarium does not allow the fish to become adjusted to the water conditions of the aquarium. When the fish are released from the bag, they will still be subjected to the stress of differing water conditions.

The best way to reduce the harmful effects of differing water conditions is to empty the contents of the bag into a clean bucket or other suitable container especially kept for aquarium use. Then, add small amounts of water, at intervals of 5 minutes, from the aquarium into which the fish will be placed, into the bucket or container until a 50-50 water ratio is achieved. Place the fish into the aquarium and dispose of the water in the container. Do not add the container water into your aquarium.

For anyone contemplating sending or taking live rainbowfishes (or eggs) out of Australia please note that the export of all native fishes requires a permit from the wildlife protection authorities. Approvals to export live rainbowfishes may be given where the fish were produced lawfully in an aquaculture facility operating in accordance with the relevant State Fisheries Acts and subordinate legislation, or taken under an approved collecting permit. To export captive bred fish, a copy of a State aquaculture permit indicating the species held in captivity will need to be provided with the application to export. The relevant authorities of the States and Territories control the collection of rainbowfishes from the wild and permission will only be granted where the taking of the species from the wild is not detrimental to the survival of that species or its habitat.

The export of native fishes from Australia is controlled by the Environment Protection and Biodiversity Conservation Act and Regulations. Controls under this Act apply to museums, zoos, scientific institutions, commercial organisations, tourists, and the general public. This Act regulates the importation and exportation of most live animals and plants. These controls are in addition to those exercised under the Quarantine Act.

All aquatic species exported from Australia also are required to meet the importing country health requirements. An export permit and a health certificate must be prepared by the Australian Quarantine and Inspection Service (AQIS) prior to export. If there are no importing country health requirements then no export permit or health certificate is required and AQIS is not involved in the export. The receiving countries set the requirements for imports and in Australia this function is managed by AQIS. However, always check with the relevant authorities, as these rules and regulations are subject to change.



# Rainbowfishes Keeping & Caring



Photo: Hans Booij

Keeping and caring for rainbowfishes in captivity is relatively simple as far as general aquarium conditions are concerned they are one of the easiest fishes to maintain. Nevertheless, rainbowfishes do have a number of basic requirements in order to survive from day to day, to breed and to maintain populations over the longer term. They require suitable habitat, proper nutrition, and a stress-free existence if they are to grow and remain healthy.

If you want to be successful at keeping rainbowfishes then you must make an effort to learn some basic aquarium principles to maintain and care for them. More than a few fishkeepers have watched the inhabitants of an unhealthy aquarium die because they had failed to learn the basics. If you do not have time to learn the basics, then I suggest that you seriously reconsider the idea of keeping rainbowfishes in captivity. Practical experience can only be gained through time and exposure to the aquarium environment. It's a combination of learning aquarium disciplines and experience that will make you a successful aquarist. The actual learning that is required is relatively easy and the rewards so much greater.

In order to design more natural aquarium environments it is essential to have a broad understanding of the biology and ecology of the fish species in question and especially the environment that the species inhabits. A feature of any pristine environment is the huge variety of habitats that are available. Few species of rainbowfishes flourish in bare, barren habitats. Habitat is provided by rocks, logs, fallen branches, aquatic and riparian vegetation. However, many aquarium environments are completely devoid of structure. They are usually featureless, monotonic enclosure with no opportunity for the inhabitants to display any natural behaviour. They bear no resemblance whatsoever to the fish's natural environment and densities can be up to 100 times greater than those in nature. Reductions in density alone seem to have mixed results but it appears that intermediate densities will produce better quality fish.

Simple measures like increasing filter performance, providing dark backgrounds, natural substrate material, submerged structures, such as driftwood, and floating plant cover could improve the aquarium environment. This also leads to lower levels of aggression, better health and improved growth rates. Aquarium-reared rainbowfishes that spend some time in outside ponds improves growth, colour and survival rates substantially. In the pond environment the fish are not only exposed to natural temperature and light fluctuations, they also have access to a range of naturally occurring live foods.

Successful set-ups can range from a standard 55-litre aquarium with a modest box filter costing very little, to a huge 650-litre aquarium with elaborate trickle filtration, carbon dioxide injection and computer operated lighting systems. Some hobbyists believe that the more expensive the life support system is, the better their success will be. However, all aquariums despite how simple or how specialised, share the same principles for maintaining rainbowfishes successfully. When choosing an aquarium the largest size one can afford is usually recommended. However, any size aquarium can be successfully maintained, although aquariums smaller than 55 litres are not worth considering. The problem with small aquariums is that the environment can degrade so rapidly that the hobbyist is unable to solve the problem in time to save the inhabitants. The small 55-litre aquarium is often the most popular size with beginning hobbyists because of its moderately low price. However, problems start when they assume that this size aquarium can do more than is realistically possible.

Rainbowfishes are mid-water to surface swimming fish and require sufficient space to swim. They are also highly social and form schools for most of the time. It is obvious, therefore, that the shape of the aquarium in which rainbowfishes are kept is of great importance to them. In a small aquarium, they will feel stressed, resulting in health problems. The actual surface area of an aquarium is more important than depth. The depth of the aquarium is of no importance, except that it should never be so shallow nor so deep that the aquatic plants fail to grow properly.

An aquarium depth for rainbowfishes may be anything from 30 cm to 60 cm, in proportion to its superficies. Aquarium width should be at least 45 cm, particularly for the larger species. Rainbowfishes can jump, and it is therefore important to keep the aquarium covered at all times. Covers should be made of clear plastic or glass and fit tightly because rainbowfishes can escape through even relatively small slits. It is advisable to use extra strong tape to secure one end of the lid to the aquarium.

# Lighting

Adequate lighting should be installed above the aquarium and is best kept on a 14-hour light – 10-hour dark cycle, using appropriate timers. Although rainbowfishes will also do well in dim light, proper illumination of the room is generally desirable. Light (excessive or rapid changes in intensity) should be minimised. Rainbowfishes will often dash frantically about the aquarium injuring themselves in response to normally harmless stimuli such as turning on the fishtank light in a dark room. Red-light lamps can be installed in fishrooms that will allow hobbyists to perform tasks at night time while the fish are 'sleeping'.

## Water Quality

The all-important factor in successfully maintaining rainbowfishes in captivity is water quality. Without this, rainbowfishes will not survive for more than a few minutes. Water quality is important to rainbowfishes as air quality is to you and me. Every particle of waste matter in the aquarium will affect the health of your rainbowfishes, and determines not only how well they will live and grow, but also whether or not they survive. Some water quality factors are more important such as nitrogenous waste levels, dissolved oxygen, pH and temperature. Others, such as alkalinity, hardness, and clarity have some affect, but usually are not significant. Each water quality factor interacts with and influences other parameters, sometimes in complex ways. What may be harmful and cause mortalities in one situation can be harmless in another. Regular testing of your aquarium water and your water source is important and it should allow you to detect and correct problems before your fish are adversely affected. Therefore, knowledge of water testing procedures and interpretation of the results are important for the successful maintenance of rainbowfishes in captivity.

The first consideration is the availability of a good quality water supply to fill the aquarium. Surface water from a natural stream or pond is not recommended as it may contain contaminants, diseases, pests or parasites, any of which may harm the aquarium's ecosystem. Rainwater drawn from a wellestablished water-tank can be used. However, rainwater is not pure water because it has gases and a range of other particles from the atmosphere dissolved in it, which may include carbon dioxide, oxygen, nitrogen oxides, sulphur dioxide, dust, pollens, bacteria and numerous other compounds. This difference varies with location and proximity to oceans, industry, cities and other contributors. Using rainwater in the aquarium can have a number of benefits by providing a much more natural source of water for your rainbowfishes. Nevertheless, rainwater should not be used exclusively. Rainwater contains none of the essential trace elements that fish need, and also have no buffering capacity to stabilise the pH. It can however, be mixed with a certain percentage of tapwater. It is possible to produce various water conditions in this manner such as lowering the water hardness or pH of your normal tapwater. If the changes are within the parameters that the fish can adjust to, then suitable conditions can usually be established.

The most common source for water is reticulated (town) water. The domestic water supply of most cities and towns if suitable for human consumption will generally be suitable for use in the aquarium. Therefore, most hobbyists will fill their aquarium with city water drawn from the household tap. Nonetheless, municipal water supplies are typically treated with chlorine compounds to control bacteria and make it suitable for drinking. If used for fish keeping, then these compounds must be removed or neutralised with chemicals designed for that purpose. One week of continuous aeration will dechlorinate most town water supplies if the chlorine source is liquid or gaseous chlorine. However, many municipal water supply authorities have switched from using chlorine to chloramine. Chloramine is a compound formed by mixing ammonia and chlorine in water.

Chloramine is very stable and can not be easily driven off, even by heavy aeration. Chloramine is very toxic and high levels can cause all rainbowfishes to die within 24 hours. The actual toxicity will depend on the individual fish, water temperature, and dissolved organic levels in the aquarium water. Most rainbowfishes will exhibit serious signs of stress or die at levels above 0.01 mg/L. Some species are particularly sensitive and will die with even the slightest amount in their water. It is worthwhile to note here that domestic water supplies often change in character, and must be tested regularly from time to time for contaminants, changes in pH values, etc., using aquarium test kits. It is not uncommon for the level of chloramines in municipal supplies to change drastically, for example, due to some local problem and during that period additional conditioner has to be used. Private water supplies are not consistent in their output either, and should be checked on a regular basis.

Many hobbyists fiddle about with various chemicals or compounds trying to maintain what they regard as natural water conditions, but this often causes more problems than it solves. Duplicating the natural water conditions under which rainbowfishes are found in the wild may seem ideal. However, creating natural water conditions in an aquarium is almost impossible. The natural environment of rainbowfishes is vastly different from that of an aquarium. Most rainbowfishes live in the tropical and sub-tropical climates of Australia and New Guinea. They occupy virtually every type of freshwater ecosystem, from slow-flowing, acidic, tannin-stained water in coastal swamps and streams to fast flowing clear-water mountain rainforest streams; riverine habitats and their tributaries; lagoons, billabongs and the waterholes of arid desert country. Intermittent streams that are subjected to seasonal or frequent drying are also a common feature in Australian inland waters. As drying proceeds, decreases in water volume concentrate the aquatic life into a reduced area. These isolated pools tend to experience physicochemical extremes in the form of elevated temperatures, fluctuating pH levels and low dissolved oxygen.

The chemistry of natural waters inhabited by rainbowfishes depends on the equilibrium reached with the normal physical, chemical and biological characteristics of the surrounding environment. Water that flows over granite rock will tend to remain soft, low in calcium and bicarbonate. Water flowing over limestone will become alkaline with hardness depending on the amount of dissolved calcium and bicarbonate that it contains. Water with a low pH (<5.0) will dissolve naturally occurring minerals from the soils and rocks. To some extent the water quality will depend on the type of vegetation covering the watershed, since the products of plant decay will find their way into the streams draining the area. Such waters may be clear, or brown with varying amounts of dissolved humic substances. Water draining these areas will be acidic, and can be extremely so with sudden rainfall after a prolonged dry period, leading ultimately to 'blackwater events'. Documented fish kills from blackwater events are common throughout Australian inland rivers and streams, particularly during extended periods of inundation and prolonged periods of low flow. Therefore, the quality of natural water is never constant; it is constantly changing in response to daily, seasonal and climatic rhythms.

Mention should be made here of the diurnal and seasonal fluctuations of temperature. Changes in water depth and flow rates can affect the rates of diurnal warming and cooling. These physical changes as such, have little effect on the chemical quality of the water. However, they can affect the natural biological community that can live in equilibrium with the particular chemical and physical characteristics of that environment.



The natural environment of rainbowfishes is complex, and it is unlikely that they will ever be fully understood. It is not as simple as having the water at a particular temperature or pH. There are so many variables in their natural environment that are either difficult to replicate or are difficult to control in a captive environment. Most rainbowfishes can adapt to these natural fluctuations of water conditions as they occur. All the same, they will not tolerate sudden changes so well. Maintaining a constant stable environment is more important for their long-term survival in captivity. The easiest way to do this is by regular water changes.

## Temperature

Rainbowfishes are poikilothermic animals, that is, their body temperature is the same as, or 0.5-1°C above or below the temperature of the water in which they live. The metabolic rate of rainbowfishes is closely correlated to the water temperature: the higher the water temperature, the greater the metabolism, and their need to take in nutrients. Therefore the temperature of their environment is a major and even the deciding environmental factor in determining growth rate, metabolism, and nutritional efficiency. In fact, temperature will influence all biological and chemical processes in an aquarium. In tropical waters, which have prevailing high temperatures, rainbowfishes generally grow faster, mature younger, and have a shorter life span than rainbowfishes in temperate waters. Rainbowfishes do, however, display a wide range of sizes, growth rates and life spans in captivity. Water temperature also has a great influence on the initiation and course of a number of fish diseases.

In their natural environment, rainbowfishes are exposed to temperatures that vary considerably. This is dependent on the size and depth of the body of water, water flow, whether exposed to the sun or in shaded rain forest streams, the time of day and the season. A deep shaded backwater, for example, can be strikingly cooler than a shallow section that is exposed to the sun. The overall range has been reported from a low of 5° to a high of 38°C, and even higher in a shallow body of water exposed to full sun at midday. Temperatures in these water bodies commonly fluctuate from  $8-12^{\circ}C$  each day.

It should be noted that the survival rates for rainbowfishes in their natural habitats declines sharply when the water temperature is high and will often die at temperatures above 36°C. Such increases in temperature are common in tropical waterbodies of Australia during the late dry season. Although, the cause of death is more likely to be caused by lowered oxygen levels rather than higher temperature levels. Dissolved oxygen and temperature are critical water quality parameters and extreme levels of these parameters can cause fish kills.

The temperature conditions of the rainbowfishes natural environment can be subdivided into extreme, tolerable and preferred ranges. In the extreme temperature range their survival time is strongly dependent on the exposure time. In the tolerable range, the fish survives without temperatureassociated problems, while the preferred range is thought to be the range where physiological processes are optimised. Each species has a preferred or optimum temperature range where it grows best. At temperatures above or below optimum, their growth is reduced and mortalities may occur at extreme ranges. Rainbowfishes can tolerate the seasonal changes in temperature in their natural environment. However, these temperatures fluctuations are not recommended for aquarium keeping.

In their natural environment, rainbowfishes can search for more favourable conditions by moving into cooler or warmer water. However, in captivity this is not possible. In captivity, a temperature range of  $22-24^{\circ}$ C is recommended while an increase to  $28^{\circ}$ C can be used for breeding purposes. However, these changes should not be abrupt; temperature shock can occur if the fish are put into a new environment where the temperature is colder or warmer than the original water. Under these conditions fish may die, showing symptoms of paralysis of the respiratory and cardiac muscles. With young fry, problems may arise even where the difference in temperature is as low as  $1-3^{\circ}$ C.

# Water Chemistry

Water chemistry can seem intimidating at first, but once a few basic principles are memorised, it becomes quite easy. The parameters of prime concern are dissolved oxygen, temperature, pH, alkalinity, hardness, and carbon dioxide. Everyone involved in fishkeeping should invest in water quality test kits. An excellent water quality management program will result in fewer fish disease problems, better growth and less use of chemical treatments. The cost of test kits will pay for themselves many times over; both in numbers of fish saved and increased enjoyment of the hobby. For aquarium keeping, the high precision of sophisticated analytical methods is not required to make informed decisions.

Intensive stocking in recirculating aquarium systems requires frequent monitoring. If fish are maintained at high densities, then temperature, dissolved oxygen, ammonia, nitrite, and pH should be monitored regularly. At lower stocking densities, water quality parameters can be monitored less frequently. However, regardless of the frequency, monitoring should be conducted at a standard time of the day. The time of measurement and observed values should be recorded, as good record keeping is essential for successful fishkeeping.

Alkalinity and hardness can be measured less frequently, perhaps once a week, as they do not fluctuate as rapidly. Carbon dioxide should be monitored closely and the means to correct problems should be readily available. It is preferable to monitor dissolved oxygen early in the morning, when conditions stressful to fish are most likely to occur. Conversely, temperature and pH in ponds are best measured during the late afternoon. Chlorine or Chloramine levels in domestic tap water should be determined so that corrective measures can be initiated.



Numerous test kits are available on the market from which you can choose, but they can vary in quality and price. You are therefore advised to select test kits carefully and not buy the first one you see on the shelf. Outdated reagents are a major source of inaccurate tests resulting from kits that have a limited shelf life. If used beyond their shelf life, then they will not give accurate, reliable results.

Some manufacturers' print use-by dates on their kits but unfortunately, these manufacturers are in a minority. If you have a test kit without a use-by date, and you have had it for over six months, then you should replace the test chemicals. Test kits containing dry reagents are usually superior to kits that contain liquid reagents. Liquid reagents are generally less expensive but over time become unstable. Reagents in a dry form generally remain stable for longer periods. The most stable way of storing dry reagents is in sealed foil pouches; this protects them from oxygen, moisture, and light until they are used. Several aquarium test kit manufacturers have adopted this type of packaging.

## Alkalinity

Alkalinity is the concentration of bases dissolved in water and expressed as parts per million (ppm) or milligrams per litre (mg/L) Calcium carbonate (CaCO<sub>3</sub>). These bases are usually bicarbonates (HCO<sub>3</sub><sup>-</sup>) and carbonates (CO<sub>3</sub><sup>-</sup>), and, in rare instances, hydroxide (OH<sup>-</sup>) ions. These ions, called buffers, are important because they slow the rate at which the *p*H changes. The magnitude of change is determined by the water's buffering capacity or its ability to absorb acids and/or alkalis (base) and is an often overlooked, though extremely important component of *p*H balance in an aquarium. The term "alkalinity" comes from the fact that these compounds, mostly the carbonates, collectively shift the *p*H to the alkaline side of the *p*H scale. This term also has little to do with hardness, waters may be very soft (low hardness) yet have high alkalinity.

Without a buffering system, free carbon dioxide will form large amounts of carbonic acid that may potentially decrease the night time pH level to 4.5. During peak periods of photosynthesis in a heavily planted tank, most of the free carbon dioxide will be consumed by the plants and, as a

# Water Quality Factors

Water quality factors, commonly used monitoring procedures, and preferred ranges for keeping rainbowfishes in captivity. Details for specific test procedure can be obtained from the supplier or appropriate literature.

	•	• •	
Water Quality	y Factor	Test Procedure	Preferred Ranges
Alkalinit	ty	Titration	50-200 ppm CaCO₃
Ammon (ionised and ur		Colorimetric (Nesslerisation or Salicylate Electrochemical	e) No detectable level
Carbon dic	oxide	Titration	<10 ppm
Chlorine/Chlc	oramine	Colorimetric	No detectable level
Dissolved O	xygen	Colorimetric Electrochemical Titration	>5 ppm
Hardnes	SS	Titration	50-250 ppm CaCO₃
Nitrate	9	Colorimetric	<20 ppm
Nitrite		Colorimetric	No detectable level
рН		Colorimetric Electrochemical Titration	6.5-7.8
Temperature		Thermometer	22-24°C (28°C Breeding)



result, drive the pH levels above 10. Rainbowfishes grow best within a certain range of pH values and either of the above extremes can be lethal to them. In aquariums where plants are non-existent, a good buffering capacity can prevent excessive build-ups of carbon dioxide and lethal changes in pH. In planted aquariums better growth rates are attained in high alkalinity waters because phosphorus and other essential nutrients become more available to the plants.

Alkalinity is not the same as hardness. Calcium  $(Ca^{2+})$  and Magnesium  $(Mg^{2+})$  are primarily responsible for hardness. However, in most waters, alkalinity and hardness have similar values because the carbonates and bicarbonates responsible for total alkalinity are usually in the form of Calcium carbonate or Magnesium carbonate. However, waters with high total alkalinity are not always hard, since the carbonates can be in the form of Sodium or Potassium carbonate.

A desirable range of alkalinity for rainbowfishes should be maintained higher than 50 mg/L (ppm) at all times, with levels up to 200 mg/L. Alkalinity in excess of 200 mg/L won't adversely affect rainbowfishes, but it can interfere with the action of certain commonly used aquarium chemicals. Alkalinity can remain relatively constant in ponds, but will decrease steadily in non-supplemented aquarium systems. Adding buffering compounds to ponds or aquarium systems will increase the alkalinity and stabilise the *p*H. The *p*H should always be monitored during alkalinity increases, as a high *p*H increases the toxicity of unionised ammonia.

The equation below shows that carbonic acid (H<sub>2</sub>CO<sub>3</sub>) dissociates into hydrogen (H<sup>+</sup>) and bicarbonate (HCO<sub>3</sub><sup>-</sup>) ions. The bicarbonate ions can further dissociate into hydrogen (H<sup>+</sup>) and carbonate (CO<sub>3</sub><sup>-</sup>) ions. When acid (H<sup>+</sup>) is introduced into well-buffered water, carbonate ions react with the hydrogen ions to produce bicarbonate. Thus, although acid is added, no change in the overall *p*H occurs. Furthermore, bicarbonate ions act as an additional reservoir for hydrogen ions. The reactions outlined in the equation below are *p*H sensitive and shift to the right as *p*H increases.

$$H_2O + CO_2 \iff H_2CO_3 \iff H^+ HCO_3^- \iff 2H^+ + CO_3$$

Test kits measure alkalinity by titrating a water sample with an acid (usually dilute sulphuric acid) to an endpoint pH of about 4.6 (varies from 4.5 to 5.1 depending on the indicator dye used and the initial alkalinity). A pH indicator dye (usually bromcresol green plus methyl red) is added to a known volume of water (indicated in the test kit instructions), and acid is added until the solution changes colour. With the bromocresol green plus methyl red dye system, the colour will change from green to pink.

The units used to measure alkalinity will depend on the test kit. Some use milliequivalents (mEq/L), °dKh (degrees of carbonate hardness), mg/L or parts per million (meaning ppm of calcium carbonate equivalents). mEq/L stands for milliequivalents per litre. A milliequivalent is 0.001 of an Equivalent, which is the weight of substance that will react with one atomic weight of hydrogen.

Aquarists often measure alkalinity in units of °dKh which is the German scale of carbonate hardness (KH = from the German "KarbonatHaerte"). The prevalence of this measurement was due to the fact that a lot of aquarium literature originated in Germany. However, °dKh shouldn't be used anymore. Alkalinity should be quoted in milliequivalents per litre (mEq/L), or milligrams per litre of calcium carbonate (mg/L CaCO<sub>3</sub>).

The term "total carbonates" may also be used by some testing laboratories to refer to alkalinity of a solution. Some laboratories assume that all alkalinity is derived solely from bicarbonates (HCO<sub>3</sub><sup>-</sup>) and will report alkalinity as bicarbonates using ppm (mg/L) or mEq/L. To convert between these two units, use the following values: 1 mEq/L HCO<sub>3</sub><sup>-</sup> = 61 mg/L HCO<sub>3</sub><sup>-</sup>.

For aquarium purposes, you can use the following conversion factors:  $1 \text{ mEq/L} = 2.8 \text{ °dKh} = 50.04 \text{ ppm CaCO}_3$  $1 \text{ °dKh} = 17.9 \text{ ppm CaCO}_3$ 

# **Carbon Dioxide**

Carbon dioxide (CO<sub>2</sub>) is a by-product of decomposition of organic matter, fish, and aquatic plant respiration and can cause problems for most rainbowfishes if levels build higher than 20 mg/L. Most can tolerate concentrations of 10 mg/L provided dissolved oxygen concentrations are high. Well-maintained aquariums normally contain less than 5 mg/L of free carbon dioxide. Although, levels may fluctuate daily from 0 to 15 mg/L.

The presence of carbon dioxide is not usually a major problem in well maintained aquariums. However, high concentrations can lower the pH of the water and limit the capacity of fish's blood to carry oxygen. Fish are able to rid themselves of carbon dioxide through the gills in response to a difference in carbon dioxide concentration between fish blood and the surrounding water. If environmental carbon dioxide concentrations are high, the fish will have difficulty reducing internal carbon dioxide concentrations, resulting in accumulation in the blood. This accumulation inhibits the ability of haemoglobin, the oxygen carrying molecule in fish blood, to bind oxygen, and may cause them to lose equilibrium, become disoriented and possibly die even if oxygen levels are high. There is some evidence to suggest that the toxicity of carbon dioxide is enhanced by low dissolved oxygen concentrations.

Problems with carbon dioxide are only likely to develop in the aquarium when rainbowfishes are maintained under somewhat crowded conditions or if being added as a fertiliser for aquatic plant growth. Carbon dioxide can build up to significantly high levels in systems with large numbers of fish and relatively slow water turnover.

Advanced aquarists may use carbon dioxide injection systems to ensure that sufficient levels are available for the plants. Some injection systems will monitor pH levels and inject carbon dioxide as pH levels rise.



In some heavily planted aquariums, carbon dioxide can be a limiting factor. If the respiration of the organisms in the aquarium cannot produce enough carbon dioxide, plants will not be able to photosynthesise. If they cannot photosynthesise, their growth will be slow; they may lose leaves, exhibit poor colour and may even die. Carbon dioxide should be monitored in heavily planted aquariums to ensure that sufficient concentrations are available for photosynthesis. Carbon dioxide can be measured directly with a relatively easy titration test kit, which is fairly accurate.

While oxygen and ammonia levels are often viewed as critical to fish health, carbon dioxide tends to be ignored and very few aquarists regularly monitor carbon dioxide levels in most fish-only or lightly planted aquariums. This is partly due to an assumption that, if the other water quality parameters, particularly oxygen, are OK, then carbon dioxide will not be an issue.

Research would suggest that poor water quality in aquariums, particularly high carbon dioxide, may lead to an increased susceptibility of fish to pathogens which later leads to disease. By paying appropriate attention to this aspect of water quality in the early stages, this may reduce vulnerability to disease. It has also been suggested that high carbon dioxide levels may cause bone demineralisation resulting in spinal abnormalities.

Adequate aeration or surface agitation, and buffering of the water will keep carbon dioxide at acceptable levels. Adequate buffering will initially remove free carbon dioxide and store it in reserve as bicarbonate and carbonate buffers. Small water exchanges will also reduce the levels of carbon dioxide.

# **Dissolved Oxygen**

Dissolved oxygen is oxygen gas that is dissolved in water. Like terrestrial animals, oxygen is essential to the survival of fish and other aquatic organisms to live. As water moves past their gills (or other breathing apparatus), oxygen gas is transferred from the water to their blood. Like any other gas diffusion process, the transfer is efficient only above certain concentrations. In other words, oxygen can be present in the water, but at too low a concentration to sustain aquatic life. Oxygen is also required by virtually all aquatic plant life, and is important for many chemical and biological reactions that occur in water.

Light, temperature, pH, the number of photosynthetic organisms, depth, turbulence, altitude, and salinity are all factors that affect the dissolved oxygen level in water. By manipulating these factors, the dissolved oxygen in water can be increase or decreased. Oxygen is produced during photosynthesis and consumed during respiration and decomposition. Because it requires light, photosynthesis occurs only during daylight hours. Respiration and decomposition, on the other hand, occur 24 hours a day. This difference alone can account for large daily variations in dissolved oxygen concentrations. During the night, when

photosynthesis cannot counterbalance the loss of oxygen through respiration and decomposition, dissolved oxygen concentration may steadily decline. It is lowest just before dawn, when photosynthesis resumes. Low dissolved oxygen concentrations in a water body can also lead to associated toxic effects, such as the release of ammonia and sulphides from bottom sediments to the water column. The consequences of these processes are sometimes seen in natural environments where the combined effects may be the death of fish and other aquatic animals. These changes in dissolved oxygen that occur every 24 hours are called the 'Diurnal Oxygen Cycle'.

Photosynthesis is a fundamental biological process that uses light energy to produce sugar from carbon dioxide and water. Submerged aquatic plants (including planktonic algae), increase dissolved oxygen levels in water. Emergent and floating plants generally release oxygen into the air but all submerged plants release oxygen directly into the surrounding water. Under adequate light, bubbles of oxygen can often be seen on the leaves of aquatic plants. However, since photosynthesis requires light, plants don't produce oxygen during the night but respire and remove a small amount of oxygen from the water which slightly decreases oxygen levels. As a general rule, submerged aquatic plants produce about six times more oxygen through photosynthesis than they consume through respiration. However, under low light conditions these submerged plants are net consumers of oxygen.

Oxygen is not as abundant in water as it is in air. Air can be regarded as having a constant percentage (approximately 20.9%) of oxygen. Wherever air is exposed to water, the oxygen in the air will dissolve in the water. The amount of oxygen that dissolves in the water depends on many factors: whether there is adequate time and adequate mixing to fully saturate the water, the water temperature, the air pressure, etc. Water that contains the maximum amount of oxygen that can be obtained from the overlying air under the prevailing conditions is said to be saturated and the dissolved oxygen concentration is 100 %Saturation. The concentration of oxygen dissolved in water can be expressed as mg/L or as percentage of air saturation value. Water temperature, atmospheric pressure and contents of salts dissolved in water have to be taken into account when the values in mg/L are converted to %Saturation or vice versa.

There are two main sources of dissolved oxygen in an aquarium: Oxygen diffuses into the water from the air especially when the surface is agitated and also from the photosynthesis of aquatic plants. On the other hand, oxygen is removed by the aerobic degradation of organic substances by bacteria and by the respiration of all the organisms present in the water. Most indoor aquarium systems lack sufficient photosynthesis. Therefore, mechanical means of aeration is the only alternative for supplying oxygen to aquatic animals maintained in these systems. Providing some form of aeration or surface agitation to aquarium water will allow more water to contact air at the surface, increasing dissolved oxygen levels and maintaining oxygen at safe levels.



Different fish species have different requirements for the concentration of oxygen dissolved in water. The oxygen requirements of fish also depend on a number of other factors, including the temperature, pH, and CO<sub>2</sub> level of the water, and the metabolic rate of the fish. The major criteria for the oxygen requirement of fish include temperature, and the average individual weight and the total weight of fish per unit volume of water. Oxygen requirements increase at a higher temperature; a higher total weight of fish per unit volume of water can lead to increased activity and thus increased respiration as a result of overcrowding. Oxygen deficiency causes asphyxiation and fish will die, depending on the oxygen requirements of the species and to a lesser extent on their rate of adaptation.

In general, it is recommended that the dissolved oxygen concentration be kept near to saturation. Typical values in a healthy aquarium system should be 8 mg/L (85–95 % Saturation at 24°C). If the level declines below 3 mg/L they begin to show signs of suffocation. In aquariums equipped with proper filtration and aeration, insufficient dissolved oxygen is seldom a problem. It is therefore generally unnecessary to test oxygen in aquariums. However, corrective measures need to be initiated if conditions become unfavourable. Remedial action is to aerate the water. Aeration can be with air or oxygen pumps, by agitating the water surface, or by increasing the input of aerated water.

Fish exposed to oxygen deficient water do not take food, collect near the water surface, gasp for air, gather at the inflow of filters where the oxygen levels are higher, become sluggish, fail to react to irritation, lose their ability to escape capture and ultimately die. The major pathologico-anatomic changes include a very pale skin colour, congestion of the cyanotic blood in the gills, adherence of the gill lamellae, and small haemorrhages in the front of the ocular cavity and in the skin of the gill covers. In the majority of fishes the mouth gapes spasmodically and the operculum over the gills remains loosely open. The only way to know for sure if low oxygen levels have caused fish deaths in an aquarium is to measure the oxygen depletion as a probable cause of a fish death include:

- All fish die at approximately the same time (often during the night or in the pre-dawn hours).
- Large fish may be affected more than small fish.
- Moribund fish may be seen at the surface "gasping" for oxygen.
- Some species may die with their back arched, gills flared, and mouth open.

Dissolved oxygen can be measured with an electronic metering device or with a chemical titration test. Dissolved oxygen meters can be expensive, so most aquarists will generally use the chemical titration method. Most aquarium test kits do not meet the requirements for precision and

accuracy needed for professional quality data. However, most are reliable if used correctly and can provide good results for aquarium use. Specific instructions on how to use kits are provided with the kits and will vary according to the manufacturer. Commercial test kits are based on the "Azide-Winkler" titration method. It is the most reliable method, against which the others are compared to test for accuracy. It's important to become familiar with water testing and know how to use the associated test kits.

Dissolved oxygen concentrations are commonly reported as milligrams per litre (mg/L) or as percentage saturation (% Saturation). They measure the same thing, but sometimes your test kit will use only one of the measurements. Most oxygen meters can read oxygen concentrations as both mg/ L (ppm) and %Saturation. If the water temperature, salinity and barometric pressure are known, either of these can be calculated from the other. There is confusion in the literature regarding which is the better measure to employ in testing aquarium water. In practice both measures are needed to fully interpret data, but %Saturation is the most readily interpreted and ecologically relevant of the two.

Percent Saturation (%Saturation) is the amount of dissolved oxygen in the water compared to the maximum amount that could be present at the same temperature. As temperature increases, the concentration at 100% saturation decreases. These factors affect the percent saturation (the highest dissolved oxygen level possible even in well-aerated water). Living organisms require specific minimum levels of dissolved oxygen to survive. Saturation values less than 60% or over 125% are undesirable. Dissolved oxygen percent saturation values in the range of 80–120% are desirable.

It is the saturation level that directly indicates how much oxygen is available for aquatic organisms to breathe, not the amount that is dissolved in the water. As a rule of thumb, a fish in water that is 100% saturated with oxygen is able to gain access to an amount of oxygen equivalent to that in the overlying air. If the concentration falls to 50 %Saturation then it can only obtain half the amount of oxygen that is present in the overlying air. The mg/L concentrations of dissolved oxygen required to achieve these saturation levels vary enormously, particularly with temperature, so results expressed in terms of mg/L are much more difficult to interpret. At saturation the partial pressure in the water is equal to that in the thin layer of moisture-saturated air at the surface layer.

Well-aerated water (in free interchange with the air) will usually be 100% saturated. In general, the colder the water the more oxygen it can dissolve, the more saline the water the less oxygen it can dissolve, and the lower the atmospheric pressure (e.g., the higher the elevation), the less oxygen it can dissolve. These generalities come from the gas laws of physics. Oxygen saturation is calculated as the percent of dissolved oxygen relative to a theoretical maximum concentration given the temperature, pressure, and salinity of the water. It is possible to get more than 100% saturation. The water can be supersaturated in an area where there are a lot of plants or algae on a sunny day (due to photosynthetic activity).



Calculating oxygen levels in an aquarium can be somewhat complicated. Water saturated with oxygen at 15°C contains about 9.8 mg/L, whereas water at 30°C is saturated at about 7.6 mg/L. A reading of 1 mg/L @ 30°C (13.15 %Saturation) is a higher concentration than 1 mg/L @ 15°C (10.2 % Saturation) and represents more available oxygen. The figures below gives the amount of dissolved oxygen in mg/L (ppm) that represent 100 %Saturation in freshwater at normal pressure and different temperatures. To calculate the percentage of dissolved oxygen, test the amount of oxygen present in your tank (in mg/L or ppm), and divide the number by the mg/L value below that correspond to your tanks temperature. The answer is the percentage of dissolved oxygen in your system. Answers over 100% are perfectly valid, and indicate supersaturation of the water. So if you have a temperature of 25°C and you measure an oxygen level of 5 mg/ L (ppm) you divide the ppm (5) by 8.3 = 60 %Saturation.

#### Freshwater:

(a) 20°C normal pressure 9.1 mg/L = 100 %Saturation (a) 22°C normal pressure 8.7 mg/L = 100 %Saturation (a) 24°C normal pressure 8.4 mg/L = 100 %Saturation (a) 25°C normal pressure 8.3 mg/L = 100 %Saturation (a) 28°C normal pressure 7.8 mg/L = 100 %Saturation (a) 30°C normal pressure 7.6 mg/L = 100 %Saturation

Dissolved oxygen levels change according to the time of day, the temperature and the weather. Levels are usually lowest in the morning and highest in late afternoon. The ability of oxygen to remain in the solution decreases as water temperature increases (e.g., dissolved oxygen saturation decreases by about 2% for each 1°C increase in temperature). Temperature also increases the metabolic rate of aquatic animals, resulting in increased consumption of oxygen. As a result water temperature has a significant influence on dissolved oxygen levels. Therefore, dissolved oxygen in an aquarium must be maintained above levels considered stressful to the fish. Prolonged exposure to low oxygen may cause a slowing in growth rates, reproductive difficulties, stress, susceptibility to disease, and in severe cases of depletion, premature death. Usually larger fish are more affected by low dissolved oxygen levels than smaller fish.

Problems caused by too much oxygen dissolved in water are seldom encountered. However, it may happen, for example, when fish are transported in polythene bags with an oxygenfilled air space. The critical oxygen level of water is 250 to 300% of the air saturation value; fish may be injured at these higher values. The gills of such affected fish have a conspicuous light red colour and the ends of the gill lamellae fray. When such fish are placed in the aquarium water they may suffer from secondary fungus infections and some of them may die. It is possible that fish adapted to such high oxygen levels need to be progressively acclimatised to more normal concentrations. This condition should not be confused with the supersaturation of water with dissolved gas, which can cause gas bubble disease.

Supersaturation with dissolved gas occurs when the pressure of the dissolved gas exceeds the atmospheric pressure. It occurs when water is equilibrated with air under

pressure, e.g., at the bottom of a lake or reservoir, in ground water, or if air is drawn into a centrifugal water pump. It can also occur if cold air-equilibrated water is warmed up without re-equilibration to the higher temperature. A bottle containing such water will show either minute bubbles forming as a cloudy suspension which will clear from the bottom upwards, or larger bubbles forming on the glass wall. This is analogous to that seen in an opened bottle of carbonated drinking water.

If fish are exposed (at a lower atmospheric pressure) to such water, their blood equilibrates with the excess pressure in the water. Bubbles form in the blood and these can block the capillaries; in sub-acute cases the dorsal and caudal fin can be affected, and bubbles may be visible between the fin rays. The epidermal tissue distal to the occlusions then becomes necrotic and cases are known where the fins have become completely eroded. In severe cases, death occurs rapidly as a result of blockage of the major arteries, and large bubbles are clearly seen between the rays of all the fins. A similar effect of gas bubbles forming in the blood can be experienced by deep-sea divers when they return to the surface. The remedy is either to remove the fish to normally equilibrated water or to provide vigorous aeration to strip out the excess gas.

There is little information available on how rainbowfishes are affected by low levels of oxygen. The minimum dissolved oxygen level that rainbowfishes can safely tolerate depends upon individual species and temperature. Different life stages (i.e., eggs, larvae, juveniles and adults) may also have different oxygen needs. Several experiments have been conducted on the effects of low oxygen on rainbowfishes and other freshwater species. One such study (Flint 2003) reported that Melanotaenia splendida and M. utcheensis died when dissolved oxygen saturation reached 7%. Lower egg production was also noted over the duration of the study in tanks with lower oxygen levels. However, the eggs of M. utcheensis and M. splendida were found to be remarkably tolerant to low dissolved oxygen. Eggs were able to survive and produce viable larvae at dissolved oxygen saturations levels lower than those that killed their parents (lowest tested was 5% at 28°C).

In another study (Pearson *et al.* 2003), experiments were conducted to identify acute threshold values for *M. splendida* exposed to various low dissolved oxygen levels. Fish exposed to oxygen levels of 25–35 %Saturation or higher for 5 days experienced negligible mortality. In contrast, all fish exposed to <10 %Saturation died within 24 hours after the start of the experiment.

Five-day exposure to dissolved oxygen levels of 25-35 % Saturation did not affect the survival, and did not appear to affect the feeding behaviour, of adult *M. splendida*, However, the fishes appeared to be more lethargic following exposure to 25-35 %Saturation than individuals exposed to higher oxygen treatments. Relative to normoxia (approximately 100 % Saturation), breathing rates of *M. splendida* had doubled by the time oxygen levels had declined to 55 %Saturation, nearly tripled by 40 %Saturation and quadrupled at approximately 35 % Saturation.

Furthermore, a 24-hour experiment on survival of M. *splendida* by Pearson et al. (2003) revealed a fine line between survival and mortality. The data suggested that M. *splendida* exposed to lowered dissolved oxygen levels for 24 hours were able to survive exposure to oxygen levels down to 13 %Saturation. Levels of approximately 12 %Saturation were lethal to 90% of fish, while exposure to oxygen levels of 9 %Saturation was lethal to all the rainbowfishes. However, it was apparent during the experiment that the rainbowfishes were making use of dissolved oxygen gradients in the test tanks that were unable to be measured. Thus, the recorded oxygen concentration that the rainbowfishes were actually exposed to.

Rainbowfishes can perform aquatic surface respiration and access higher oxygenated water at the air-water interface to survive lower oxygen concentrations for short periods of time. They stay just below the surface, put their snout at the air-water interface, and breathe in the film of water that is in direct contact with the air. This thin layer of water is comparatively rich in oxygen. Typically, rainbowfishes will wait until a very low threshold of oxygen concentration is before starting surface respiration. This reluctance to breathe near the surface is easy to understand when we consider that, in nature, many predators of rainbowfishes are terrestrial or aerial animals that attack from above, and therefore being close to the surface has some risks.

## Water Hardness

Water hardness is commonly confused with alkalinity. The confusion relates to the term used to report both measures, milligrams per litre as calcium carbonate equivalent (mg/L CaCO<sub>3</sub>). Calcium carbonate is a general term that indicates the total quantity of divalent salts present and does not specifically identify whether calcium, magnesium and/or some other divalent salt is causing water hardness.

Calcium (Ca<sup>2+</sup>) and magnesium (Mg<sup>2+</sup>) ions are the most common factor that comprises hardness and aquarium test kits usually determine both ions as "total hardness". Despite the much confused state of misinformation prevalent in the hobby, permanent or general hardness is true hardness; carbonate hardness (°dKh) is not hardness at all but alkalinity. Water hardness should be expressed as a concentration of divalent ions in mg/L (ppm). The terms "permanent", "general", and "carbonate" hardness should be discontinued.

Calcium and Magnesium are essential in the biological processes of fish (bone and scale formation, blood clotting, and other metabolic reactions). Fish can absorb calcium and magnesium directly from the water or from food. However, calcium is the most important environmental divalent salt in rainbowfish culture. The presence of calcium in aquarium water helps reduce the loss of other salts: for example, sodium and potassium from the fish's blood. Sodium and potassium are the most important salts in fish blood and are critical for normal heart, nerve, and muscle function. Research has shown that environmental calcium is also required to reabsorb these lost salts. In low calcium water, fish can lose substantial quantities of sodium and potassium into the water. A recommended range for free calcium in aquarium water is 25 to 100 mg/L (50 to 250 mg/L CaCO<sub>3</sub> hardness). A low CaCO<sub>3</sub> hardness value is a reliable indication that the calcium concentration is low. However, high hardness does not necessarily reflect a high calcium concentration. It is not clear however, whether calcium plays an equally important role in embryonic development.

Water hardening (swelling process of flaccid newly shed eggs when they first contact water and absorb water) of fertilised eggs varies according to the water hardness. When hardness is low, increase in egg diameter is greater. However, the effect of water hardness on larvae hatching and growth of rainbowfishes is unknown. For those species, which are found in hard water in their natural environment, hard water may be required for good development, while in others it might ameliorate the deleterious effect of non-optimal conditions.

Water hardness is expressed in a confusing array of scales, although in the aquarium hobby the influence is to express them in terms of milligrams per litre of calcium carbonate (mg/L CaCO<sub>3</sub>), which is also equivalent to parts per million, or in the obsolete German units of "degrees of hardness" (° dH). °dH or Deutsche Hartgrad, loosely translated, means German hardness gradient. As a result of the different systems, it's not uncommon to see water hardness incorrectly reported as dH = 85 ppm (an erroneous combination of both systems).

For aquarium purposes, you can use the following conversion factors:

°dH X 17.9 = ppm (mg/L) ppm x 0.056 = °dH

## **Dissolved Salts**

Confusion can arise from the fact that salinity is sometimes equated to one of the terms 'total dissolved salts' or 'total dissolved solids' – both are sometimes abbreviated TDS.

*Total Dissolved Salts* is determined by calculation from the results of analysis for common ions (sodium, calcium, chloride).

*Total Dissolved Solids* is determined by filtering a sample, drying at a specified temperature, and weighing the residue. It includes non-ionised substances if present (example: sugars, other organics, colloidal particles too small to be retained by filter medium), so it can be greater than Total Dissolved Salts.

TDS should properly be interpreted as Total Dissolved Salts, because non-ionised species do not contribute to electrical conductivity of the sample.

Total Dissolved Salts (TDS) is a measurement of the total amount of dissolved salts in water and expressed in ppm of NaCl. TDS is essentially the same as conductivity.



For aquarium applications it is recommended that the conversion formula TDS (in ppm) x 0.68 = EC (in  $\mu$ S/cm) be used. The conversion formula is only an approximation. TDS meters are calibrated with a Sodium chloride solution.

Conductivity or specific conductance is the measure of the water's ability to conduct an electric current. Electrical conductivity is an inherent property of most materials, and ranges from extremely conductive materials like metals to very non-conductive materials like plastics or glass. About halfway between the two extremes in conductivity are aqueous solutions, such as aquarium water. In metals, the electrical current is carried by electrons, while in water it is carried by charged ions. In both cases, the conductivity is determined by the number of charge carriers, how fast they move, and how much charge each one carries.

Thus, for most aquarium waters, the higher the concentration of dissolved salts, which will lead to more ions, the higher the conductivity. This effect continues until the solution gets "too crowded", restricting the freedom of the ions to move, and the conductivity may actually decrease with increasing concentration. This can result in two different concentrations of a salt having the same conductivity.

The specific conductance is measured by passing a current between two electrodes (one centimetre apart) that are place into a sample of water. The basic unit of conductance is the Siemens (S) and was formerly called the mho. The normal unit of measurement for conductivity is expressed in microsiemens per centimetre ( $\mu$ S/cm) or millsiemens per centimetre (mS/cm). Conductivity meters are calibrated with a Potassium chloride solution.

In natural waters the distributions of freshwater macrophytes, invertebrates and fish are not noticeably affected by variations in conductivity if concentrations remain below 1000  $\mu$ S/cm (roughly equivalent to total dissolved salt concentration of 500 mg/L). Monitoring of intermittent inland streams has shown that conductivity levels fall dramatically during storm events, and rise gradually during periods of base flow, achieving maximum values when flows stop.

## pH of Water

The term *p*H derives from a combination of *p* for the word power and H for the symbol of the element Hydrogen. Together the meaning is the power or exponent of hydrogen. The *p*H value can be defined as 'a number used to express the concentration of ionised hydrogen in an aqueous fluid and is thus indicative of the reaction of that fluid, that is, the neutrality or the degree of acidity or alkalinity'. According to the theory of electrolytic dissociation all liquids of which water is a constituent contain free, positively charged hydrogen (H<sup>+</sup>) ions and negatively charged hydroxyl (OH<sup>-</sup>) ions.

Much mystery has been made about the pH value of aquarium water, but, in reality, there is nothing very mysterious about it, nor is the subject so complicated, as some would have us believe.

*p*H serves as a convenient way to compare the relative acidity or alkalinity of a solution at a given temperature. Absolute neutrality has a *p*H value of 7.07 (usually taken as 7.0). The addition of acid increases the hydrogen (H) ion concentration; consequently the *p*H of all acid solutions is less than 7.07. The addition of alkali increases the concentration of the hydroxide (OH<sup>-</sup>) ions, and decreases that of the H<sup>+</sup> ions, so that the *p*H of all alkaline solutions is greater than 7.07. The range of *p*H values extends about equally on each side of 7.07; for the complete range of *p*H values forms a graduated scale from about -0.3 to 14.5.

Since the pH of water is critical to the survival of most aquatic animals and plants, monitoring pH values in the aquarium is an important part of successfully maintaining rainbowfishes in captivity. The testing is quick and easy and can establish a valuable baseline of information so that unanticipated water quality changes can be better understood.

# Practical Application of pH

pH is one of the most common aquarium measurements because many chemical processes are dependent on the pH. Aquarium conditions can often be significantly altered by changing the pH of the water. The solubility of many chemicals in solution, and their bio-availability is dependent on pH. The physiological chemistry of living organisms usually has very specific pH boundaries.

A simple test kit which exhibit characteristic colour changes at different pH values or a hand-held electronic meter can be used to test pH. Always remove a sample of water from the aquarium to measure the pH with an electronic meter. Measurement by immersing the electrode directly in the aquarium can be severely compromised by other undetectable electrical currents from power filters, heaters, etc. pHelectrodes which are not routinely cleaned and standardised will not provide accurate readings and will be no better than, and often far worse than, a colorimetric measurement made with the cheapest liquid-reagent test kit.

pH electrodes must be routinely checked against known pH standards to insure accuracy and need to be replaced every 9 to 12 months. The popular pocket pH "pens" are disposable meter/electrode combinations which can be inaccurate, particularly if not calibrated correctly, and do not compensate for changes in temperature. Therefore, the selection of measuring devices for pH is largely a situation in which "you get what you pay for". If you are unable to recognise the inadequacies of pH meter measurements, you are better off using dye methods. Only dyes with clear-cut colour changes around the target pH should be used.

The commonly used pH indicators for freshwater testing are bromothymol blue (yellow to green to blue as the pH increases) and phenol red (yellow to orange to red with increasing pH). The colorimetric method is the least expensive but can suffer from interferences due to discoloured water samples, salinity, organic matter, and substances that can oxidise or reduce the reagents. In water with very low alkalinity, the indicators themselves may actually alter the pH



of the sample. However, for the purposes of routine aquarium testing, colorimetric indicators are more than adequate. Some scientific supply houses now sell narrow-range litmus paper, which allows for low-cost, rapid estimation of pH.

In well-buffered aquariums with alkalinity levels above 50 mg/L, the pH will be more stable. In the morning, carbon dioxide levels are high and pH is low because of respiration during the night (carbon dioxide forms a mild acid when dissolved in water). When a suitable light source is provided, algae and other aquatic plants will produce carbohydrates and oxygen from carbon dioxide and water by photosynthesis. As carbon dioxide is removed from the water, its pHincreases. In aquarium systems, the pH will generally drop in relation to the fish load, biological filtration, feeding, and maintenance schedules. Therefore, acidic water in an aquarium system is biologically different from that found in nature. In an aquarium, acids derive primarily from two sources. The first is when carbon dioxide (directly dissolved into water or released as a respiration by-product) mixes with water to form carbonic acid.

$$H_2O + CO_2 \iff H_2CO_3 \iff H^+ + HCO_3^-$$

water + carbon dioxide <=> carbonic acid <=> hydrogen ion + bicarbonate

The other is when ammonia undergoes nitrification by bacteria.

$$2 \text{ NH}_3 + 3 \text{ O}_2^- > 2 \text{ NO}_2^- + 2 \text{ H}^+ + 2 \text{ H}_2\text{O}$$

ammonia + oxygen » nitrite + hydrogen ion + water

If the aquarium water is not well buffered any acid that is added serves to drive down the pH. Consequently, the daily pH swings caused by photosynthesis can combine with longer-term acid accumulations and cause the pH to suddenly drop with catastrophic results for the fish. There are indirect consequences that can also affect fish. Changes in *p*H will affect the toxicity of many dissolved compounds. For example, ammonia becomes more toxic as pH increases. Nitrifying bacteria, essential in the conversion of ammonia to nitrate also have a pH range preference, which is between 7.5 and 8.6. Variations in pH will also have an effect on some disease treatments. Fluctuations in pH, even though they may still be within the preferred range, can be stressful and damaging to fish health. Therefore it is important to monitor pH. The actual time to measure pH will depend on what you hope to achieve with your tests.

It is well-established that levels of pH fluctuate throughout the day, and a single pH measure taken during the day may not draw a very accurate picture of long-term pH conditions in the aquarium. Photosynthesis by aquatic plants removes carbon dioxide from the water; this can significantly increases pH. A pH reading taken at dawn in an aquarium with many aquatic plants will be different from a reading taken six hours later when the plants are photosynthesising. Likewise, in waters with plant life (including planktonic algae), an increase in pH can be expected during the growing season. For these reasons, it is important to monitor pH values at the same time of day if you wish to compare your data with previous readings. It is also important to monitor pH values over a long period of time to provide useful data.

Most rainbowfish species can survive pH changes down to 4.0–5.0 or up to 9.0-10.0, but exposure to more acidic or alkaline waters can be lethal within a few hours. There is no definite pH range for maintaining rainbowfishes in captivity, but a gradual deterioration of their health is likely as the pH values are removed from their preferred range. Water with a pH range of 6.5–7.8 is usually considered best for rainbowfishes in captivity. However, studies regarding the survival, growth and reproduction of rainbowfishes in acidic or alkaline water conditions are still inadequate.

Rainbowfishes will survive reasonably well in waters with a pH range of 6.5 to 8.3. If pH readings are outside this range, growth is reduced; their slime coat can suffer, making them susceptible to disease. At values below 5.0 or above 9.0, mortality, impaired growth and reproduction can be expected. The gas exchange in the gill membranes will be so reduced that the fish may suffocate. From my own experience, most rainbowfishes in captivity do not seem to be comfortably in water below pH 6, certainly not for any extended period.

There may be some isolated populations that have adapted to extreme conditions as low as pH 4.0. However, acidification of water is thought to have a major impact on fish mortality and the structure of their populations. Field and laboratory studies have shown a clear correlation between low pH and declining fish populations. The recruitment failure by embryo and larva mortalities is considered as a primary factor leading to gradual loss of fish stocks of aquarium as well as of wild populations. In addition, exposure of larvae at pH 4.5 or lower may impair growth and reduced survival. Behavioural responses, such as reduced swimming and feeding activities, have been observed in fish larvae exposed to acidic water conditions.

Experimental studies with eggs from different fish species showed that the sensitivity of embryos to low water pH is related to the developmental stage. Highest mortality occurs immediately after fertilisation of the eggs ('green' egg stage) and at the time of hatching. Freshly fertilised eggs, which show no external signs of cleavage, are called 'green'. When eye pigmentation and further development are visible through the chorion, eggs are 'eyed' and these are less vulnerable to low pH. Most studies on egg development and effects of low pH indicate a delay of the hatching time and an elongation of the hatching period. Fertilised embryos may also develop deformities when exposed to pH 4.5 or lower.

Numerous laboratory studies have tested tolerance of fish species to low pH. In most fish species, the 96-hour LC<sub>50</sub> was reported to range from pH 4.0 to 5.0 for early and adult life stages. Fish mortality caused by low pH has been thought to be associated with disturbance of water and ion balance, which may eventually lead to disruption of ion homeostasis. The chloride cells in the gills, opercular epithelium, and skin of fishes are known to play a key role in regulation of ionic balance.



Although the effects of low pH on fish have been extensively studied, relatively little is known about the effects of high pH. Environmental high pH values can be caused by enhanced photosynthetic activity of aquatic plants, and can be accompanied by high temperatures and supersaturating of dissolved gases. The latter too may contribute to fish mortality, which makes it difficult to relate observed effects to the high pH value alone. From field studies it appeared that mortality of adult fish is more pronounced after episodic pH changes, e.g., after heavy rainfall.

Acidification of water also decreases photosynthetic activity in aquatic plants and phytoplankton. Studies (Allen 1995) show that a pH of less than 4.0 is directly toxic to the roots of aquatic plants. Some species of insects have been noted to avoid depositing eggs, thereby reducing an important food source for other species. Many freshwater invertebrates fail to reproduce in acidified waters. Some species will avoid entering acidified waters if they have an alternative.

#### pH in Natural Waters

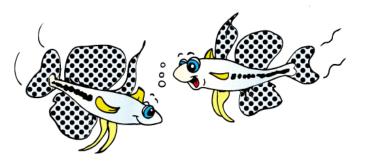
In Australia the *p*H of freshwater streams naturally varies between catchments due primarily to differences in catchment geology and vegetation. However, in general, most freshwater streams have a *p*H range of 6.5 to 8.0. There are, of course, exceptions to this general rule. Coastal streams generally range from about *p*H 4.5 in tannin-stained streams associated with coastal 'wallum' heathlands, to *p*H 8.8 in streams at the headwaters of some catchments. Forested areas and coastal areas with high rainfall generally have the lowest *p*H.

Naturally low pH also occurs due to seasonal wetting and drying of peaty soils in wetlands and waterways. In contrast, waterways in the Pilbara and Kimberley areas of Western Australia generally have a pH range of 8.0–8.5. The pHrecorded from a number of coastal streams in Queensland including several relatively undisturbed rivers i.e., the Endeavour and Daintree range from pH 6.5 to 7.15. Other river recordings include the Russell-Mulgrave, Tully, Herbert, Ross, O'Connell and Burrum, all of which have relatively intensive development and floodplain modification. The pH range for lowland and upland streams was 6.5-8.0 and 6.5-7.5 respectively. In areas with highly alkaline subsoils natural pH is generally in the range of 7.0–8.5. Similar conditions are found throughout northern topical Australia. Data for streams in New Guinea are generally not available.

Large stretches of dune field and coastal heathland (wallum) swamps and streams are found dotted along the eastern Australian coast. These 'blackwater' habitats are generally acidic, with pH levels from 3.9 to 6.8, have low conductivity, and vary in their dissolved organic matter, ionic composition, and colour. Alkalinity and hardness levels are very low. Factors contributing to these variations are age, formation, layers of low permeability and peats, proximity to the sea, surrounding vegetation, and the extent to which leaf

litter accumulates and decays in the water. These water bodies are usually well oxygenated but highly oligotrophic (low nutrient levels due to the surrounding infertile soils) and of low biological productivity. The dominance of humic acids among this organic material and the relatively low *p*H are not conducive to bacterial degradation, so particulate and dissolved humic compounds are metabolised very slowly. The brown (tannin) colour of the water severely limits penetration of light, which, together with low concentrations of inorganic ions, restricts photosynthetic activity in aquatic plants. Limited photosynthesis and slow bacterial degradation results in low zooplankton and phytoplankton development.

Rainbowfishes often found in these habitats include *Iriatherina* werneri, Melanotaenia maccullochi, Pseudomugil gertrudae, Pseudomugil mellis and Rhadinocentrus ornatus.



#### Summary

There are numerous water chemistry parameters in the aquarium that need to be monitored. Those primarily concerned with keeping rainbowfishes are dissolved oxygen, temperature, pH, alkalinity and carbon dioxide. However, many dissolved substances, which cannot be measured by the average hobbyist, rapidly accumulate in an aquarium especially if the water is not changed on a regular schedule. These dissolved substances remain largely unidentified but include organic acids, sugars, microbes, phenolics, proteins, hormones and fine particles of detritus. The toxicity of these dissolved substances to rainbowfishes is not completely known; however research has indicated that certain components will inhibit the growth and development of rainbowfishes and increases their susceptibility to disease. Some researchers believe that there is a direct relationship between high levels of these substances and high populations of disease organisms in aquaria.

A variable proportion of these accumulated substances is readily degradable and consumes significant amounts of oxygen, increasing carbon dioxide levels, lowering the pH, and contributing towards the deterioration of water quality. Therefore the reduction of these compounds ultimately leads to improved water quality and healthier specimens. Only regular waterchanges will lower the concentration of these substances and restore a stable environment. The more frequently the water is changed, the lower the stresses on the system and its inhabitants. Most rainbowfishes enjoy waterchanges; they seem stimulated by them, so long as they do not involve so great a change in water parameters that they induce stress.



## Stocking Density

The stocking density, that is the volume of water available to a single fish, can be a significant health factor. Unfortunately, people who commence keeping aquaria are usually too anxious to have as many fish as possible, and most of the problems which overtake their endeavour arise from such over-stocking. Large numbers of fish confined in the relatively small space of the aquarium can often lead to impaired growth and health conditions, and before long it ends in death. This is due to reduced oxygen levels and increased toxic substances such as ammonia and nitrite. What follows is that many enthusiastic beginners become so thoroughly depressed by their first mistake that they never try again! There is one sure guidance to a beginner in these matters - have too few animals rather than too many. Most important of all, do not add too many fish to the aquarium at the same time.

Although there are numerous mathematical formulas to calculate the fish holding capacities of aquariums, they are essentially without merit. The maintenance of water quality is the best method to determine the true carrying capacity of the aquarium. Start with a very small number of fish and check the water quality using test kits for ammonia, nitrite, and pH. More fish can be added gradually, building up over the months as your aquarium matures, as long as the water quality is not diminished. Ultimately, your own observations and test results should help you determine what is a safe number of fish for your particular situation.

The hobbyist who undertakes a regular program of water changes and maintenance will be able to successfully keep and maintain larger aquarium populations than those who are unable or unwilling, to perform such activities. The more fish you have, the more water changes will be needed. Each hobbyist must determine for themselves the level of effort they are willing to spend on their fish tanks and adjust the fish population accordingly. This will result in both healthier fish and greater enjoyment for the hobbyist.

## **Aquarium Filtration**

An effective filtration system needs to remove waste solids, oxidise ammonia and nitrite, remove carbon dioxide, and aerate the water before returning it to the aquarium. Waste solids are generally removed via some form of mechanical filtration, ammonia and nitrite via biological filtration, and carbon dioxide by the provision of an air/water interface. Aeration of the water is also achieved across the same air/water interface. In the average aquarium, all these processes are done within the tank. In more specialised aquarium systems, most if not all of the processes are undertaken external to the aquarium.

Proper biofilter design is critical for the success of aquarium filtration systems. A number of biofilter types have been developed and tested in aquacultural systems, but very little research has been conducted with aquarium filters. The ability of a filter to remove harmful nitrogenous compounds is affected by media type, flow rate/retention time, dietary protein source/content, water quality, and filter design. Media type and quantity determine the amount of surface area available for bacterial growth. Dietary protein source and content directly affect the amount of nitrogen that will enter the water. Interactions between these factors and with the biology and chemistry of the system must be considered. Filter media should provide maximum surface area with media particles and pore spaces large enough to minimise clogging. Retention time, which affects filter efficiency but not necessarily nitrification rates, is one of the most significant factors that must be considered in filter design and operation. Ammonia removal efficiency is most affected by retention time in the filter, hydraulic and ammonia loading, and amount of organics in the water.

Flow rate significantly affects the rate of ammonia removal. More ammonia was removed in filters with the flow rates of 57.5 L/hour than for 111 L/hour. The slower flow rate provided a longer residence time and allowed time for bacterial reactions to take place. Filter flow rate does not significantly affect nitrite levels, nitrate levels, fish growth rate or fish mortality. Dietary protein content will significantly affected nitrate level because of the varying nitrogen contents of the food rations, but will not significantly affect nitrite level or fish mortality (Brunty *et al.*, 2005).

Maintaining healthy fish in an aquarium involves establishing adequate dissolved oxygen levels, removal of wastes, and sufficient ammonia nitrification. Aquarium fish produce a variety of wastes including faecal solids, ammonia, carbon dioxide and other materials e.g., uneaten food and dissolved substances that will accumulate in an aquarium. These wastes must be removed from the aquarium water or they become toxic to the fish. These accumulated wastes act as a nutrient source for bacteria that generate nitrogenous wastes, increasing the demand for oxygen, increasing carbon dioxide levels, lowering the pH, and contributing towards the deterioration of water quality.

While poor water quality may not be lethal, little or no growth as well as increased incidence of disease can result from poor water quality. Some researchers believe that there is a direct relationship between high levels of wastes and dense populations of disease organisms in aquaria, thus increasing the susceptibility of the fish to disease. Maintaining good water quality is of primary importance. Therefore, effective filtration or bioconversion must be provided to eliminate the effects that these waste products have on the rainbowfishes health and survival.

A properly designed filtration system will remove or reduce such wastes, and enable you to maintain your rainbowfishes for extended periods without drastic procedures, such as extensive water changes. All systems should have a method of removing particulate waste, a method to re-oxygenate the water and a method to recirculate the water. Each component of the filtration system must work in conjunction with other components of the total system. In choosing any filter component, you should keep in mind that it must be capable of maintaining an excellent environment for the fish. However, don't expect any filtration system to provide perfect water conditions. Over a period of time the water quality will deteriorate and therefore must be changed on a regular basis and is a necessary part of your aquarium management program.



Aquarium filtration can generally be accomplished by a number of methods:

- Mechanical removal of undissolved, particulate matter.
- Chemical removal of dissolved organic matter.
- Biological conversion of toxic wastes to less toxic materials.
- Any combination of the above.

Removal of particulate matter can be accomplished by mechanical filtration through porous material such as sponge, screen, sand or gravel. Particles that are larger than the pore sizes in the filter media can clog the filter, and lead to reduced filtering capacity and efficiency. Most of the particulate are made up of organic compounds that will gradually break down in the system from bacterial activity. Although this process adds additional oxygen demand to the system, it reduces the need for frequent cleaning if the solids do not become resuspended or interfere with normal water flow. Mechanical filters require regular cleaning since they are prone to clogging.

Filtration systems should be designed for simplicity of operation. Sufficient time must be allowed for conditioning of the biofilter prior to introducing fish. Ammonia and nitrite concentrations must be checked frequently. Dissolved oxygen should be sustained above 5.0 mg/L and periodically verified. Alkalinity, hardness, and pH need to be measured and adjusted, if necessary, at regular intervals. Filters should be inspected and cleaned as required. Medications used to treat fish diseases may be toxic to bacteria in the biofilter. An ability to isolate fish tanks for disease treatment should therefore be provided.

# **Biological Filtration**

The ability to maintain rainbowfishes in good health is a function of the efficiency and health of the aquarium's biological cycle and the aquarist's management of the system. Providing and maintaining a suitable biological filtration system, together with regular water changes, will facilitate the removal and detoxification of dissolved wastes. These dissolved compounds remain largely unidentified but include organic acids, phenolics, proteins, hormones, and other compounds. The toxicity of these dissolved wastes to fish is not completely known, however research has indicated that certain components will inhibit the growth and development of fish. As rainbowfishes breathe and metabolise feed, wastes are released into the water column. If these wastes are allowed to accumulate they will increasingly degrade the water quality.

Aquarium systems use biofilters and a continuous flow of recirculated water to bring oxygen to the fish and detoxify nitrogenous wastes. Nitrogenous wastes, particularly ammonia, can rapidly accumulate to dangerous levels unless biological filtration is properly employed. Biological filtration uses naturally occurring bacteria to detoxify nitrogenous wastes and, provided it works efficiently, it does so to the extent that nitrogenous wastes are virtually undetectable. Thus, the primary purpose of biological filtration is to remove nitrogenous wastes and is therefore, a critical component in every aquarium. There are a number of technologies available to remove nitrogenous wastes, but the most commonly used method is biological filtration.

The generation of nitrogenous wastes in aquarium systems occurs with the breakdown of proteins from excess feed, excretion from the gills of fish as they utilise feed, and decomposition of organic waste by bacteria. Fish may excrete nitrogen in the form of ammonia, urea, uric acid, amines and amino acids. Nevertheless, fish and other aquatic organisms, particularly those in freshwater, release their nitrogenous wastes primarily as ammonia (NH<sub>3</sub>) excreted across the gill membranes. Ammonia is also released through decomposition of dead animals and plants, uneaten food, bacteria and other organic matter.

Ammonia is both highly toxic and highly soluble in water. At a pH range of 6–8 approximately 90% of the total nitrogenous waste is excreted across the gills, with ammonia accounting for approximately 85% of this total. Because of its high solubility, ammonia becomes effectively diluted by the environment as soon as it is excreted and it thereby rendered harmless. However, the natural environment of rainbowfishes is a much larger aquatic system than an aquarium, and it would be difficult for ammonia to reach toxic concentrations, unless, of course, the water was polluted.

On the other hand, in an aquarium, which is far from a balanced ecosystem most rainbowfishes enjoy in nature, ammonia can build up fairly quickly, especially if overstocked and overfed. The amount of ammonia generated by fish varies with the amount of food put into the aquarium, accelerating as stocking and feeding rates increase. Generally, 1 to 3 mg of ammonia is produced for every 100 mg of feed.

An excess ammonia concentration can detrimentally effect fish growth and health, and ultimately lead to mortality. Ammonia has been reported toxic to freshwater organisms at concentrations ranging from 0.53 to 22.8 mg/L. Mortality results from gill hyperplasia, a condition which decreases gill surface area and thereby leads to inadequate transfer of toxic metabolites from the fish to the aquarium water. Although acute ammonia toxicity values vary between fish species, most aquatic organisms experience significant growth reductions, and lower resistance to disease at concentrations between 0.05–0.20 mg/L.

The LC<sub>50</sub> toxicity of ammonia for *Melanotaenia splendida* at 96 hours is 1.99 mg/L [3.49 mg/L @ 24 hours and 2.33 mg/L @ 48 hours]. Rainbowfish fry can only tolerate up to about 0.57–0.75 mg/L, before they all die. Therefore, ammonia concentrations must be consistently maintained below toxic levels. Preferably, levels should be lower than 0.02 mg/L.

In well-planted aquariums most of the ammonia is taken up directly by the plants (including algae), as it is the preferred nutrient form of nitrogen for most plant species. Existing research suggest that approximately 50% ( $\pm$ 20%) of the total ammonia load is assimilated by plants.



In the average aquarium however, most of the ammonia will be converted by nitrifying bacteria to nitrate (nitrification). If the water quality is allowed to decline or the aquarium fish population is suddenly increased, ammonia and nitrite levels can increase rapidly.

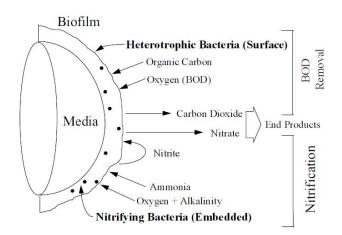
Total ammonia-nitrogen consists of ammonia (NH<sub>3</sub>) and ammonium (NH<sub>4</sub>) ions. At any given time there will be both ammonia and ammonium present. The conversion between volatile ammonia and ammonium ions strongly depends on the pH and temperature. When the *p*H of the water is acidic (<6.9) or neutral (7.0), the majority of the nitrogen is ammonium. When the *p*H increases over 8.0, the nitrogen is mostly ammonia. For a normal condition of 25°C and a *p*H of 7.0, ammonia amounts only to 0.6 % of the total ammonia-nitrogen present. At a *p*H of 9.5 and a temperature of 30°C, the percentage of total ammonia present in the ammonia form increases to 72%.

Ammonia-nitrogen is an energy source for autotrophic (nitrifying) bacteria that oxidise the ammonia-nitrogen to nitrite and nitrate. Nitrate is a stable end product with low toxicity and does not harm the fish in the concentrations typically present, but ammonia and nitrite are both highly toxic at low concentrations.

Both heterotrophic and autotrophic bacteria are common bacteria found in the aquarium. Organic waste such as fish faeces, uneaten food, dead plant tissue, and other organic material serve as the primary source of nutrition for heterotrophic bacteria, which metabolise the waste into ammonia-nitrogen (ammonification). Bioconversion of organic material by heterotrophic bacteria is a precursor to nitrification as high levels of organic material can inhibit nitrification. Therefore the removal of organic wastes from the water will enhance the efficiencies of the biological filter. To prevent excess amounts of solids from accumulating in the biofilter, particulate matter is usually removed prior to, or as the first component of biofiltration.

The autotrophic (nitrifying) bacteria are particularly important because they oxidise ammonia to nitrite and ultimately to nitrate, which is comparatively innocuous except at very high concentrations. Nitrifying bacteria must remove ammonianitrogen at a rate equal to production to maintain water quality at a level adequate to prevent exposure to the fish. The rate of reduction corresponds to the rate of growth of the nitrifying bacteria. When water quality is sufficient to meet their environmental needs and they are given enough time to reproduce, the nitrifying bacteria will flourish. Their concentration in aquarium systems therefore becomes the limiting factor of biological filtration.

Nitrifying bacteria oxidises the ammonia-nitrogen  $(NH_4 + NH_3)$  to nitrate, allowing the aquarium water to be recycled many times before a water change is required. The nitrate can be ultimately converted to nitrogen gas and oxygen by denitrifying bacteria, thus completing total ammonia conversion. However, the denitrifying process is not well known in the aquarium hobby and also nitrate is less harmful than ammonia or nitrite in a practical point of view. Nitrates are relatively non-toxic unless the levels are extremely high, and they are typically maintained at low levels by regular water changes.



Heterotrophic bacteria exist on the surface because of their higher growth rates while the slower growing nitrifiers become embedded in the biofilm. A filter with more media surface will naturally support more nitrifiers, which will convert more ammonia, as long as a sufficient water flow rate is maintained that delivers ammonia, oxygen, and alkalinity to the nitrifying bacteria (Golz 1995).

However, the biofilter is not the only place in the aquarium where nitrification takes place. Up to 30% of nitrification in a typical aquarium system takes place outside of the biofilter (e.g., in biofilms on piping, plants, substrate and tank walls). Nevertheless, the majority of biochemical reactions pertaining to heterotrophic and autotrophic bacteria occur within biofilters. Biofilters are specifically designed for concentrated bacterial attachment and nitrification via biofilms. Because of its advantages, biofilm nitrification has become the standard treatment method for recirculating aquarium systems.

Heterotrophic and autotrophic (nitrifying) bacteria compete for available surface area in biofilters. Under optimal growth conditions, heterotrophic bacteria grow very efficiently, doubling in population from about 15 minutes to 8 hours. Comparatively, nitrifying bacteria are much less efficient, and typically require 15-29 hours for ammonia oxidising bacteria and 10-21 hours for nitrite oxidising bacteria to double in population. High organic wastes can result in establishment of large heterotrophic bacteria populations on the filter media, enabling them to outcompete nitrifying bacteria for available surface area, potentially decreasing nitrification efficacy of the filter. Ammonia removal will decrease as organic loading increases. Therefore, a biofilters nitrification capacity depends first upon the amount of surface area available to the nitrifying bacteria.

The concept of biological filtration is to provide a substrate with a large surface area to encourage the growth of nitrifying bacteria. As the bacterial population develops, it coats the surface upon which it is growing. Water containing ammonia and/or nitrite flow over this media (and the bacteria attached to it). Bacteria use the ammonia as an energy source to drive their life processes. These bacterial excrete nitrite, require oxygen and produce carbon dioxide as by-products of their respiration.



A different group of bacteria use the released nitrite and convert it to nitrate. These bacteria use the nitrite to nitrate conversion for an energy source; they use nitrite and oxygen, and produce nitrate and carbon dioxide. The ammonia to nitrite conversion produces hydrogen and uses up alkalinity.

In aquarium systems, nitrification occurs at a rate of around 200 to 400 mg of ammonia per square meter of biofilter surface area per day. Thus, it is apparent that an important factor in biofilter design is to get the maximum amount of surface area into a given volume. Increasing the surface area in the filter increases the number of bacteria leading to more efficient nitrification. Therefore the nitrifying capacity of the biological filter is largely a function of the type of medium and the volume of the filter. Gravel, sand, plastic beads and rings, and plastic plates are the substrates most commonly used. Each type of biological filtration medium has a defined specific surface area per unit volume.

Predicting the performance of biofilters is an engineering challenge. The task is complicated by the wide variety of environmental conditions in the aquarium system. A myriad of biofilters designs have been generated that attempt to maximise specific surface area and oxygen transfer within the context of biological filtration. In designing biofilters, the principal concerns should be maximum surface area for bacterial growth, high dissolved oxygen levels, uniform water flow through the filter, sufficient void space to prevent clogging, and proper sizing to ensure adequate ammonia removal capability. Mechanical filtration also must be employed to ensure consistent removal of particulate matter. Particulate matter within the aquarium environment can significantly increase biofilter clogging.

There are four basic types of biofilter designs: submerged bed, rotating biowheel, trickle and fluidised bed. While all biological filters rely upon the same species of bacteria for bioconversion, its how the different filter types operate that determines their effectiveness. There are numerous aquarium filters that utilise biofiltration, ranging from the humble box filter, which gets little recognition these days, to high tech designs with computer control. However, it is helpful to remember that the filter itself only provides a suitable "home" for the bacteria to colonise. Media for biofilters can be virtually any substrate which provides maximum surface area for bacterial growth: gravel, plastic media and sponge foam pads are among popular choices.

A good biological filter should have the following criteria:

- Large surface area for bacterial growth.
- Adequate open area to avoid clogging.
- Adequate water movement through biological filter and around the surfaces designed for bacterial growth.

The selection of substrates on which bacteria can form colonies has led to a reduction of the size of biological filters. From the initial substrates of gravel or shell used in submerged or trickling filters, now inert fibre cushions, similar to those used in the filters of air conditioners, or small moulded pieces of plastic for packing purposes or else blocks of undulated PVC sheets are utilised. The continuous search for ideal substrates is oriented towards materials offering the highest surface/volume ratio, limited weight, strong mechanical resistance and limited clogging characteristics in addition to being cheap and easy to maintain.

The smaller the particle size, the more the surface area per unit volume. However, when the particle size is reduced, the probability of filter clogging increases and the ability to mix the water within the biofilter decreases. In submerged-bed biofilters, such as undergravel filters, the relationship between surface area and performance is probably asymptotic, because blockage and diminished circulation increasingly hamper performance as substrate particle size diminishes. The rate of water flow through the biological filter should range from 30 to 100 percent of the volume of the entire system per hour. Higher stocking rates will require the highest turnover rate (70–100% per hour).

Basically, a biological filter is simply a surface on which the bacteria grow. While growing, the bacteria convert toxic wastes produced by the fish to less toxic wastes. However, in reality, it is a complex system comparable to a living organism. The biofilter must be "fed" and supplied with oxygen in order to remain healthy and function properly. It also releases carbon dioxide and hydrogen ions as waste products. It can take weeks for bacteria to establish or colonise a biofilter and they grow, age, and die like any other life forms. Although a larger number of water quality parameters affect nitrification.

Dissolved oxygen, pH, water temperature, ammonia concentration, filter flow rate and chemical treatment are the dominant factors affecting nitrifying bacteria efficacy. Surfaces for bacterial growth should be protected from light, which inhibits nitrifying bacteria.

When an aquarium system is first set up or restarted after cleaning, time must be allowed for colonisation of the biofilter. This is a critical time in aquarium systems, because ammonia levels may increase faster than their removal. Biofilters are often started by artificially adding ammonia, such as ammonium chloride, into the system thereby allowing establishment of the nitrifying bacteria prior to stocking with fish. This would also be expected to encourage the growth of autotrophic bacteria because without an organic load, competition for attachment surfaces from faster growing heterotrophic bacteria would be minimal. However, the effect of exogenous addition of ammonium chloride can extend the cycling time dramatically; in some cases, as much as 50–60 days (Shimura *et al.*, 1996)

It may be advantageous to pre-activate the cycle. Pre-activation is accomplished by seeding the filter(s) with a 'starter' bacterial population from an established aquarium. Seeding of nitrifying bacteria has a positive effect on the nitrification process by improving performance and stability. Nitrifying bacteria are usually abundant in the gravel substrate or filters of an established aquarium; so many hobbyists use a small amount of this as a bacterial seed. Previous studies have shown that seeding freshwater systems with 3% or 10% of wet filter media from an established filter decreased the start-up time of a new filter by 48% and 81%, respectively.



The addition of 10% wet filter media from an established system can reduce the start-up time to around 4-7 days compared to 20–25 days for ammonia removal and a similar time scale for nitrite removal (Gross *et al.* 2003).

In recent years, pre-coating of nitrifying bacteria on filter media has also been used in aquarium biofilters to enhance nitrification, using mixed bacterial cultures from natural water and aquarium systems. However, seeding with a wet media of an established filter has a major disadvantage because it might cause transfer of diseases even after the nitrifying enrichment process, as bacteria and pathogens might survive such conditions.

As an alternative to preactivation, a staggered stocking regime can be used. Once the aquarium water is stable, you can increase the animal load to the desired level. The aquarium is stocked with a small number of fish. The bacteria will grow on the wastes from the animals and plants, and a balance will be established between the quantity of the wastes produced by the fishes, invertebrates and plants and the bacterial population. An increase in wastes will result in an increase in bacteria, but the response is not instantaneous. The time required is a function of the conditions under which the bacteria are growing and the nature of the bacteria themselves. When the biofilter bacteria populations grow to meet this ammonia load, additional fish can be added. However, when this technique is used, water changing may be required. You must monitor water quality and be able to correct any problems when and if they occur, there is not much forgiveness and catastrophic failures can happen.

Often ignored by those unfamiliar with the metabolic capabilities of bacteria is the fact that many heterotrophic bacteria and fungi are able to oxidise nitrogen compounds such as ammonia. The pathways of nitrogen transformation are very complex. The ability to denitrify ammonia is a facultative trait spread among a wide variety of physiological and taxonomic groups of nearly 130 bacterial species within more than 50 genera (Zumft 1992).

Newly discovered 'Anammox' bacteria have been found that convert ammonium and nitrite directly to dinitrogen gas. Anammox is an abbreviation for anaerobic ammonium oxidation (ANaerobic AMMonium Oxidation). It is a very recent addition to our understanding of the biological nitrogen cycle and is the most unexplored part of the cycle. Recent studies have also revealed the widespread existence of unique ammonia-oxidising archaea (bacteria-like organisms), belonging to the Archea domain.

New research is showing that archaea capable of ammonia oxidation are ubiquitous in marine and fresh water environments. The phylogenetic diversity and species richness of ammonia-oxidising archaea and bacteria were examined in aquarium biofiltration systems. The results showed that species richness of ammonia-oxidising archaea is greater than those of ammonia-oxidising bacteria. However, the relevance of ammonia-oxidising archaea in aquarium systems remains unknown. **Thus, the nitrogen cycle is much more complex than indicated in most aquarium textbooks.** Even after much intensive study, modern microbiologists still do not fully understand just how the nitrification process functions. Under certain conditions, however, it is conceivable that the complete nitrification process could become disrupted, with elevated levels developing for one or more of the intermediates. Under normal operating conditions, there are a variety of factors that, individually or in combination with each other determines the efficiency of biofilter nitrification. These factors include fish density, ammonia concentration, flow rate, filter surface area, filter media type, temperature, *p*H, salinity, alkalinity, dissolved oxygen, total dissolved solids, and filter type (submerged, trickling, fluidised bed, etc.).

## Loss of Biological Filtration

The ability of biological filtration to adequately control ammonia and nitrite in aquarium systems depends on a variety of factors. Under normal operating conditions, there are a variety of factors that, individually or in combination with each other, will reduce the efficiency of biofilter operation. If the causes of these problems are avoided, the operation of a biological filter will be trouble free. Several common causes of problems with the operation of biological filters include:

- Overloading
- Overfeeding
- Loss of biological filtration (filter media change and/ or chemical treatments)

Overloading — When we start a new system a certain time is required for the bacteria to establish themselves and develop an adequate population within the aquaria. The bacteria will grow on the wastes from the animals and plants, and a balance will be established between the quantity of the wastes produced by the fishes, invertebrates and plants and the bacterial population. An increase in wastes will result in an increase in bacteria, but the response is not instantaneous. The time required is a function of the conditions under which the bacteria are growing and the nature of the bacteria themselves. Suboptimal conditions for the microorganisms slow their activity and growth time. New systems should be started with less than the maximal biomass that the system is designed to handle. Once the aquarium water is stable, you can increase the animal load to the desired level.

Overfeeding — The most common problem in aquarium management is overfeeding, which generates more wastes than the biological filter can handle. This results in cloudy water, spikes, or continuously high levels of ammonia and/or nitrite, rapid mulm accumulation, excessive growth of algae and increased stress and disease susceptibility.

Loss of Biological Filtration — bacteria can grow on all surfaces within the aquarium, but the most important part of the population will be within the biological filter where water flow patterns have been designed to assure that water is rapidly renewed around the bacteria. This is particularly important because water-purifying bacteria have no significant mobility and unless the water is rapidly changed around them, they will deplete their nutrients in the microenvironment around them resulting in reductions in their growth and water purification. The water flow patterns are thus important in the design of a biological filter.



A properly designed filter should contain multiple elements, such as sponge and plastic or porous porcelain, on which the bacteria can grow. All elements should not be replaced at the same time, as this will remove most of the active water purifying bacteria and nitrification may essentially cease for a short period of time. If your filter contains multiple elements such as sponges or cartridges, to remove accumulated solids, simply rinse them off in water and return the filter element to the filter. When it is necessary to replace a worn element, replace only one of the multiple elements at the same time. In this way you will not throw out your most active population of water purifying bacteria. Studies have shown that aggressively-washed filters elements require longer intervals to produce optimal nitrification, and their performance suggests that this is due to substantial biofilm removal. Research has shown that gently-washed filters achieve their highest nitrification rates at relatively short intervals, due to the effect of a low biomass-loss rate.

Another way in which bacterial activity of the biological filter can be lost is with medications containing bactericides. Ideally, sick fish should be quarantined and treated in a separate aquarium from your main tank. If it should become necessary to treat the fishes in the main tank but not sterilise the entire system, stop feeding the fishes prior to the treatment. This will reduce ammonia production. Immediately, prior to adding medication, remove the biological filter element (cartridge or sponge), and place the element in another aquarium or container with aeration; add enough clean water or water from the aquarium to cover the filter elements. When the treatment has been completed and residual bactericidal products have been removed by activated charcoal or otherwise, the bacterial filter elements can be returned to the aquarium. Feeding can now be restarted, but slowly without overfeeding.

## Filter Turnover Rate

The calculation of the turnover rate most frequently used is simply: Volume of system in litres / Rate of pumping litres per minute = turnover time in minutes. Thus in an aquarium with a volume of 1000 litres and a pump rate producing 100 litres per minute, the result is 1000 / 100 = 10 minutes. However, that does not mean that in 10 minutes all the water in the aquarium has passed through the filter (pump) attached to it. It takes much longer to ensure that all the water has passed through the filter.

The explanation is that in the first minute 100 litres (10% of the total volume) of water passes through the filter. At the end of that minute the system contains 100 litres that has been through the filter and 900 litres which has not. Assuming there is good mixing in the next minute, 100 litres passes through the filter but 10% of this will already have passed through the filter. Thus at the end of two minutes 190 litres not 200 has been treated.

The formula to use to work out the time taken for all the water in the aquarium to pass through the filter (pump) is: Volume of system in litres / Rate of pumping litres per minute x 9 = timein minutes for all the water to pass through the filter. Thus in this example 1000 / 100 = 10 x9 = 90 minutes. This is of course assuming there is sufficient mixing of the aquarium water. You can expect an even further decrease in flow rate from 10 to 50 percent once filter material is added.

#### Aeration and Circulation

Aeration and circulation of the water is one of the most essential functions in aquarium systems. Water must be aerated to maintain adequate dissolved oxygen concentrations for fish and for proper functioning of the biological filter. Fish utilise oxygen dissolved in the water and release carbon dioxide and since the biofilter also utilises oxygen, a successful maintenance system requires a method to aerate or oxygenate the water. The amount of oxygen consumed by the fish is a function of fish size, feeding rate, activity level of the fish, and temperature. Consumption of oxygen by the biofilter bacteria is most closely related to the amount of ammonia entering the filter.

Aeration, circulation, and filtration are usually performed at the same time by a well-designed filtration unit. Aeration is usually applied in the fish culture tank and again prior to or within the biological filter, that portion of the recirculating system where organic waste products are broken down through bacterial decomposition. Trickle filters and revolving biofilters are designed to be self-aerating. Vigorous aeration of submerged filter beds is not recommended because beneficial bacteria can be dislodged from the substrate decreasing the filter's effectiveness. Air lift pumps are often used to move water through the tanks, accomplishing both aeration and pumping. Super-intensive systems may use pure oxygen injection although this is seldom used in aquarium hobby. The level of aeration should be sufficient to sustain dissolved oxygen levels above 5.0 mg/L throughout the system.

In an aquarium, a number of functions are performed by aeration or circulation:

- 1. It adds oxygen directly to the water (oxygenates).
- 2. It circulates or mixes the water top to bottom to ensure that the oxygen and temperature levels are uniform throughout the system.
- 3. It moves aerated water away from the immediate area around the filter while dragging in unaerated water rather than retreating the same water.
- 4. Circulation encourages other harmful gases and excess nitrogen and carbon dioxide to escape to the atmosphere.

## Diffused Air (Airstones)

The oxygen transfer efficiency from air diffusers is a function of the bubble size from the diffusers and the way in which they are installed (loose or in airlifts). The aeration effect is usually localised around the diffuser. An airlift improves mixing and helps generate circulation. The finer the bubble the greater the efficiency but fine air-diffusers block more rapidly. However, reducing bubble size from 6 mm to 3 mm can increase the mixing of the water and oxygen transfer efficient by almost 5 times, bubble size is therefore very important.



The size of the bubble is a function of the quality and chemistry of the water. In seawater, or water with a high ionic content, the bubble size can be smaller than 1 mm, as the water becomes fresher, the size of the bubble will increase to around 3 mm in clean freshwater. If the level of fats and lipids is very high, the lipids will tend to reduce water surface tension and increase the size of the bubble.

Air diffusers are usually a solid stone type diffusers or a flexible membrane type diffuser. High levels of carbonates and iron present in the water can foul solid diffusers. If the water is very hard, with an alkalinity over 400 mg/L as calcium carbonate, then the carbonate will tend to come out of solution and form a precipitate on the diffusers. The carbonate deposit will block solid type diffusers. Flexible membrane diffusers are usually resistant to this type of fouling because the surface of the diffuser is flexible, and hard carbonates will tend to crack off the surface. If solid diffusers such as the stone type become fouled by carbonates they can become very difficult to clean.

Air diffusers connected to a compressed air line are usually placed into various culture tanks for live-food production, egg incubation, larval rearing and fry raising. The bubbles they produce help in maintaining the still passive yolk-sac larvae and the first-feeding larvae afloat, and in homogenising the rearing medium (rotifers, microalgae). When an additional quantity of oxygen is required (for example, when there is a temporary failure of the water supply system or when there is a temporary high density in the rearing tanks), one or more air diffusers can be placed in the rearing tanks, connected to a separate compressed oxygen line. The porous material of the diffusers should be periodically cleaned, since small particles (algae, food residues, faeces) in the rearing water can easily clog the pores and reduce (or even block) the air/oxygen flow.

## **General Maintenance**

Fundamental to the success of a thriving aquarium is a stable environment made possible with a regular maintenance program. This includes regular removal of particulate matter (faeces, uneaten food, detritus, etc.). The removal of algae from tank walls, removal of particulate matter from the filter, and regular water changes.

Waterchanges provide regular removal of wastes not normally removed by filtration or bacterial decomposition, and restores a stable environment. The more frequently the water is refreshed, the lower the stresses on the system and its inhabitants. Most rainbowfishes enjoy waterchanges; they seem stimulated by them, so long as they do not involve so great a change in water parameters that they induce stress. Weekly changes of between 25–35% should be employed to avoid any major changes in water quality and chemistry.

Weekly changes of at least 50% will be required for rainbowfishes maintained at high population densities. Waterchanges are one of the easiest things you can do for your fish, and will do more to support their health and longevity than the most high-tech filtration and control systems can do without waterchanges. No systems exist, despite misleading claims to the contrary, that can replace waterchanges. Substratum in heavily fed, overstocked or neglected aquariums can rapidly accumulate organic wastes, especially if the water is not changed on a regular schedule. These wastes will stimulate bacterial growth that uses up oxygen. As the dissolved oxygen in the water is depleted the water becomes polluted and the fish can die. The best way to avoid this problem is to 'vacuum' the gravel each time you do a waterchange. This process removes organic wastes, which otherwise might clog the gravel bed.

The most common apparatus for vacuuming the gravel is a siphon with a large cylinder on one end attached to a long, narrow tube. These devices are generally available from aquarium suppliers. The large end is placed in the tank, the small end in a bucket that is below the level of the tank. Move the large end up and down in the tank to start a water flow into the bucket. Then insert the large end of the siphon into the gravel and slowly move it up and down. If used correctly, it will remove the accumulated wastes from the gravel without removing any gravel. Repeat this over the entire bottom of the tank, until the bucket is full. Then pull the siphon end out of the tank to stop the water flow. If more water is to be changed, this can be repeated. Replace the removed water with new, conditioned water that has been adjusted to the same chemical parameters as the tank.

However, should the gravel becomes contaminated, the aquarium should be drained, cleaned and the gravel thoroughly washed before re-establishing the aquarium. Incidentally, if no more contaminants enter the aquarium, eventually the bacteria most suited to the existing conditions will convert all waste to gases and trace elements (denitrification) and the dissolved oxygen in the water will begin to rise to the point where the water will be safe for fish again.

Although I have detailed a number of routine jobs in the maintenance of an aquarium, the work is not quite as laborious as a reading of it may imply. The less an aquarium is fussed with the better, and, as a rule, if it has been set up in the correct manner in the first place it requires very little further attention. It does not mean that a great deal of time has to be spent every day in looking after the fish and plants. A few minutes each day, to feed the fish, to remove uneaten food, to see that all the fish are in good health is ample. Once a week the aquarist will have to spend half an hour, perhaps a little longer, overhauling the plants, removing excess sediment, water changing, cleaning the filter, and cleaning the front glass. It is important to note that such attention as is necessary must be given regularly, once you neglect an aquarium the whole thing becomes a nuisance rather than the attraction that it can be with a little care.

Finally, let me say, that in all the details of the care and keeping of rainbowfishes in captivity, there is no teacher as good as experience. I can only suggest some general guidelines I have acquired from my own experience over the years. Each hobbyist will have to discover those conditions suited to his own circumstances by experience and practice. Those who treat an aquarium carelessly or indifferently will soon become tired of it, and cast it away accordingly.



In suitable climates, rainbowfishes can be maintained and will breed in outdoor ponds. Although a tropical to sub-tropical group, all rainbowfishes can handle temperatures down to 20° C, and even as low as 10°C for short periods without problems. If you live in a cooler climate you could maintain your rainbowfishes in an outside pond during the summer, as they will all benefit from even a short period outdoors. Generally, rainbowfishes kept in outdoor ponds develop better colouration and will often grow to a size not attained in aquariums. This is because in a pond situation the rainbowfishes have access to the full range of natural pond organisms for feeding. In general, rainbowfishes should have at least some live foods to be healthy and grow rapidly.

An ornamental pond or watergarden provides a wonderful opportunity to enjoy both the natural beauty of rainbowfishes and waterplants. The soothing, visual beauty of ponds is enhanced by waterplants, with an ever-changing view as the rainbowfishes swim among the plants, and the play of light and shadows are reflected in the water. Rainbowfishes are naturally camouflaged and difficult to see from the surface, but the inclusion of waterplants will assist in preventing predation by birds and other animals. All fish tend to prey on each other, and the larger naturally eat the smaller. Nevertheless, some small fry will survive and the overall number will increase if part of the pond is thickly planted. Waterplants serve many roles in ponds; they produce oxygen, which is used by the fish, and help remove waste nutrients. They provide cover for small fish, spawning habitat for adult fish, and home for small aquatic animals, which can be food for the fish. On the negative side, plants while creating habitat/shelter for small fish also provide an ideal habitat for dragonfly larvae, and other aquatic creatures, which can be highly predatory on fry.

Plants available for use in ponds are many, but there are certain considerations to be taken into account. Such things as water depth, amount of sunlight, and whether the plant chosen will survive in the pond, need to be considered. Floating leafed and submerged plants are necessary for a healthy pond and must be included in your selection. Pond size and construction is also very important. The smaller the pond, the greater the impact seasonal and diurnal temperature fluctuations have and the less stable the overall pond environment will be. Minimum size for a healthy balanced pond is considered to be about 5 m<sup>2</sup> of surface area. Another important factor in the overall health of the pond is the depth. Depth of the pond should range from 45 to 60 cm. Greater depths are not necessary and may cause maintenance problems.

Locate your pond to avoid direct sunlight at midday during the warmest months. Rainbowfishes can become stressed by high temperatures unless shade is provided by waterplants. A minimum of 5 to 6 hours of direct sunlight each day is recommended for the best growth and establishment of all waterplants. However, there are endless options for planting a pond and its surrounding area.

Floating leafed plants are usually waterlilies. Plant enough to cover 50 to 75 percent of the surface area of the pond to keep the growth of algae in check. Submerged plants are the oxygenators of the pond - a must if your pond is to be healthy and support fish.

Free-floating plants, such as *Azolla, Lemna*, or *Ricciocarpus* species, though not necessary, add the finishing touch to the natural appearance of the pond. These plants move with the breeze and produce an ever-changing pattern in the pond. However, floating plants can smother the air/water interface resulting in reduced oxygen/carbon dioxide exchange and aeration. Surprisingly a cover of floating plants does not reduce evaporation, as people believe; it actually increases evaporation due to the transpiration/respiration cycle and is called evapotranspiration. They also reduce the amount of light penetration into the water column due to the effects of shading as well as competing for nutrients, resulting in lower phytoplankton productivity in the natural food chain.

Bog or marginal plants are also suitable for the pond. These plants can generally tolerate as little as three hours of direct sunlight. Some grow best in constantly moist to boggy soils, while others actually grow in standing water. There are many different species of bog plants with varying heights, textures, and colours to their foliage. Waterplants, just like other garden plants, will need periodic pruning, dividing, repotting, and fertilising. Fertilisers used in the pond should be slow release pellets that can be pushed into the base of the plants. Caution should be used, as any fertiliser leaching out into the water will cause an algal bloom.

Ponds can be built out of several types of materials. Some of the more common construction materials are earth, liners, fibreglass, and concrete. Choice of construction materials should take into account the life expectancy of the material and installation requirements. The liner is generally the most important and most expensive component of a watergarden.

Some examples of material in order of life expectancy are: PVC (fish grade) – 7 to 15 years Butyl or Rubber (fish grade) – 30 years Fibreglass – 50 years Concrete – Lifetime, if done correctly.

Ponds can be relatively expensive to build and maintain, although many hobbyists start with little expense by using an old wash tub, bath or wading pool. However, it doesn't matter whether your pond is an old truck tyre or a backyard masterpiece with waterfalls and hidden lights, good water quality must be maintained. If not, the pond declines in beauty and the fish become stressed and susceptible to diseases. Once the basics of water quality are understood and practiced, maintenance will become second nature and require only a few hours per week.





"Ponds can be relatively expensive to build and maintain, although many hobbyists start with little expense by using an old wash tub, bath or wading pool. However, it doesn't matter whether your pond is an old truck tyre or a backyard masterpiece, good water quality must be maintained."



The most common water quality problems are oxygen depletions and the build-up of toxic nitrogenous wastes. Oxygen depletions occur because the total amount of plant and animal life has exceeded the carrying capacity of the pond or because of an excessive rate of decomposition. Fish gasping at the surface is almost a sure sign of oxygen depletion. Oxygen consumption depends on the respiration of aquatic organisms, including plants, and the aerobic decomposition of organic material by bacteria; these rates also increase with temperature. This balance needs to be clearly understood; a satisfactory oxygen level recorded during the day is no guarantee that the levels will be maintained during the night. Moderate levels recorded on a warm, sunny afternoon will almost always indicate that severe oxygen deficiencies will occur during the night. Also, lower than expected daytime pH values due to high levels of CO2 may indicate high levels of bacterial respiration which could lead to low night-time oxygen levels.

The other common water quality problem is the accumulation of toxic wastes such as ammonia and nitrites. This problem occurs because of over-feeding, rapid decomposition, or biofiltration failure. Waterplants are active biological filters, and, if a balance is maintained between the number of plants, the number of fish, and the amount of nutrients the pond receives, no other filtration should be necessary. Ponds with abundant waterplants and a modest number of fish should become a balanced system on its own. The key is to maintain water quality and relatively clear water so your fish can be seen and enjoyed.

# The Pond Ecosystem

All factors occurring in the pond, whether physical, chemical or biological, influence the pond ecosystem. The pond ecosystem is of course extremely complicated and intricate. As aquarists, we need to manipulate the ecosystem so as to produce an optimal environment for the rainbowfishes. Food is just one component of this complex system. Some ponds will support adequate plankton communities without any assistance. However, most ponds require some form of fertilising in order to promote plankton development. A better understanding of the pond ecosystem will assist in the management of the pond to promote the natural blooming of favourable microalgal (phytoplankton) species. Different microalgal species have widely varying abilities and demands for nutrient uptake and light utilisation. Parameters that will influence the phytoplankton ecology will include fertilisation (nutrients), temperature and light.

When ponds are first filled with water, there are few living organisms and few nutrients. The water rapidly gains nutrients, particularly if soluble inorganic fertilisers are added. It also gains nutrients, but more slowly, as organic fertilisers are decomposed by bacteria. Phytoplankton and other bacteria rapidly use the released nutrients. Within a few days, growing populations of phytoplankton may provide a green tinge or "algae bloom" to the water. What turns the pond green is innumerable single celled algae. These are present in all water and will create a bloom in any water left undisturbed in full sunlight. The long filamentous algae that grow on the bottom and sides of the pond are not responsible for the discolouration of the pond. However, if you feel the algae is unsightly and needs to be removed, manually pull one end loose and roll it up on a stick, or just pull it up by the handful. Some rainbowfish species will eat filamentous algae, but don't expect it to be rapidly consumed. In time the waterplants will cover most of the pond's surface denying light to the algae.

Established waterplants will eventually out-compete the algae for the available CO<sub>2</sub> and soluble nutrients. Sometimes the pond will suddenly clear overnight as the algae succumb and sink to the bottom. Occasionally throughout the pond's life, this algal bloom may reoccur for a short time. This may happen when the temperature of the water is increasing, or the nutrient levels are up. Algal blooms of short duration are to be expected. This indicates that there is a growing food base for single-celled protozoans and other zooplankton. In many ponds the water first appears brownish. This happens when the bacterial food levels are large enough to cause huge protozoan or rotifer blooms without much phytoplankton being present. Water quality in ponds changes continuously and is affected by physical and biological characteristics. With this in mind water quality should be monitored regularly.

It is important to maintain the pond properly during each season, paying attention to the specific requirements, since the tasks differ widely from one season to the next. In long periods of hot, dry weather, you may need to top up the water level in the pond. Use stored rainwater, if possible. Water from the house supply is likely to have higher chlorine/chloramine content in summer, and topping up with it may encourage alga blooms, and induce stress in the fish. If tap water is all you have available, introduce it in small quantities - no more than 5% of the total pond volume, and no more than once a week.

## **Feeding Fish**

Rainbowfishes may have to be fed some artificial feeds because some ponds just won't have enough natural foods to sustain ideal growth. Large amounts of artificial feeds however, should not be used to feed rainbowfishes in outdoor ponds as it can be detrimental for water quality and is an ineffective feeding method. Feed just a small amount more than they immediately consume and later check to see if the additional feed is eaten. Adjust the amount of feed offered accordingly. The pond will need to be inspected periodically to check natural food populations.

Rainbowfishes should be removed from the ponds, or restocked at lower densities, at the time when the natural food in the pond can no longer support the number of fish. It is not possible however; to give daily dietary requirements for feeding rainbowfishes in ponds as the dietary requirements under these conditions will depend on stocking density, and the availability of natural food organisms.

Feeding should be reduced at water temperatures above 32° C. At high temperatures rainbowfish do not feed well and are easily stressed by poor water quality. Also, do not feed at water temperatures below 10°C. Rainbowfishes will not feed at lower temperatures because their metabolism decreases.



The natural food items that are available in ponds can be divided into three broad categories, these being plant material (phytoplankton), animal material (zooplankton) and detritus (decomposing fragments of organic material derived from both plants and animals), as well as organisms that are not easily classified into any of these groups (such as protozoans and bacteria), Rainbowfishes will feed on all these organisms.

There are a number of different species of aquatic animals that will inhabit the pond. The most important live food found in a pond is aquatic insect larvae and zooplankton. These animals are very high in protein which is necessary for the growth of rainbowfishes. Zooplankton consists mostly of rotifers, cladocerans or copepods. The ability of rotifers and cladocerans to reproduce parthenogenetically (asexually) enables them to react quickly to favourable and unfavourable environmental conditions. Rotifers (40–600  $\mu$ m) have the shortest life span (5–12 days) and can reach their peak reproductive level in about 3-5 days. At 20-25°C, the egg-to-egg span is 1-3 days. Cladocerans (0.2-3.0 mm) and copepods (0.3-3.0 mm) have similar life spans of approximately 40-50 days, but with different peak reproductive periods. Egg-to-egg generation times are 7-14 days for copepods compared to 6-8 days for cladocerans at 20-25°C. To reach their peak reproductive capacity, cladocerans require 14-15 days while copepods require 24 days at 20-25°C. Copepods, which have only sexual reproduction, require longer periods to increase their population levels.

Cladocerans are desirable live food since they have high protein value and are readily consumed by most rainbowfishes. However, cladoceran populations usually decline rapidly when subjected to predation in ponds. On the other hand, copepods, because they are swift swimmers are better able to maintain their populations during the later stages of pond culture.

Rotifers are often the earliest visible zooplankton to appear in ponds. Rotifers feed on bacteria and phytoplankton, and then reproduce to form huge populations. That usually happens 2-3 weeks after the ponds are filled and when water temperature is 20-28°C. As rotifers eat their own food supply the population drops drastically. Then copepod nauplii, adult copepods and cladocerans make their appearance. Together, copepods and cladocerans prevent a re-bloom of the smallest rotifers. However, modest populations of larger rotifers may appear after several weeks, particularly when the fish prey on the rotifers' competitors and predators - cladocerans, copepods and aquatic insect larvae. Rotifers hatch from "resting eggs" that survived on the pond bottom during unfavourable conditions. Most of them hatch into females that reproduce asexually until pond conditions become unfavourable. Then sexual reproduction occurs and resting eggs are again produced.

For larger juvenile rainbowfishes, the smallest rotifers may not provide enough nutrients to make chasing and ingesting them worth the effort. Juvenile rainbowfishes are more predatory than



the adults and require a higher proportion of animal protein in their diets. Most juvenile rainbowfishes will eat zooplankton. For the small rainbowfishes, such as the newly hatched larvae, small rotifers may be the only zooplankton small enough to eat. Although copepod nauplii can also be important first foods for rainbowfish larval. Protozoans may also be eaten, but little is known about their contribution to rainbowfish larvae diets.

Although little live phytoplankton is eaten directly by rainbowfishes, it is one of the most important components of the pond food chain. Plant material can come from many sources including microalgae, aquatic plants, reeds and rushes. With all of these fresh plant materials, especially ones containing low protein, the actual nutrient value to rainbowfishes is relatively low. In many cases when they are eating plant material they are only acquiring a few vitamins and minerals. Microalgal species can vary significantly in their nutritional value, and this will change under different culture conditions. Nevertheless, microalgae can offer an excellent nutritional food for larval rainbowfishes, either directly or indirectly (through enrichment of zooplankton). Plant materials become far more nutritious after they have been in the pond for a couple of weeks after they begin to decompose. At this stage they are colonised by tiny aquatic animals, bacteria and fungi and begin to break down into detritus.

As plants decompose they become broken down into tiny fragments. The fragments become colonised by bacteria and fungi which feed off the decomposing material. These tiny fragments and the microscopic plants, animals, bacteria and fungi associated with them are known as 'detritus'. Detritus is a major component of the diet of rainbowfishes at all stages of their life cycle. The tiny plant fragments themselves are not very nutritious but the micro-organisms associated with them are a readily digestible, nutritious, protein rich food source. The naturally occurring detrital food available can be supplemented by adding small amounts of organic plant materials such as hay and lucerne which will break down most rapidly and effectively to form healthy detrital communities.

## **Outdoor Growing Ponds**

Outdoor ponds are also perfect for raising newly-hatched rainbowfish larvae and/or juveniles. An abundance of zooplankton is particularly important for larvae to develop into juveniles and for juveniles to develop into sub-adults. Rainbowfish larvae will feed on zooplankton through to the transition to adults. The larvae are not particular about the types of live foods they eat, but the organisms must be small enough to fit into their mouths.

The successful rearing of rainbowfish larvae in an outdoor pond does however, requires specific management of the pond to enhance phytoplankton and hence zooplankton development. The aim of pond rearing therefore is to maintain high densities of desirable zooplankton species until the fish are removed from the pond or weaned onto artificial feeds. These ponds are usually fertilised with organic or inorganic nutrients to encourage the development of phytoplankton blooms which, in turn, produce zooplankton blooms upon which the stocked larvae feed. Rainbowfishes larvae can be hatched or transferred directly into outdoor ponds to feed on these naturally produced live foods. However, there may be some advantages in having an initial 10–15 day rearing phase indoors, before moving them into on-growing ponds. Larvae can be stocked at densities of  $\sim$ 100 larvae/m<sup>2</sup> of pond surface area.

The proper timing of fish stocking is also important for optimum growth of the fish. The pond must contain the appropriate type and size of food when the fish are stocked. Larger juveniles (>25 mm) stocked into ponds with very tiny zooplankton may grow slowly because the fish must expend so much energy to catch an adequate amount of food. Likewise, if the zooplankton is mostly too large for larvae rainbowfishes to eat they may starve. Most rainbowfish larvae (4-6 mm or less) fall into this category. When ponds are filled and fertilised, the plant and animal populations that invades or hatches from within the pond pass through a somewhat predictable change in sizes and species. At first there are usually a few small species in large concentrations. Later there will be many species in an array of sizes, but each in moderate concentrations. The average size of organisms also gets larger with time. The early community is unstable and great changes can occur quickly; later, the greater diversity of organisms makes the community more stable.

Although some protozoans may be large enough for tiny rainbowfish larvae to eat, it is the next stages in succession that are of greatest importance for growth. To maximize survival, stock any larvae just as populations of zooplankton small enough for the larvae to eat are rapidly increasing. The larvae will then have the right size food for rapid growth and can better escape from any carnivorous aquatic predators that may begin to populate the pond. Stocking even larger juveniles into a pond that has been established for some extended period of time can result in predation.

In an established pond, a variety of fish predators would have colonised the pond and begun to reproduce. These include insects such as back-swimmers, diving beetles and whirligig beetles. Later, even larger insects such as water scorpions, giant water beetles and the larval stages of dragonflies will appear. Insects begin to colonise as soon as ponds are filled during the warmer months. However, it usually takes several weeks for their populations to reach levels threatening to small fish. Rainbowfishes are active predators and are well adapted to catching the smaller free swimming forms of aquatic insect larvae. Rainbowfishes will also seek out and eat aquatic worms, snails, ants and flying insects of allochthonous origin that may have fallen into the pond. These organisms form a significant component of the diet of rainbowfishes.

## **Pond Fertilisation**

The purpose of fertilisation is to promote an algal "bloom" without necessarily trying to promote a particular alga species. There is no point in fertilising ponds that have very low *p*H values (<5.0) or very low total alkalinity (<20 mg/L). Alkalinity stabilises *p*H and facilitates the uptake of inorganic carbon by algae. Carbon can also be supplied to the algae when



carbon dioxide is released following the decomposition of organic fertilisers. Likewise ponds with very high clay turbidity will not respond to fertilisation. However suspended clay particles can provide suitable sites for active bacterial colonisation and these ponds often have very good natural zooplankton populations.

The dynamic characteristics of zooplankton populations have led commercial aquaculturists to use particular fertilisation techniques and species-specific zooplankton inoculations in culture ponds. The intent of these management techniques is to maintain high densities of desirable zooplankton species in the ponds. Some aquaculturists have had considerable success in managing zooplankton populations through phytoplankton management. The most important diet component of these animals has been shown to be small algae (1–25  $\mu$ m). Algae larger than 50  $\mu$ m or algae with spines or in colonies are usually rejected.

Fertilisers may be either inorganic or organic based. Inorganic fertilisers may be either inorganic or organic based. Inorganic fertilisers are those that take the form of granular or liquid fertilisers having high phosphorus content and, to a smaller degree, nitrogen (phosphorus is often the limiting nutrient in freshwater). The premise behind using inorganic fertilisers is that by applying needed nutrients, phytoplankton populations' increase. These increased populations of phytoplankton will then increase the number of zooplankton in the pond, which then eat the phytoplankton. However, it has been shown that large phytoplankton populations; zooplankton will eat more fungi and bacteria associated with decaying organic substances than phytoplankton directly. In fact, these large populations of phytoplankton often lead to reduced water quality.

Organic fertilisers may be animal manures, hay and lucerne (ground or meal), or soybean meal. Organic fertilisers should have small particle sizes to allow rapid decomposition. They can be broadcast over the pond or placed in porous mesh bags for slow release into the water; this will help prevent the organic matter from floating around the pond. Another method is to pre-soak the dry material for several hours, and then distribute the wet material over the bottom, allowing it to slowly decompose. As previously indicated, zooplankton will consume fungi and bacteria associated with decaying organic material. However, be aware that the use of organic matter may cause pH fluctuations, dissolved oxygen and ammonia problems during the initial decomposition.

Ponds should be fertilised as they are being filled. Using a combination of organic and inorganic fertiliser results in a greater diversity of plankton than if either fertiliser type is used alone, and reduces the potential for a bloom and bust (crash). Organic fertilisers are the basis of the food chain that nourishes bacteria, protozoans, zooplankton, and eventually the fish. As organic fertilisers decompose, their nutrients are used by phytoplankton, which will be consumed by the rainbowfish larvae and smaller juveniles. The phytoplankton will also be eaten by protozoans and/or zooplankton before they are eaten by the fish. Nutrients from organic fertilisers are released over time, so they produce less drastic changes in plankton populations than

do inorganic fertilisers. Inorganic fertilisers add nutrients to the pond instantly. A phytoplankton-based food chain can develop very rapidly without the need for bacterial action. However, the nutrients are often used up very rapidly by the phytoplankton, and the risk of a bloom and bust is greater than it is with organic fertilisers.

Fertiliser nutrients are used quickly in the pond environment. Some nutrients are trapped in the bottom sediment or otherwise lost from the water. Therefore, nutrients should be replenished often. Frequent applications of small amounts are more effective than a single large application for maintaining a constant supply of food organisms.

The succession patterns and species composition of zooplankton in natural environments may not be the same as in intensively fertilised culture ponds. In a study of fertilised culture ponds without fish, it was found that copepod adults and nauplii, and daphnia populations reached maximum mean densities in an average of 23.5 days. Rapid population declines of copepod adults and nauplii occurred in 5.3 days, respectively, while daphnia and bosmina populations decreased significantly within 7.3 days after reaching maximum densities.

Aquaculturists have different recommendations concerning the time between filling the ponds and fish stocking. Some recommend that culture ponds be filled 2–3 weeks prior to stocking to allow time for maturation of zooplankton populations. However, not all fish species require the same size of prey at the onset of feeding. For instance, some species have very small mouths that require them to consume small prey, such as rotifers and early instars of cladocerans. Improved survival may be achieved by stocking these fish species into culture ponds filled only 2–3 days before stocking.

Cladocerans, which are coloured a deep red are often indicators of low dissolved oxygen conditions, and quickly become clear when placed into well-oxygenated waters. This coloration is based on the increased amount of haemoglobin that these animals have to compensate for low oxygen levels in the environment; however, this increased amount of haemoglobin comes at a cost. The increased number of diapause eggs in cladocerans also indicates another indication of poor environmental conditions. These diapause eggs are often quite large and dark and are produced when these animals are forced to undergo sexual reproduction in preparation of unfavourable environmental conditions.

When cladocerans are food-limited, they mature at a smaller size and produces smaller offspring. The main response of *Daphnia pulex* to low food levels was a reduction in size specific food intake and egg size. Cladoceran populations also consist of smaller individuals in water bodies with large populations of vertebrate predators. Large-bodied species, e.g., *Daphnia pulex*, tend to be fewer in ponds with large predator bases. In these situations, smaller species or smaller individuals within a given species have improved chances of escaping predation than larger individuals (based on prey visibility). However, smaller animals can also be selected when predators are other invertebrates, such as midge (Chironomid) larvae, or backswimmers.



It is not the intention of this section to discuss the cultivation of aquatic plants, their structure, or classification. These details are readily available in any basic aquatic plant book. Nevertheless, since the natural environment of all rainbowfishes includes waterplants; live, flourishing plants are an important part of maintaining a wholesome captive environment.

Aquatic plants are recognised as an integral component of natural aquatic ecosystems because they increase habitat diversity and directly modify aquatic habitats. They are important habitats for the swarms of tiny animals that inhabit waterbodies. The cyclical uptake and release of carbon dioxide and oxygen during photosynthesis and respiration produce cyclic fluctuations in the concentrations of these gases and hence cyclic fluctuations in dissolved oxygen concentration and pH. Aquatic macrophytes further alter in-stream habitat by reducing water velocities, stabilising river substrates and influencing water temperatures. In Australia's inland river systems, where relatively few waterplants now grow, there are relatively few aquatic animals. Rainbowfishes that do live in these river channels cluster around submerged snags and tree roots.

In their natural environment rainbowfishes use aquatic plants for shelter and refuge, as a food source either directly or indirectly, in the form of epiphyton (periphyton) and associated invertebrates, and as spawning and nursery sites. Rainbowfishes larvae need aquatic macrophytes as shelter and protection from predation or to avoid cannibalism. In general the macrophyte habitat not only provides protection from predators, but also rich foraging microhabitats for larvae rainbowfishes, allowing higher growth rates and fecundity, and lower mortality. Rainbowfishes that inhabit aquatic plant habitats feed mainly on autochthonous items, represented by diatoms, chlorophytes, microcrustaceans, and aquatic insect larvae.

In an aquarium, living plants serve most of the same functions that they do in nature. Plants provide shelter, shade, and for some rainbowfishes - food, especially the duckweeds. Plants in an aquarium also contribute to the oxygen content of the water and assists in maintaining water quality. In addition to this, plants provide huge surface area for colonisation by other micro flora and fauna. In tanks where plants are growing well, rainbowfishes behave more normally and display better colouration. Finally, aquatic plants add to the beauty, interest, and naturalness of the aquarium.

Aquatic plants are not difficult to keep healthy and attractive, as long as their basic requirements are met. As is the case with terrestrial plants, these include adequate light and fertilisation, a suitable growing medium, proper water conditions and temperature. The importance of proper lighting cannot be over-estimated, but too much light is almost as bad as too little light. Keep the aquarium well away from a window or natural light and use artificial light. In this way you can control the light independently of the vagaries of natural weather; and should not be affected by a rampant growth of algae.

Planted aquariums are often referred to as "natural" aquaria. However, the "natural" aquarium is an unattainable ideal. It implies an aquarium in which all the inhabitants are interdependent on each other, as they are in a natural stretch of water, and it takes very little thought to understand that this result could not be achieved in an aquarium. Strictly speaking a natural aquarium would be one where we wouldn't have to feed the fish, change water, learn water chemistry, provide lighting or any of the other things we have to do to maintain an aquarium successfully. No matter how much we may wish it, regrettably, there is no such thing as a natural aquarium. Keeping rainbowfishes in an aquarium cannot, even under the best conditions, be anything equal to their life in the wild. At best, an aquarium is an artificially controlled environment that is suitable for maintaining fishes, plants, and other aquatic life forms for extended periods.

In a loose sense, however, a "balanced" aquarium is attainable, by not overcrowding the fish, by stocking with plenty of plants, and in a way we are imitating Nature, even if we have to help her by feeding the fish, trimming the plants, and removing from time to time much of the sediment that collects on the bottom. A state of balance would occur if everything (food, lighting, fertiliser, etc.) that we supply the aquarium is taken up by the fishes, aquatic plants, algae and other life forms to the same degree as they are being supplied.

Many hobbyists are disturbed by the presence of algae and do their best to try and remove it. Perhaps this should be taken as evidence that a 'state of balance' is occurring. Maybe not the particular state of balance that you desire, but it is a biological balance all the same. However, the truth is that no matter how biologically balanced your aquarium is, over time water quality will deteriorate and must be changed on a regular timetable.

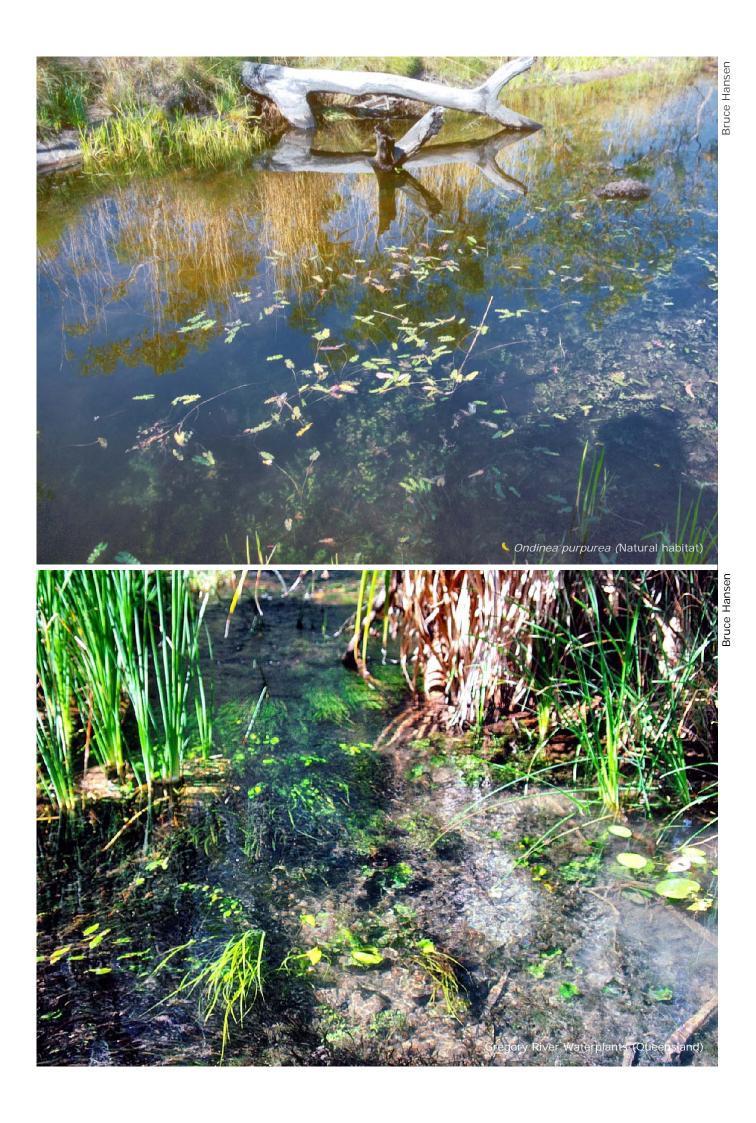
## Australian & New Guinea Aquatic Plants

Many waterplants are unique to Australia, although some are also found elsewhere in the world, with the Australian populations representing local varieties rather than distinct species. However, the aquatic flora of Australia is not dissimilar to that found in most other tropical or subtropical regions of the world. Although some endemic species are found, most species are cosmopolitan. This relatively low degree of endemism and diversity in aquatic flora is probably because of the cosmopolitan nature of many aquatic plant species. A mixture of distance dispersal, barriers and local speciation, can explain the development of Australian aquatic flora. Climate is obviously the most significant barrier. However, very few comprehensive aquatic flora surveys have been conducted within Australian freshwaters.

The aquatic flora of New Guinea has not been studied to any significant degree neither. Many of the river mainstreams are turbid, which precludes the establishment of submerged aquatic macrophytes. The number of species is probably less than 200. However, species-level treatments exist for only a small portion of the flora.







Cryptocoryne is a genus of perennial aquatic plants found growing in a variety of tropical riverine or swamp habitats, adapted to fluctuating water levels during seasonal flooding and drought. Leaves are attached at the crown by sheathed petioles as long as or longer than the leaf. Leaves ovate to lanceolate, tapering at the tips and rounded at the bases. Leaf surfaces smooth, margins entire, commonly wavy. Submersed leaves 4–10 cm long, 1–4 cm wide. Upper surfaces to green to brown often marbled in darker brown or patterned with prominent venation. Lower surfaces nearly green to brown and tinted with a glistening violet. Inflorescence enclosed within a spathe, a fleshy ornate bract. Immature spathes are sometimes present on submersed plants and appear as tightly rolled tubes. Mature spathes will unfold only when emersed from the water. Several features of mature spathes, specifically the colour of the collar and the twist of the terminal limb, are used in species identification.

There are no *Cryptocoryne* species found growing naturally in Australia. In New Guinea however, there are at least three species that have been reported. *Cryptocoryne versteegii* have been found in the Lorentz and Kikori River systems. *Cryptocoryne ciliata* are reported from the Merauke and Kikori River systems, and *Cryptocoryne dewitii* has been collected from the Kiunga region in the Fly River. Unidentified specimens of *Cryptocoryne* have also been reported from the Wapoga River.



The typical habitats of *Cryptocoryne* are mostly streams and rivers with slow-flowing water in the lowland forests. They also live in seasonally inundated forest pools or on river banks submerged only at high water.





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The vegetation of New Guinea is more closely allied to the flora of western Asia than to that of the Australian continent. For this reason it has been termed "Malesian", part Asian and part Melanesian.

Authors inevitably differ in their concept of which species should be included in a taxonomic treatment of aquatic plants. Each has their own definition for the term "aquatic". Virtually all authors recognise those plants with a submersed or floating growth habit as aquatic. Most also apply the term to the common emergent species. The definition is harder to apply consistently for (a) plants growing in marginal zones of wetlands, e.g., floodplains, swamps, streambanks, etc., and (b) plants displaying a wide-ranging ecological amplitude which enables them to grow in either wet or dry situations. Whether a plant is to be designated as aquatic or not is thus based upon the plants' growth habit and the types of habitats in which it is found. How much weight is assigned to either of these criteria is a subjective decision which largely accounts for differences in the taxa treated by various aquatic plant manuals.

Algal and aquatic moss commonly comprise Charophyta (stoneworts) and Chlorophyta (green algae) which forms macroscopic mats either attached to plants or in open water. Floating vascular/leaved plants have part or all of the leaves at the waters surface. Examples include *Azolla, Lemna, Spirodela* and *Wolffia* and the genus *Utricularia*. Members from the family Potamogetonaceae (pondweeds) are also common floating plants and can be found in a variety of habitats. Rooted vascular plants are those rooted in the sediments with either a major proportion of material above water (reeds, rushes and sedges) or totally under water. Typical genera include *Baumea, Bolboschoenus, Carex, Cyperus, Gahnia, Schoenus, Juncus, Triglochin, Blyxa, Myriophyllum* and *Vallisneria*.

Floating and floating-leaved plants occur in permanent waters in most coastal rivers and wetlands across Australia. Typical northern assemblages include *Nelumbo nucifera*, *Nymphaea gigantea*, *Nymphoides indica*, *Ottelia ovalifolia*, *Azolla*, *Ludwigia*, *Marsilea* and *Pseudoraphis* species. These may be found in the river channels and pools in the tropical coastal plain. On the floodplains of the Northern Territory and the Kimberly, such assemblages may be fringed by *Melaleuca* swamp forests.

In coastal floodplain systems, species include Azolla filiculoides, Nymphaea gigantea, Potamogeton tricarinatus, Ottelia ovalifolia, Nymphoides indica and Lemna species. In wetland landscapes, species include Potamogeton tricarinatus, Azolla pinnata, A. filiculoides, Ottelia ovalifolia, Lemna trisulca and L. disperma. In south-western Australia, Najas marina, Myriophyllum propinquum, Lemna disperma, Azolla filiculoides, Spirodela oligorrhiza, Potamogeton tricarinatus, P. pectinatus, Nitella congesta, Chara baueri, and Ottelia ovalifolia dominate.

Floating and floating-leaved communities are associated with the inland Murray–Darling river system and include *Azolla filiculoides*, *A. pinnata*, *Potamogeton tricarinatus*, *Spirodela oligorrhiza* and *Lemna disperma*. While the water level maintaining such communities usually remains between one and two metres in depth, it may dry out completely for periods. Coongie Lakes supports floating plants of *Ludwigia peploides*, *Azolla filiculoides* or *Lemna disperma*, while Goyder Lagoon supports a system dominated by *Polygonum spp.*; these plants may be entirely dependent on groundwater. The same holds for the *Marsilea* species in the river pools of the Pilbara.

Most of the above wetlands are fringed by submerged and emergent herblands in shallower waters. In northern Australia, species include *Triglochin procera*, *Caldesia oligococca*, *Limnophila brownii*, *Ludwigia adscendens*, *Ceratophyllum demersum*, *Monochoria cyanea*, *Vallisneria nana*, *Utricularia*, *Myriophyllum*, *Eriocaulon* and *Chara* species. Species associated with such communities on the east coast of Australia include *Ludwigia peploides*, *Najas marina*, *Vallisneria australis*, *Triglochin procera*, *Myriophyllum propinquum*, *Potamogeton crispus*, *P. ochreatus*, *Nitella* and *Utricularia* species.

In south-western Australia, communities including *Halosarcia* halocnemoides, Sarcocornia quinquefolia, Wilsonia humilis, Triglochin procera, Lepilaena preissii, Najas marina, Ruppia maritima, Potamogeton pectinatus, P. ochreatus, Villarsia albiflora, Persicaria decipiens, Chara baueri and Nitella species occur in coastal sand dune swamps and on the fringes of lakes. Equivalent emergent and submerged herblands exists in coastal and highland Tasmania, in the extreme southeast of South Australia, and on the tablelands of south-eastern Australia.

The genus *Aponogeton* is a group of freshwater aquatic plants occurring in Australia and New Guinea. *Aponogeton* are popular aquarium plants and for many years they were collected from the wild. However, conservation measures introduced by Australian Governments now restrict this practice. Members of the *Aponogeton* genus listed as threatened species include *A. bullosus*, *A. prolifer*, *A. cuneatus*, *A. queenslandicus* and *A. elongatus* subsp. *elongatus*.

Many specimens of Aponogeton from northern Australia were originally assigned to A. elongatus, but are now assigned to A. euryspermus, A. vanbruggenii or A. tofus. These species initially produce submerged leaves in flowing or clear water but later in the year plants often develop floating leaves. Species in this group can be separated by their seed shape and size; A. euryspermus has large broad seeds that can become very thick when mature, A. tofus has narrower almost cylindrical seeds, while A. vanbruggenii has even narrower seeds with a distinct knob at one end. All have seeds with an outer testa that is easily removed (thick in A. euryspermus and A. tofus – thin in A. vanbruggenii). All species are confined to the tropics with A. euryspermus more westerly in its distribution, A. vanbruggenii more easterly and A. tofus growing between the two but overlapping with A. vanbruggenii. A. tofus is closely related to A. euryspermus (as it had been previously identified); however, it is distinct and quite divergent at the molecular level from that and other species.

The genus *Limnophila* commonly known as Ambulia, has given the aquarium hobby some beautiful and well-known aquarium plants. *Limnophila* is a genus of aquatic or semi-aquatic plants that are found in Australia and New Guinea.





In Australia five *Limnophila* species are currently recognised: *L. aromatica*, *L. australis*, *L. brownii*, *L. chinensis* and *L. fragrans*. Reports of *L. indica* found growing naturally in Australia are *L. brownii*.

The genus *Myriophyllum* contains species of hardy and adaptable plants for both temperate or tropical aquariums and watergardens. They are amongst the best submerged aquatic plants for a pond where rainbowfishes are kept. With their finely divided foliage in dense swirling masses, they provide the perfect place for rainbowfishes to deposit their spawn and for the fry to start their early life. Species more suited to somewhat shallower waters includes *M. papillosum*, *M. simulans and M. verrucosum*. Other water milfoils remain mostly submerged at all times.

*Potamogeton* is a genus that is strictly aquatic, and is rooted in the substrate with either floating (*P. tricarinatus*) or submerged leaves. They produce flowers just above water level and a number have floating foliage for some part of the year. This is a small but cosmopolitan family, found almost anywhere permanent, still or flowing, fresh or slightly brackish waterbodies are found. Similarly, in Australia they can be found anywhere there is suitable habitat, including the arid inland.

*Rotala* species are typically found in damp soil adjacent to water and occasionally in shallow water. It is assumed that they germinate in response to inundation and reach maturity after water has receded. Six species have been recorded in Australia: *R. diandra*, *R. mexicana*, *R. occultiflora*, *R. rosea*, *R. rotundifolia* (naturalised) and *R. tripartita*.

The taxonomy of *Vallisneria* in the past has been confusing and inconclusive with numerous species being described and these names (often incorrectly) have been widely used in the aquarium hobby. The genus *Vallisneria* was reviewed in 1997 (Jacobs & Frank) with the addition of new species and some clarification of existing names. Recent phylogenetic analyses (Les *et al.* 2008) of *Vallisneria* material collected throughout Australia, Asia and North America has revealed considerably higher diversity in *Vallisneria* (11–13 species) than previously described with the addition of two new species and the transfer of *Maidenia rubra* to *Vallisneria*.

These studies have identified Australia as the centre of diversity for this genus. It is speculated that *Vallisneria* species were once considerably more abundant than is the case today. The depauperate riverine flora within the regulated inland rivers of south-eastern Australia suggests that their preferred habitat has suffered greatly from altered flow and water quality.

Confusion over names and identifications has been mainly resolved by detailed examination of specimens collected throughout Australia. *V. nana* was formally separated from *V. americana*. *V. nana* has over the years been described as *V. spiralis* and both *V. nana* and *V. annua* have been previously described as *V. gracilis*. Both *V. nana* and *V. annua* have basal leaves and tufted shoots. *V. nana* is usually perennial and from perennial habitats, whereas *V. annua* is mostly an annual from ephemeral habitats. Female shoots of *V. nana* only flower

when several years old and have fewer flowers per plant. *V. annua* produces many flowers per shoot (>12) when the plant is still small and young. Reliable identification however, is only possible by examining floral structures. The two species also have distinct leaf characteristics.

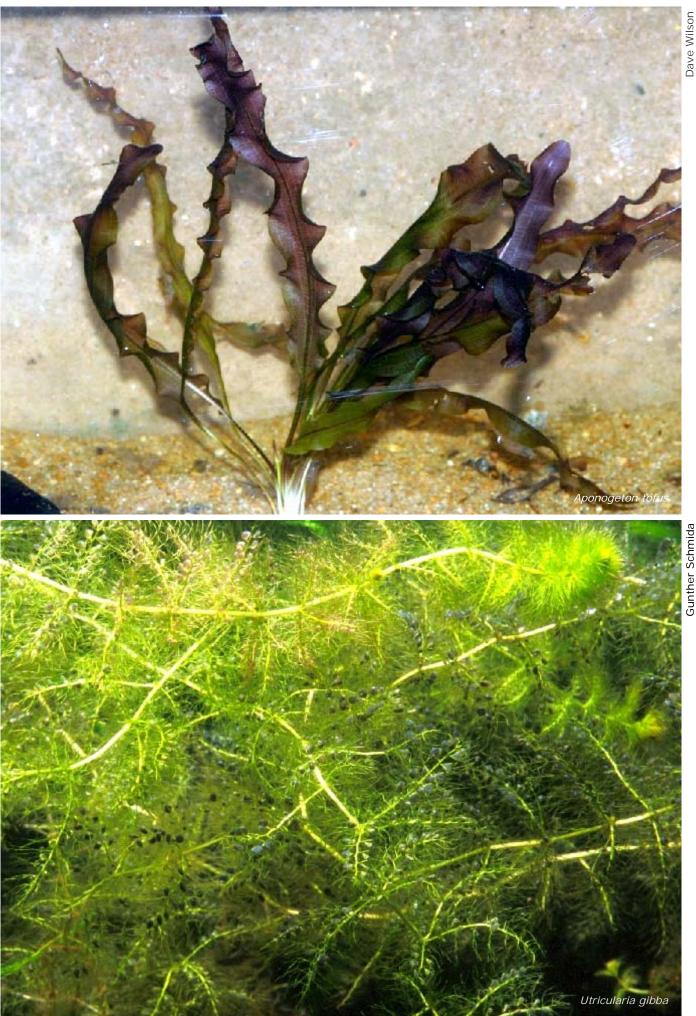
*V. caulescens* and *V. triptera* are two closely related species from northern Australia that grow as stem plants, with leaves arranged alternately along the stems, rather than as compact, basal rosettes. Both are more closely related to other *Vallisneria* species than to species in any other genus. The only exclusive character they share within *Vallisneria* is the cauline leaf arrangement, a character also shared by the florally distinct *Maidenia* (*Vallisneria*) rubra (stamen 1, 3-locular) and *Nechamandra* (female flowers sessile with a long delicate hypanthium).

*Marsilea* species are semi-aquatic fern allies with distinctive fronds shaped like clover leaves, which may be emergent or floating during inundation. At least seven species are known in Australia. They are mainly distinguished from each other by their reproductive structures, which are called sporocarps and are located towards the base of the fronds. One species, *Marsilea latzii*, is moderately salt tolerant and is rare. Many of the species readily grow in highly temporary water bodies such as very small clay depressions; however, *Marsilea mutica* may favour longer term and more frequently inundated wetlands. Standing water is probably required for the spores of all species to germinate, and many or all species thrive in shallow water for extended periods of many months.

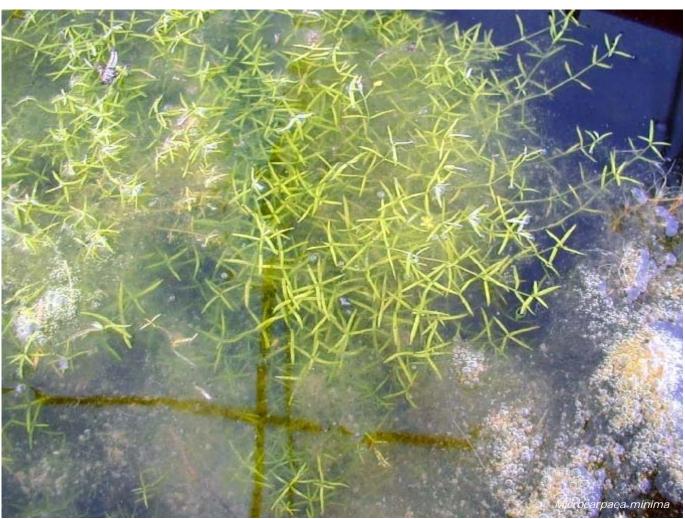
*Isoëtes muelleri* is a fern-like plant that is semi-aquatic or amphibious but has more of the appearance of a grass. It may not require free water to complete its life cycle. *Hygrophila* species occur in tropical regions in Australia and New Guinea. They are common on floodplains, swamps and waterholes with apparently three species in Australia. *Hygrophila angustifolia* is the most common.

Pond plants can include the Australian water lilies which include at least 9 species, all of which are much more coldsensitive than the exotic tropical hybrids available through water garden nurseries. Many of these Australian species have been lumped together under *N. gigantea*. The true *N. gigantea* has round petals, and varies from blue to white to pink, fading with age. *N. violacea* is sharper-petalled, blue to mauve or sometimes white to pink. White species include *N. elleniae*, and *N. pubescens* which is usually tinged pink. *N. immutabilis* may be blue or white, always with a blue tinge. The sacred lotus (*Nelumbo nucifera*) is one of our largest aquatic plants.

In cooler climates other plants can be used in place of water lilies, including the related *Brasenia schreberi* with its unusual elliptical leaves and purple flowers. *Nymphoides* species are occasionally referred to as water lilies due to a general resemblance to *Nymphaea* species. Many *Nymphoides* species have very waterlily-like leaves, but the flowers are much smaller and often heavily fringed. *N. indica* is white-flowered with a yellow centre. *N. crenata* has serrated leaf edges in most forms. *Ottelia ovalifolia* is also very decorative but not easy to maintain.



**Gunther Schmida** 



Other shallow waterplants can be suitable, particularly *Alisma plantago-aquatica* with rather heart-shaped leaves held high, although the shivering masses of tiny white flowers are also very decorative. *Mimulus gracilis* produces carpets of bluish, yellow-centred flowers for months on end in a good season. *Mimulus* occurs worldwide, but most of the ~160 species are part of two large radiations centered in western North America and Australia. The Australian genera *Peplidium* (14 species), *Glossostigma* (seven or eight species), *Microcarpaea* (two species) and *Elacholoma* (two species) are considered to be members of Mimuleae. However, the systematic placement of the genus and the relationships among species within it remain unresolved.

There are many aquatic and wetland plants found in Australia and New Guinea that are suitable for the rainbowfish aquarium or watergarden. They are not restricted in variety—its just that they are poorly known!

Most aquatic plant species lack the mobility necessary to travel directly from one catchment to another, to colonise new areas and to disperse to neighbouring water bodies in different catchments. Despite this apparent isolation of freshwater habitats, many of these aquatic species have widespread distributions consistent with the frequent dispersal by migratory waterbirds. Many aquatic plants can survive intervening drought periods as dormant stages, or recolonise temporary dry wetlands via hydrological connections, especially via floodwaters entering from rivers.





Rainbowfishes have often been reported in the aquarium literature as being suitable for brackish water aquariums. However, all rainbowfishes are permanent inhabitants of freshwaters. There are, in fact, few recorded observations of rainbowfishes in saline waters and can generally be considered as a species most intolerant to salinity. Where rainbowfishes have been collected in "brackish" water conditions it is probably due to stratification of the freshwater flow (lens) above a saltwater wedge caused by substantial freshwater runoff. Surface water sampling does not provide any information on the flow stratification nor on the firesh/brackish interface characteristics.

Rainbowfishes are therefore not a suitable candidate for a brackish aquarium. However, brackish conditions are the preferred environment for a number of *Pseudomugil* (blue-eye) species. *P. cyanodorsalis, P. inconspicuus, P. majusculus,* and *P. signifer* are regularly found in brackish mangrove estuaries. They are found in estuarine and coastal freshwater habitats across northern Australia and southern New Guinea. Although *P. cyanodorsalis* are mostly found in brackish mangrove areas, I successfully bred and raised them in freshwater over a period of three years. Many hobbyists, however, report that they will live longer, breed more freely, and produce more offspring if maintained in brackish water.

The various species of fish found in oceans, lakes, rivers and streams have evolved over millions of years and have adapted to their preferred environments over long periods of time. Fish that can tolerate only very narrow ranges of salinity are known as stenohaline species. These fish die in waters having a salinity that differs from that in their natural environments. Fish that can tolerate a wide range of salinity at some phase in their life-cycle are called euryhaline species. These fish can live or survive in wide ranges of salinity, varying from fresh to brackish to marine waters. A period of gradual adjustment or acclimation, though, may be needed for euryhaline fish to tolerate large changes in salinity. Ultimately, fish adapted to or inhabited marine, fresh or brackish water because each environment offered some competitive advantage to the different species.

*Pseudomugil cyanodorsalis* have been collected from around Broome and Wyndham in northwestern Australia and from the vicinity of Darwin, Northern Territory. They have also been collected from coastal areas around the Gulf of Carpentaria in Queensland. I have no doubt that this species will eventually be found in southern New Guinea. *Pseudomugil inconspicuus* has been found at the mouth of the Fly River, southwestern Papua New Guinea and Bintuni Bay, Irian Jaya. They can also be found in scattered localities around northern Australia. *Pseudomugil majusculus* have been collected from brackish water on the northern coast of New Guinea, near Cape Ward Hunt. *Pseudomugil signifer* are widely distributed along the eastern coast of Australia and are usually abundant in brackish mangrove estuaries. Most of the Australian fish fauna is contentiously hypothesised to be derived from marine descendants, and are sometimes labelled as "secondary" freshwater fish. The rainbowfish family are one of the groups considered to have been derived from relatively recent marine ancestors, and therefore could be expected to retain at least moderate salinity tolerance. However, there is limited scientific information of salt tolerance available for rainbowfishes.

One study (Kefford, et al.) on the sub-lethal effects of salinity on both male and female fish of M. s. splendida was undertaken in 2006. Preliminary results found that better growth rates occurred at low salinities than in higher salinity. It was also found that egg numbers and hatching rates were reduced at the higher salinity levels. Poor hatching rates of eggs and survival rates of fry has also been reported from moderate increases in salinity. It is important to note the sensitivity of earlier life stages because the occurrence of adult fish at a given salinity at a particular site on a given sampling day may not necessarily indicate a viable population. The most sensitive stages appear to be pre-hardened eggs, post-hardened eggs and fry (particularly when the yolk sac is exhausted). Another study found deaths occurred at salinities between 4.4 and 13.2 mS/cm. There is also evidence that the salinity tolerances of younger fish is less than the acute tolerances of older fish.

Salinity tolerance refers to the ability of an animal to withstand exposure to salinity for an indefinitely long period without dying. Tolerance to salinity is in part due to the physiological mechanisms and morphological adaptations that act to balance concentrations of salts in the cells and tissue of an organism against the external environment. In this way, salinity tolerance can vary between species, populations and to some extent can vary between individuals of the same population. Salinity tolerance may also be due to environmental factors that affect the duration of exposure and rate of increase in salinity concentrations.

Vast stretches of mangrove estuaries are common around Australia and New Guinea where fresh and salt-water mix producing a unique brackish environment containing its own distinctive fauna. These estuaries are tidal, but during the wet season, freshwater flowing in from the flooding rivers dilute the saltwater to fresh, water thus varies from saline through brackish to fresh. The estuarine sections include deep-water over bottom sand and mud, encompassing densely vegetated mangrove zones. Subtidal and intertidal wetlands are found adjacent to riverine channels and are generally shallow, probably less than 0.5 m., with subsequent aquatic beds consisting of algal and vascular species.

The thought of setting up a brackish water aquarium may seem more trouble than what its worth. However, a brackish water aquarium suitably set-up with treated mangrove roots, rocks, and sandy substrate can make a very attractive display. It is very simple to maintain a brackish water aquarium. However, like any aquarium, if you do not understand the animals, their needs and what it takes to keep them successfully, your brackish water aquarium can rapidly turn into a nightmare. The secret of success is to provide good quality water and to keep only a limited number of compatible animals in the aquarium.

Brackish water conditions can be obtained by mixing freshwater with seawater. Seawater is more than just Sodium chloride, and for this reason adding ordinary salt is not an adequate substitute. Seawater contains a variety of salts, and the best way to make brackish water is to use sea salt (marine mix) sold for saltwater aquaria.

Typically seawater has a density of around 1.025, which means one litre of seawater weighs 1.025 kilogram (approx). For the record, seawater has about 35 grams of sea salt dissolved in one litre of water. If you want one-tenth seawater, or about 1.002 to 1.003, you would make full strength (1.025) seawater and then dilute it with freshwater in the ratio one part salt to nine parts fresh. One half seawater, about 1.012, would be made by mixing one part salt water to one part fresh.

The salinity of the water required will depend on the fishes being kept. Blue-eyes are from the fresher end of the range and will enjoy a salinity of less than 1.005. Fishes that live mostly in seawater prefer a salinity of over 1.010. One litre of seawater to every nine litres of freshwater makes an acceptable mixture for most brackish fish. Always make the saltwater up separately, following the instructions on the package. Saltwater is highly corrosive and most metals will corrode quickly when continuously splashed by saltwater. It is therefore important to make sure that no metal objects are used inside the aquarium and metal lighthoods and stands should be avoided.

Water temperature should be maintained between  $20-24^{\circ}$  Celsius. A *p*H of 7.8–8.4 will be acceptable. Like any other aquarium, levels of ammonia, nitrite and nitrate should be kept to a minimum. Most brackish water fishes appreciate moderately hard, alkaline water conditions. Sea salts contain carbonates and bicarbonates which tend to buffer the water well, so hardness and *p*H shouldn't be too much of a problem. A good way to buffer the water is to use calcareous media in the filter. This can consist of crushed coral, shellgrit, or similar material in the filter or as part of the substrate.

The most natural substrate for a brackish aquarium is fine sand, but this is not ideal for use with an undergravel filter. If you wish to utilise an undergravel filter, you could use a sheet of fine mesh plastic flyscreen to stop the sand from being pulled below the undergravel plate. Coarse-grained calcareous beach sand is also useful (the type used in marine aquariums). This will produce a buffering substrate to a moderate extent.

Partial water changes should be made every 14 days and when changing the water do not forget to add salt supplemented water. Remember that this is for doing a water change, not for replacing evaporated water. Do not use brackish water to replenish water lost to evaporation. Use freshwater only as dissolved salts do not evaporate with the water. If brackish water is used to top up the tank, the salt concentration will increase, and before long you will have a saltwater aquarium.



# Rainbowfishes Breeding & Raising



Photo: Leo O'Reilly

With declining habitat conditions in many parts of Australia and New Guinea, saving the existing aquarium gene pool and the integrity of each individual species should mean more to rainbowfish keepers than their just being a passing aquarium adornment. Therefore, the breeding of rainbowfishes should be a central role for the serious rainbowfish keeper.

Unfortunately, the initial numbers of each individual species collected from New Guinea has been very small resulting in a limited genetic base from which to establish larger aquarium populations. This can be clearly seen in several of the New Guinea species where the quality and colouration has diminished greatly in captive stocks. Aquarists must give careful consideration to the choice of brood stock if genetic 'pollution' of the aquarium stock is to be prevented. Ideally, a number of individuals should be sourced from the wild population every so often.

To maximise genetic diversity captive populations need to be perpetually managed so an adequate number of separate brood stocks are maintained with frequent transfers between them. For the most part, aquarium breeding has effectively resulted in domesticated strains of rainbowfishes that may be well adapted to life in captivity, but are far removed from their life in the wild.

Very little is known about the biology, ecology and natural life history of rainbowfishes in the wild. Most information is mainly based on aquarium observations. Rainbowfishes are highly social and form schools for some or most of their lives. Strong sexual dimorphism is present in the species with males typically being larger and brighter in colouration. The behaviour between the sexes also appears to vary with females and juveniles forming small social groups that stay together while solitary males cruise in search of mating opportunities. Males can also be highly territorial and will engage in spectacular fin-flashing displays during contests with rivals over the acquisition and defence of spawning sites (submerged logs, rocks, vegetation) close to the water's edge. Females that are ready to spawn often move between territories, inspecting males along the way.

Males play an active role in courting females and will often swim over to display to passing females. If successful in his efforts, the female will follow the male to his territory and scatter her eggs amongst the spawning site. In the wild they tend to spawn amongst stems and roots of marginal aquatic vegetation, especially where the substrate slopes up to the bank. After spawning, the female will leave, while the male remains displaying to passing females and thus defending his territory and the fertilised eggs. The eggs are attached by adhesive chorionic filaments or tendrils to a range of submerged physical structures, including gravel substrates, woody debris, root masses, aquatic vegetation and submerged marginal (riparian) vegetation, which hide them from predators. Spawning frequency in the wild is unknown, but it is likely that the number of spawnings per year would be much less than that observed in captivity. In their natural environment rainbowfishes are generally aseasonal breeders, spawning opportunistically at intervals over a period of about four to five months of the year. Most however, show a peak in reproductive development during the early-wet season (November to May) in floodplain rivers, but can also occur in streams during the dry season (August to November). Rainbowfishes usually move out of their dry season habitats to take advantage of the extensive flooding, which often coincides with an increase in food resources.

Flooding also increases the area and diversity of aquatic habitats available. The young are spawned when food is plentiful and when aquatic plant communities are most dense, affording them protection from predation. Although, spawning will vary from region to region and in some cases from headwaters to lowland reaches when flow rates are reduced and the occurrence of a water flow resulting from sudden rain is significantly reduced. However, rainbowfishes will normally breed when environmental conditions ensure maximum fertilisation and larval survival. They generally spawn over a large area in slow-flowing waters and the backwaters of floodplain areas. The presence of extensive spawning enables them to 'spread the risk' from predators. This strategy increases the chances of some eggs surviving.

Rainbowfish larvae are subject to enormous mortality, with estimates as high as 99.99% mortality during the early larval stage. This mortality occurs in various ways. Displacement from favourable nursery areas can occur during the larval stage and has been shown to cause mortality. Likewise the condition or health of larval and juvenile fish is an important factor regulating mortality during the first year of life. Indeed, it has long been proposed that levels of recruitment and subsequent survival are determined during the 'critical period' which occurs between hatching and first-feeding, with survival related to the period between when fish require exogenous nourishment and when such nourishment becomes available.

It has been suggested that in lowland Australian rivers, spawning of many native fish is a predictable event that occurs regardless of variations in environmental conditions such as hydrology (Humphries & Lake 2000). This suggests that recruitment failures originate from poor larval survival rather than a lack of spawning activity. Larval growth generally has a positive relationship with temperature, due to the direct metabolic advantage of warmer water and also because increasing temperatures are often associated with increased primary and secondary production.

The precise regulatory role of environmental factors on the reproductive cycles of rainbowfishes is not known. Spawning is regulated by external environmental factors that trigger internal biological functions. The internal biological functions that regulate spawning are similar for most fishes. External environmental factors that control spawning, however, vary considerably.





Environmental factors that have been shown to play a significant role in the reproduction cycle of rainbowfishes are: photoperiod; water temperature; water quality; flooding and water flow; rainfall and availability of food. These factors do not function independently of each other, but are interrelated. Actual spawning occurs in response to short-term stimuli such as colouration or pheromones of a mate. Pheromones are chemical messengers like hormones but instead of carrying information within an individual they carry information between individuals of the same species.

Fortunately, rainbowfishes in captivity are very adaptive and will breed under a variety of conditions, and are in the main, influenced by photoperiod and temperature. Subtle changes in water chemistry will also have some influence on the spawning of rainbowfishes. Therefore, maintaining them in an appropriate captive environment will generally ensure successful breeding.

Successful breeding will depend on a number of things such as nutrition, temperature, the number of fish in the aquarium, the age of the fish, and sex ratio. To breed rainbowfish successfully you need to provide them with suitable conditions and they will do the rest. Water temperature can be maintained at around 28° C ( $\pm$  1°C). A photoperiod of 14 hours light: 10 hours of darkness usually gives the best result. Other water quality components are not a major factor providing they fall within that shown in the accompanying water quality table.

In addition to the regular aquarium, breeding usually requires one or more separate aquariums for conditioning the breeders, spawning, and rearing the juveniles. Before attempting to breed your rainbowfishes, you should feed them with the best food you have available for at least two preceding weeks. Most species will breed successfully when fed commercially manufactured fish food (43% protein; 12% lipid), but feeding with live and high-protein frozen foods will maximise egg numbers. Feed at least every day making sure you don't overfeed.

To increase your success, and prevent uncontrolled spawning, try separating the males from the females during this two-week period. Separation of the sexes elicits a synchronisation of spawning that result in a larger number of eggs.

Java moss or any similar 'aquatic moss' is a suitable live plant for spawning rainbowfishes and grows well under the low light conditions of most breeding set-ups. Even so, spawning mops are my preferred spawning medium. By simulating a spawning substrate (plants, etc.), they serve as egg collectors and provide a place for egg attachment. Spawning mops can consist of bundles of fibrous material arranged in a variety of forms and made from a variety of different materials. However, one of the easiest methods is to use acrylic thread (about 8 ply), boiled to remove excess dye, cut to the desire length and tied together in the middle. For a more natural appearance, you could use green coloured thread to simulate aquatic vegetation.



Spawning Mops (photo: Leo O'Reilly)



Typically, spawning mops are suspended in the water column or laid along the bottom or sides of the breeding aquarium. The mops can be attached to a block of styrene foam and floated in the water. You can provide several mops to offer the males a choice of spawning sites and females a choice of hiding places. Egg-filled mops are removed from the spawning aquarium and placed in another aquarium for incubation and larval rearing. Eggs are best left attached to the spawning medium to minimise handling stress.

Before being used for different rainbowfish species, mops should always be boiled or sterilised to destroy any eggs or pests, which may still be attached to the mops. This procedure also effectively precludes inadvertent hybridisation resulting from accidental carryover of eggs from the breeding aquarium of one species to another, and demonstrates a significant advantage that artificial spawning medium has over live plants.

Rainbowfishes are sexual dimorphic and can be sexed by their colour and finnage. Males are generally larger, more colourful and have extended finnage, while females are pale by comparison, and have smaller or more rounded appearance in the fins. Generally, the larger males can usually be identified from the elongation of posterior rays in the second dorsal and anal fins. In addition, males generally are larger and have a deeper body than females.

These physical differences make it relatively easy to identify mature males. However, female characteristics such as plumpness of the abdomen are subjective and can be misleading. If you are unable to sex your fish, then have someone with more experience do it for you.

Male rainbowfishes can also be highly territorial and will engage in spectacular fin-flashing displays during contests with rivals over the acquisition and defence of spawning sites. Males display to each other by extending their dorsal and anal fins, while at the same time intensifying their colouration. The extension of the fins is an illusionary aspiration by the males to increase their overall body size as they complete with each other for the attention of a female. This is often accompanied by a side-slapping action while swimming together side by side throughout the aquarium. Extended fin and colour intensity is also undertaken by the male when displaying to a female, only this time it is an attempt to increase their overall attractiveness to the females. Group spawning with multiple males and females is the preferred method of choice for breeding rainbowfishes because it produces less stress on all the participants. Breeding males can be overly aggressive at times. Therefore, it is best to have three (or more) males, as with two males, one will dominate the other, and in effect prevent him from breeding, while a single male may cause undue force on the females. Three on the other hand will keep inter-male aggression within bounds and spares the females physical abuse.

In the wild, a female has the choice of either spawning or fleeing. In captivity, the flight of the female is reduced or confined to the size of the breeding aquarium. Non-receptive females will move away and swim to the surface or hide amongst the aquatic plants or spawning mops, remaining motionless to avoid detection. Therefore, the size of the breeding aquarium is vitally important and should be appropriate for the species being bred.

In a large aquarium, it is possible for males to have their own spawning space. This should allow the females to choose their own mate and in doing so; a variety of genes will be passed on to the next generation. In addition, this method should provide you with more eggs. Females spawn a limited number of eggs at a time, and on average, 60–80% of females set up for group spawning will produce eggs.

Pair breeding is also an acceptable method and is especially well suited for selecting individual fish for genetic reasons. Breeding aquaria should not be less than 50 litres in size, and are probably best left completely bare with a small internal sponge or box filter. Choose your breeders and place them in the breeding aquarium. Spawning activity should commence within 24 to 48 hours. Spawning behaviour typically begins half an hour after the lights are turned on in the aquarium, with a peak of activity around 1–2 hours later. Activity then gradually declines until all spawning activity has finished, usually by mid afternoon.

During pre-spawning activity, males can become quite aggressive and actively pursue the females. Actual spawning is preceded by vigorous coercing by the male. The male swims around the female with all his fins expanded, making repeated sideways motions or "nods" in the direction of the female, while at the same time intensifying his colouration. During this procedure the colours in both sexes become more intense, but to a lesser extent in the female. Males frequently "flash" an instantaneous brightly coloured band that runs from the upper lip to the first dorsal fin, which they flash on and off like a neon sign. Depending on species, the colours range from white to yellow, orange, rustic red and light blue.

When receptive, the female will enter the spawning site first, closely followed by the male. In the final phase of spawning, the male presses against the side of the female and, accompanied by much trembling action from both fish, eggs are expelled directly among the plants or spawning medium. The eggs are spherical and opaque with many filaments originating from one small area of the shell. On coming into contact with a surface such as plants or spawning medium, the filaments adhere and contract so that the eggs become suspended by a fine thread.





The eggs are negatively buoyant in freshwater and become clear to light amber a few seconds after spawning. Eggs average in size between  $0.5-1.5 \pm 0.5$  mm in diameter. Water hardening (The swelling process of flaccid newly shed eggs when they first contact and absorb water) of fertilised eggs varies according to the water hardness. When hardness is low, increase in egg diameter is greater.

Usually one to three eggs is deposited at a time, during which time 50–100 eggs can be produced. In captivity, however, with limited space and artificial substrate, females may spawn all their eggs at the same time. The number of eggs shed by a single female is directly related to the size of the female with large females spawning from 40–250 eggs, with a fertilisation success rate generally around 70–80%.

The total number of eggs released will increase with the maturity and size of the fish. Large females (>50 mm TL) produce more than 100 eggs per day at the peak of their spawning. Smaller females (30–35 mm TL), which were only just sexually mature shed fewer eggs, 20–30 per day and do not spawn each day. Females usually only spawn once each day; however, males will often spawn with more than one female in one day. Spawning may be repeated every 3–5 days depending on nutrition, temperature, the age of the fish, and sex ratio.

Survival of eggs is often reduced by the predatory activity of the parent fish during and after spawning. Despite what you may hear or read most rainbowfish will eat their eggs, maybe not all of them, but they will certainly eat what they can find. I have never seen a rainbowfish that doesn't eat their eggs. The breeding male will attempt to keep all the other fish away from his spawning site and eggs, but by and large it doesn't work too well.

To insure maximum egg survival, check the spawning medium regularly. If you have a continuing problem with egg eating, you might like to use a stiff bottle brush as a spawning medium. Commercial breeders use these to spawn tropical fishes that regularly eat their eggs. The stiff bristles function as a medium in which the adhesive eggs can be laid, while at the same time, discourages the breeders from eating the spawned eggs. Feeding live foods can also reduce any egg-eating behaviour. A notorious egg-eater often encountered when breeding rainbowfish is a little freshwater flatworm called planaria. If you get them in a breeding aquarium, they can consume a whole spawning of eggs within hours. The eggs hatch into well-developed larvae after an incubation period of around 5–7 days, depending on temperature (151–152 hours @  $25^{\circ}C \pm 1^{\circ}C$ ). At hatching, the larvae measure around 3–4 mm, and are well developed. They are strong swimmers with well-developed pectoral fins and a continuous median fin fold, beginning dorsally and continuing around the tail. The larvae swim at the surface of the water, generally within the upper 1-cm water layer. The mouth is well developed and functional, and they begin feeding within hours of hatching.

They can be fed small amounts of greenwater, infusoria or finely powered fry foods such as Sera Micron<sup>®</sup> or TetraMin<sup>®</sup> Baby Food, four or five times a day. From the end of the second week (12–14 days) brine shrimp nauplii and/or microworm can also be given to the developing young to supplement their diet.

In their natural environment, such as tropical waters, which have prevailing high temperatures, rainbowfishes generally grow faster, mature younger, and have a shorter life span than rainbowfishes in temperate waters. In captivity however, rainbowfishes characteristically display a wide range of sizes, growth rates and life spans, depending on conditions such as food, space, numbers, competition and water temperature.

The growth rate of the rainbowfish larvae is initially slow, with little variation until around 12–14 days. After that time growth rates increased. Initial slower growth might be related to either absorption of the yolk sac or diet. As the larvae increased in age, the variation in length between individuals also increased. Suitable water temperature and adequate food generally results in higher growth rates.

Food is an important factor affecting growth, especially in the early larval stages. The preferred size of prey for larval fishes increases as mouth size and feeding competency increase. Providing natural green-water with resident zooplankton (which contains various invertebrates including rotifers, paramecium and nematodes) as food for the newly hatched fish has several advantages. The larvae are easily able to switch to different sized prey, a feature not present in monocultures of organisms such as rotifers or brineshrimp. Green water also enables the zooplankton to feed on resident algae and microbes, thus retaining their nutritional value for greater periods of time.



An interesting subject matter is hermaphroditism in rainbowfishes. Gerald Allen (1982) noted that *Chilatherina fasciata* are sometimes hermaphroditic; that is both male and female reproductive organs are present in the same individual. Nick Romanowski (1994) reported his observations of apparent hermaphroditism in *Melanotaenia fluviatilis*. This could well indicate that hermaphroditism may be more widely deployed in rainbowfishes than previously thought.

#### **Breeding Blue Eyes**

Breeding blue-eyes in captivity is essentially the same as for rainbowfishes. Blue-eyes are a relatively short lived species in the wild and females may only spawn once, usually at around one year of age, rarely living to spawn a second season. Males often live around two years. In captivity, life expectancy can increase to four years or more if appropriate aquarium management procedures are employed.

The secret in maintaining long-term captive populations of all the blue-eye species is to constantly breed them. There is a reduction in the frequency and intensity of spawning activity in fish over one year of age. If you fail to notice the change they become too old and then you just lose them. Try to obtain young specimens and breed them early and regularly and you will always have some around to enjoy. Because each female only lays a few eggs each day, it can take a while before you have significant numbers. If possible, start with 6-12 juveniles rather than adults pairs.

In nature, blue-eyes are essentially seasonal breeding fishes, commencing spawning activity as temperature and day length increases. In captivity, they will breed throughout the year if given suitable conditions. The easiest way to breed blue-eyes is to set up a pair in a small 50 litre tank containing an over-abundant growth of Java Moss and some floating Water Sprite (*Ceratopteris thalictroides*). With a bit of patience and small feedings of live foods to condition the female, they should do what comes naturally.

Daily observation of the tank should eventually find some fry hiding in the Water Sprite. You can either remove the parents or take a chance that the fry will survive. If you wish to raise larger numbers, it would be best to set up a small group of eight or more in a larger aquarium and provide them with artificial spawning mops. You then check the mops twice a day if possible. If you find eggs attached to the mops you can either exchange the egg-laden mop for a new one or remove the eggs by hand. The fertile, waterhardened eggs of blue-eyes are reasonably hard and rather large for a small fish.

Incubation times vary both among species and with temperature. At 25°C, hatching usually takes between seven and twenty-one days. During this time, you should be able to observe the development of the eggs. In addition to periodic movement within the egg, developing embryos display prominent eye-spots, and it is usually refer to eggs that are close to hatching as being "eyed-up". Discard any eggs that become white and fluffy as they will be infertile.

Once hatched and after the period of yolk absorption, the larvae move to the surface to feed. Carefully removed them to a previously set-up aquarium making sure that the water conditions are as close to the hatching water as possible. Alternatively, you could move the egg-laden mop to a previously established raising tank and allow the eggs to hatch naturally. This way you would not have to collect the newly-hatched larvae, which can sometimes be a bit tedious.

Although the blue-eye species are only small their newly hatched larvae are rather large and can be fed finely powdered dry or liquid fry foods, microworms, newly hatched brineshrimp or similar fine foods upon hatching. They should also be fed several times a day if possible. Their growth rate will depend on the water and food quality provided with maturity being reached within one year.

#### Artificial Incubation of Eggs

Whether you have an unintentional spawning in your aquarium or a well-planned breeding program, to raise rainbowfishes successfully you must be able to provide the right conditions. Developing embryos and newly hatched larvae (fry) are the most sensitive and delicate of the developmental stages in the life of a rainbowfish. Therefore, great care must be taken to provide them with a proper incubating and hatching environment.

High mortality rates can often occur, especially during the early stages. Mortality can be the result of several factors including inbreeding, inferior water conditions, improper incubation conditions and poor nutrition. However, hatching rates and survival can be increased using artificial incubation. Collecting eggs from the breeding aquarium is the best way to ensue the largest number of offspring from your selected breeders. Also, removal of the eggs may increase egg production by shortening the time for another spawning to occur.

For those species that spawn a large number of eggs each day, best results will be achieved by providing them with spawning mops and then moving the egg-laden mops to a previously established nursery aquarium for incubation and larval rearing. Alternatively, for those species that only spawn a small number of eggs each day the simplest method would be to just hand pick the eggs from the spawning mops. Fertile, water-hardened eggs are reasonably hardshelled and can easily be collected from the spawning mop with clean fingertips. You will find that they can be rolled between your fingers without damage. Any eggs that burst while being collected will most likely be infertile.

Spawn-laden mops should be softly squeezed or allowed to drip dry. Individual eggs will stand out like tiny glass beads against the darker-coloured strands of the spawning mop. Place the eggs into clean plastic hatching containers for incubation and provide gently aeration. Once they hatch the larvae move to the surface to feed. The larvae should then be carefully transferred into a previously set-up nursery aquarium as soon as possible.





Males frequently "flash" an instantaneous brightly coloured band that runs from the upper lip to the first dorsal fin, which they flash on and off like a neon sign. Depending on the species, the colours can range from white to yellow, orange, rustic red and light blue.



Alternatively, the hatching container can be floated in the aquarium in which the larval fish will be reared. The larvae will swim at the water surface of the container after hatching and can be gently poured out of the hatching container into the surrounding water in the aquarium. This way you won't have to collect the newly-hatched larvae, which can sometimes be a bit tedious, and it eliminates the need for any physical handling of the larvae.

The amount and incidence of light received during incubation can affect both fish development and larval survival. Therefore, all types of egg incubating containers should be stored in such a manner to protect developing embryos from direct light.

A wide variety of devices and methods can be used for incubating rainbowfish eggs. Depending on your needs, you can utilise a system of several small containers attached to a common recirculating system or a few small individual hatching aquariums.

The simplest method is to just place them in a small shallow tray or container filled with pre-conditioned water. A gently movement of the hatching water with an airstone should be provided to insure adequate water circulation to all the eggs. This will prevent the accumulation of waste products, and allow gas exchange between the egg and the surrounding water. Because of their size and permeability, fish embryos and larvae are susceptible to many types of organic or inorganic materials dissolved or suspended in the water. Therefore, it is essential to provide good water quality for the embryos and larvae.

The spawning of rainbowfishes, embryo development, survival, and growth of larvae all occur within a narrow range of water temperatures. Temperature is one of the major factors in determining the embryonic period for rainbowfishes. The development and hatching is delayed at low temperatures, and accelerated at high temperatures. Incubating temperatures are also known to modify the behaviour of larvae and determine certain morphological characteristics. There is an optimum temperature range required for each developmental life stage.

Water temperatures should be maintained with minimal fluctuations  $\pm 1^{\circ}$ C. In general, optimum temperatures for hatching and rearing all rainbowfish species are within the temperature range of 24–28°C. Avoid temperatures above or below this range. Poor embryo survival, low hatch success, reduced growth rates, larval deformities, and increase in larvae diseases may result from temperature fluctuations or temperatures outside the optimum range for the species. If needed, the hatching container can be floated in a heated aquarium to maintain the correct temperature.

Depending on species, the hatching time for rainbowfishes and blue-eyes is around 6 to 21 days within the temperature range of 24–28°C. Freshly fertilised eggs, which show no external signs of cleavage, are called 'green'. When eye pigmentation and further development are visible through the chorion, eggs are 'eyed-up'. At hatching, when the rainbowfish larva leaves the egg it is provided with a yolk sac, and from this, it derives sufficient nourishment to tide it over the first few hours or days of its free-living existence. This means that there is a certain period of time where it is not necessary for a larval fish to obtain their nutrition from external sources.

Rainbowfish larvae can survive for as long as 8–10 days without external food sources providing that the yolk sac contained enough nutrients. However, if an egg does not hold adequate nutrients, larvae hatching from that egg would have a shorter period of unfed larval life than larvae hatching from an egg with a good supply of nutrients.

At hatching, rainbowfish larvae measure around 3 to 4 mm, and are reasonably well developed. They are competent swimmers with well-developed pectoral fins and a continuous median fin fold, beginning dorsally and continuing around the tail. The larvae swim at the surface of the water, generally within the upper 1-cm water layer. The mouth is well developed and functional, and they usually begin feeding within hours of hatching. Therefore, you will have to commence feeding them as soon as possible.

#### **Incubation & Hatching Problems**

A small percentage of eggs will fail to develop, usually because they were not fertilised in the first place. Dead (opaque) eggs rapidly develop a fluffy appearance due to fungal infection and should be removed regularly. Chemical treatment during incubation is commonly done for the control of fungal infection.

The common water fungus, *Saprolegnia* starts to grow on dead eggs and if not controlled will spread to live eggs, killing them. If the growth becomes too much to control by hand, chemical treatment with a methylene blue treatment can be administered at any time after the eggs are water-hardened, until one or two days before hatching is anticipated. Eyed-eggs are generally not affected by fungal infections. Add sufficient methylene-blue to produce a concentration of 3 ppm. One treatment is usually all that is necessary, and should continue for at least 3 days.

Removing dead eggs with an eyedropper is probably more effective than chemical treatment at controlling fungus, but it can be very time-consuming. Very high fungal infections within the first 2 or 3 days after spawning typically indicate a high percentage of infertile eggs. This may be due to unfavourable conditions within the spawning tank or adult infertility. A sudden upsurge in losses later usually signals improper water conditions in the hatching container, or egg collection damage. Eggs can also be very sensitive to changes in pH, dissolved oxygen, and temperature.

It is suspected that vertical transmission (transmission from parent to offspring) of disease can occur through rainbowfishes eggs. Vertical transmission of pathogens allow the continuous and simultaneous spread of a disease from one generation to another. Although they are not affecting the adult fish, these organisms can have devastating effects if transferred to the larvae. The transfer from one generation to the other can



happen in two ways: on the egg and in the egg. For most pathogens however, the route of infection is on the eggs and then to the larvae when they hatch. Disinfection of the eggs with iodine can be carried out for the various fish species but it is most commonly used in aquaculture. For rainbowfish species, preliminary tests should be conducted to determine when and at what concentration disinfection can be carried out safely.

A problem often encountered when raising rainbowfish larvae is a little freshwater cnidarian called hydra. They are usually tan or brown in colour and are not readily seen against a background of natural coloured gravel or on plants. Within the confines of a small nursery aquarium, these little monsters can be deadly, and can ingest a batch of newly hatched rainbowfish larvae in less than a week.

Another pest that you may encounter is a free-living flatworm known as Planaria. They are a very small black or brown flatworm that look very similar to leeches and often appear in freshwater aquariums. They are generally around 3 to 5 mm long, but some grow as large as 10 mm. In a normal aquarium situation they usually don't cause any problems and probably even go unnoticed. However, if you get them in a breeding or hatching aquarium, they can destroy a whole spawning of eggs within hours. They can usually be seen crawling around on the front of the aquarium at night time after the lights have been turned off.

#### **Nursery Aquarium**

The ideal nursery tank should be bare with only a sponge filter. In this way the tank is easily kept clean and helps prevent any disease or water quality problems. During this time, rinse the sponge regularly under lukewarm running water to keep the surface area clean and free of blockage. A small trickle of air bubbles is sufficient because more air will agitate the water too much and the juvenile fish will have to fight the current. As they grow you can increase the airflow rate.

To achieve the best growth and survival, an initial stocking density of not greater than one juvenile fish per litre of water is recommended, and water temperature should range between 24–28° Celsius. Juvenile fish can remain in the nursery tank until they are large enough to be transferred into a regular aquarium. However, this period should not exceed 90 days due to the increased growth, and the potential for the occurrence of adverse conditions of water quality. Generally, 95% survival should be expected at the end of the nursery period.

General maintenance consists of changing a little of their water every second day with a small siphon tube, removing any uneaten food and faeces, and adding pre-treated replacement water. Any mortalities or deformed fish should be removed regularly. Clean and disinfect all hatchery equipment with a chlorine solution, or other suitable disinfectant before using them for another batch of fish.



Mystery Snails (*Pomacea bridgesii*) can be a useful addition to the nursery aquarium as they help clean the tank of detritus and eat any surplus food, which has a very positive effect on water quality. As an added bonus, Mystery Snails will often breed in the tank in which the juvenile fish are being raised.

A native Australian snail (*Notopala sp.*) can also be used. Notopala River Snails are endemic members of the family Viviparidae and are found predominantly in the northern tropical region of Australia. However, their distribution includes the large drainage basins of the central and northern regions, and in much of south-eastern Australia. They are absent from Tasmania, southern Victoria and south-western Australia. They are mainly found in riverine habits inhabiting permanent and ephemeral waterholes, where they can be found along the banks, attached to logs and rocks or crawling in the mud. They appear to be able to resist drought, possibly by burrowing themselves in drying mud and sealing the operculum shut in order to reduce the risk of dehydration. However, it is unlikely that they would be able to survive prolonged drying periods.

These large snails resemble Mystery Snails; however, they do not lay eggs. They release fully developed small snails, hence the name "viviparous" (live-bearing snail). Like other species in the family Viviparidae, the females brood their young to a crawl-away stage, rather than having drifting or swimming larvae. The young remain with the female until they are large enough to survive independently. Very little is known of the growth rates or longevity of the species.

The body of the animal is similar to other snails but it possesses a prominent snout and short eye stalks on the outside of the tentacles. The radula is shaped like a rake and is used to scrape soft organic matter from surfaces. As a filter feeder the river snail feeds on bacteria suspended in the water and also grazes on the bacterial 'biofilms' that occur on hard surfaces in free flowing waters. They can function both as grazers, consuming algae growing on any submerged surface, and detritivores, utilising fine particulate organic matter and the bacteria and other microorganisms therein. They also filter feed on suspended matter.



Hybridisation is the interbreeding of individuals from two populations, or groups of populations, which are distinguishable on the basis of one or more heritable characters. When two populations of distinct but closely related rainbowfishes come into contact, members of those populations may spawn with each other and successfully reproduce.

The process of hybridisation can create problems for taxonomists, but it can also be a sign of the continuous nature of the process of speciation — the evolutionary formation of new species. If hybrids are formed between two populations that are barely differentiated, they may remain undetected, since their features may fall within the range of variability of one or both of the populations. One would expect, if those populations were to remain in contact, that they would blend together and lose their distinctness. On the other hand, two populations may each have diverged so far from their common ancestor that their individuals no longer recognise each other as potential mates. In that case, biologists are agreed that the two populations should be considered separate species. They will not fuse back into a single species.

It is between those two extremes of complete blending and total distinctness that hybridisation can provide glimpses of the complex process of differentiation — of evolution in action. Speciation normally occurs in geographic isolation, but the past distribution of rainbowfishes is complicated. Populations once isolated may come into contact, and when they do, the amount, duration, and results of hybridisation will vary from instance to instance.

It has long been thought that streams straddling the Great Dividing Range, which extends down the east coast of Australia, have captured drainages on the opposite side. Captures of western flowing streams by those flowing east and vice versa have been variously debated over the last fifty years or more. For example, it has been suggested that the Barron and the Burdekin Rivers in northern Queensland captured previously western flowing streams. The Clarence River area of north-eastern New South Wales has also characteristics which suggest that drainage rearrangements may have occurred, although the precise nature and timing of these events is still uncertain. The Fitzroy/Dawson system is believed to have drained a coastal area spanning latitudes 20°S to 23°S. Presently, much of the coastal area near the mouth of the Fitzroy River is drained by short coastal streams, although the Fitzroy River drains a large inland area.

Climate has also had a significant affect on the landscape in Australia, particularly freshwater environments. Rainfall in Australia is seasonal and also very variable, with drought in some years and floods in others. Ephemeral floodwaters allow dispersal of species and populations, both within and between river systems. Floodwaters from Queensland can end up in South Australia, covering a distance of 3000 kilometres. In the distant past Australia experienced very wet and humid conditions. Much of the inland was inundated by large freshwater lakes. The climate then developed more extreme and frequent periods of aridity. Much of the coastal regions inhabited by rainbowfishes probably experienced alternating cycles of wet and dry conditions. The outcome for river drainages was periods of drying and isolation followed by extensive flooding and connectivity.

These climate changes and related environmental changes have probably shaped the distribution of rainbowfishes. During the extreme wet conditions it is plausible that rainbowfishes were able to disperse across these river drainage boundaries. In contrast, the drier conditions may have prevented dispersal particularly where the effects of acidification were the strongest. These periodic connections and disconnections among catchments are predicted to have facilitated expansion and subsequent isolation of rainbowfish populations in new habitats.

There are numerous reports in the aquarium hobby that rainbowfishes which share the same habitat in the wild are quite capable of producing fertile hybrids in aquaria, but because of behavioural or colour differences they generally won't hybridise in nature. Kin recognition in many fishes has been demonstrated by laboratory experimentation. For example, Arnold (2000) examined shoaling behaviour in rainbowfish (*Melanotaenia eachamensis*) and found that females preferentially spend time associating with relatives when in an all female shoal, but avoided male relatives in a mixed shoal. The former result is consistent with the expectation of kin-biased behaviour, whereas the latter is suggestive of an innate tendency towards inbreeding avoidance.

However, hybridisation among rainbowfishes in their natural environment has traditionally been viewed as an unusual event. Allen & Cross (1982) previously recorded only two hybrids between *Chilatherina campsi* and *Melanotaenia affinis* in their study of rainbowfish taxonomy covering all species known at that time. However, hybrids are not easy to detect. They are much easier to identify and are generally recorded more often if the hybridising species are distinctively coloured. Recent genetic studies have shown that hybridisation between rainbowfishes in their natural environment occurs more commonly than originally believed.

Zhu et al. (1994) and McGuigan et al. (2000) tentatively identified hybrids using mtDNA data for two *Melanotaenia australis* populations in the Northern Territory (Blackmore and South Alligator Rivers). Further evidence for hybridisation was found in Lake Tinaroo between *Melanotaenia eachamensis* and *Melanotaenia splendida* (Zhu et al., 1998), and other populations were found containing a mix of mtDNA genotypes and morphologies (Pusey et al., 1997; Zhu et al., 1998; McGuigan, 2001).





There is also some evidence that natural hybridisation has occurred between *Melanotaenia splendida tatei* / *Melanotaenia fluviatilis*; *Melanotaenia duboulayi* / *Melanotaenia splendida splendida*; *Melanotaenia nigrans* / *Melanotaenia australis*; *Melanotaenia australis* / *Melanotaenia exquisita*; and *Melanotaenia exquisita* / *Melanotaenia splendida inornata*. However, it remains unclear whether these hybridisations represent sympatry, ongoing hybridisation, or historical introgression.

Based on mtDNA, nDNA and allozyme data it would appear that hybridisation and introgression has been common and has involved nearly all Australian rainbowfishes. Studies have shown that at least ten rainbowfish species in Australia have been involved in some degree of introgressive hybridisation. In some cases this was between sympatric species, but in others it occurred at the boundaries between more closely related species (P. J. Unmack 2005, *pers. comm.*).

The "Eachamensis" complex is north Queensland is really confusing, and even the "experts" can't seem to agree. One study reported that while *Melanotaenia eachamensis* and *Melanotaenia splendida* were shown to be genetically, meristically and morphologically distinct, many specimens in the study exhibited an intermediate set of characters suggesting that hybridisation between these species may be common place if intermediate body morphologies are indicative of hybrids. Despite the research that has been undertaken to date, the specific status and distribution of *Melanotaenia eachamensis* and *Melanotaenia splendida* still remains unclear.

Four rainbowfish populations were sampled from the Fitzroy River in Queensland. Two populations from the upper reaches of the Comet and Dawson river tributaries, and two populations from the lower portion of the Fitzroy River drainage from the upper Conner River and a lowland tributary of the Fitzroy River were sampled. Based on allozymes and mtDNA, the lower two populations were consistent with *Melanotaenia splendida*. The populations from the upper reaches were both consistent with *Melanotaenia fluviatilis*, with some *Melanotaenia splendida* alleles based on allozyme data, although they did have *Melanotaenia duboulayi* mtDNA. However this mtDNA type is common in *Melanotaenia fluviatilis* populations in the northern Murray-Darling catchment, the most likely source of these populations.

Each of these populations represents extremes in terms of separation by river distance within this drainage. At some point(s) within this system both species likely came into contact. No information currently exists relative to where this contact is likely to be! (P. J. Unmack *pers. comm.*).





Considering all the reported hybridisation events known so far between rainbowfishes in the wild, hybridisation might also have played an important role in the success of the spread of rainbowfishes across Australia and New Guinea.

There are several possible evolutionary consequences of hybridisation. Hybridisation may occur due to human impact, such as between wild and translocated species. Hybridisation due to human disturbances can compromise the genetic integrity of existing species to the point of causing extinctions. In extreme cases, parental taxa may be lost in the process and/or new taxa formed. A third possibility is that a stable hybrid species will form, with limited introgression. Introgressive hybridisation among taxa is known to quickly increase levels of variation, allowing more rapid responses to environmental changes.

Although, for the most part, the different species of rainbowfishes can be distinguished from one another, there is the possibility that many species may only be examples of population variation within a single species. There is much argument and discussion amongst biologists as to what a species actually is. The classic definition of a "species" is related organisms that share common characteristics and are capable of interbreeding. For a long time, biologists have almost universally used the biological species concept. This definition of "species" is based on species being reproductively isolated from each other. Reproductive isolation is the failure of populations to interbreed or to form viable or fertile hybrids.

Some years ago a controlled breeding trial demonstrated that *Melanotaenia fluviatilis* and *Melanotaenia duboulayi* could interbreed and produce viable offspring. Therefore, should these fish be considered different species or just different subspecies? Subspecies are simply populations within a species that are sufficiently distinct that taxonomists have found it convenient to formally name them, but not distinct enough to prevent hybridisation where two populations come into contact.

Under the biological species concept, distinctive geographical forms of the same sort of fish are usually grouped together as one species. This is because the geographic forms interbreed (or probably would, if they had the chance). Thus, they should be considered the same species. The phylogenetic species concept says that diagnosable geographic forms of the same sort of fish should be treated as distinct species. This is because these forms have evolved separately, and have unique evolutionary histories.

Obviously, the phylogenetic species concept is less restrictive than the biological species concept. There would be many more species of fish under the phylogenetic species concept than under the biological species concept.

These complications are a natural result of applying a hierarchical taxonomic system to the results of a continuous evolutionary process. It is possible that neither definition can be applied consistently in nature.

Fish of one species are, under most circumstances, incapable of interbreeding with individuals of other species. Indeed, the "biological species concept" centres on this inability to successfully hybridise, and is what most biologists mean by "distinctly different". However, the debate over how species should be defined will continue.

Most rainbowfishes show distinct geographic variation both in colouration and characteristics. This is inevitable among populations of any species with extensive distributions. It is largely the result of populations responding to different pressures of natural selection in different habitats. If populations of a single species become geographically isolated, those different selection pressures may, given enough time, cause the populations to differentiate sufficiently to prevent interbreeding if contact is re-established.

In nature, degrees of differentiation and of abilities to hybridise fall along a continuum, so one finds what is expected in an evolving fish fauna - some populations intermediate between subspecies and species, populations that have differentiated to the point where they will not hybridise but have not yet regained full contact, and populations so distinct that they can be recognised as full species whether or not they occur together.

Because of this great variation in colours and body forms in many species of rainbowfishes, especially in Australia, all rainbowfishes should be bred within their own localised group. Regardless of their various colour patterns, they are capable and willing to breed together if permitted to do so. The serious hobbyist intent on maintaining pure lines must keep every variety in separate aquariums. Unless this is done, members of the different varieties will interbreed and complicate future breeding programs and identification. Also, females of many rainbowfish species are very similar and can easily be confused for one another.

I kept rainbowfishes for more than 30 years and always kept populations, even if they went by the same species name, separated for breeding. History has shown that this is the prudent thing to do. In the 1970s and 80s, many populations of rainbowfishes found their way into the aquarium hobby. As time went on and the various populations were more carefully studied, we learned that several of these populations were, in reality, separate species.

Most experienced rainbowfish keepers will not bring new fish into their breeding program unless the breeder/collector has retained the location details. These hobbyists, for example, would not obtain a species with the name given only as *Melanotaenia trifasciata*, because the buyer does not know what he/she is getting. We may learn in the future that the different populations have some significant genetic or morphological differences that justify their recognition as a new species. Breeding such fish would have diminished the long-term viability and integrity of the species. Therefore, it is important to include a location name, such as Wonga Creek, Goyder River, etc. If the location name is lost, the fish should be distributed as an "aquarium strain".



Before being used for different rainbowfish species, spawning mops should always be boiled to destroy any eggs, which may still be attached to the mops. This procedure effectively precludes inadvertent hybridisation resulting from accidental carryover of eggs from the breeding set-up of one species to another, and demonstrates a significant advantage of using artificial spawning medium rather than living plants.

Most rainbowfish hybrids that hobbyists will come in contact with will just be unintended crossings. Although there are a number of "commercial" hybrids available, they are usually sold and distributed under trade names such as "Red Boesemani" etc. However, there are some hybrids



being sold as true species. "M. marcii", "M. hammeri" and "M. greetii" are such examples of commercial hybrids. These three hybrids were originally bred by a commercial aquarium dealer and breeder in the Netherlands. These hybrids, particularly "marcii", often sold as the Marci Rainbowfish, have been widely distributed in Europe and North America.





"Inbreeding is deleterious because it increases overall genetic homozygosity, and thus the expression of recessive deleterious mutations in offspring." (Arnold, 2000)

Although not generally well know, Australian rainbowfishes have been maintained in home aquaria at least since the beginning of the last century. On the other hand, New Guinea rainbowfishes have only been available since the mid 1950's. They were being maintained by only a handful of enthusiasts and were virtually unknown to the international hobby. During the 1960's and 70's a small trickle continued to arrive in Australia from New Guinea. The importation of New Guinea rainbowfishes into Australia during this period did not have any significant restrictions and a number of different species were brought into the country by private collectors, which were subsequently distributed in the hobby. However, the publication in 1982 of Rainbowfishes of Australia and Papua New Guinea by Gerald Allen and Norbert Cross, greatly increased the popularity of keeping rainbowfishes and the desire for the newly discovered New Guinea species, turned that trickle into a flood.

Unfortunately, the initial number of wild-caught fishes that came in to the hobby was very small. This resulted in a very small genetic base from which to establish larger aquarium populations. The problem with having such a small genetic base is that most of the aquarium populations are closely related. Since the initial population was small, they are likely to have a higher prevalence of recessive genetic disorders, as the parents are likely to share many genes. In addition, a small population base may not be truly representative of the original wild population.

Another problem with rainbowfishes kept in captivity is that instead of natural selection, selection is done by the aquarist; because only a relatively small number of fish can be kept, the aquarist tends to select for those which grow best and look best under aquarium conditions. In the long term, the fish being kept may be genetically a long way from the original wild fish. For the most part, aquarium breeding has effectively resulted in domesticated strains of rainbowfishes that may be well adapted to life in captivity, but may look very different than the species in the wild.

This can be clearly seen in a number of New Guinea species where the quality and colouration has diminished greatly, as well as increases in breeding failure. This also applies to Australian species collected from remote locations where the initial numbers of wild-caught specimens was low.

Almost every day I see photos of rainbowfishes posted on Internet forums asking for identification. Most replies given are in the negative, suggesting that they are hybrids. This may well be true; however, what is happening for the most part is that continuous inbreeding or selective breeding has effectively resulted in domesticated strains of rainbowfishes that no longer resemble their congeners from their natural state. When fish are removed from the natural environment and placed in the aquarium environment different selective forces act upon fish in the aquarium environment compared with the natural environment and reduces their genetic variability through both selective processes and random genetic drift. Inbreeding depression and reduced levels of genetic diversity are expected to be more prevalent in small populations where breeding between close relatives is more likely and the effects of genetic drift more pronounced. Both inbreeding depression and the impacts of reduced genetic diversity have a sound basis in theory and have been well documented by animal and plant breeders over many decades. There are many studies that describe the deleterious effects of inbreeding in fish, generally in captive conditions or in association with artificial selection.

The limited gene pool caused by continued inbreeding means that deleterious recessive genes inherited from both parents become homozygous. The condition can manifest as reduced fertility, growth and disease resistance, lower hatching rates and survival, behavioural changes and high occurrence of abnormalities to name but a few. Selective breeding can produce similar effects.

Selective breeding is a breeding technique in which the breeder chooses the next generation's broodstock, based on some predetermined criteria. The process by which crosses are accomplished between the parental stocks representing different strains of the same species is referred to as crossbreeding, and can also be called intraspecific hybridisation. Crossbreeding can produce strains of superior performance by introducing greater genetic variability.

In wild populations, outbreeding may also result in a reduction of fitness, because populations can become adapted to living in particular areas with a particular climate, diseases, and so forth. If individuals from other populations interbreed with the adapted population, new alleles are introduced. These alleles may not be as well adapted to the local conditions and may reduce the fitness of the population. For example, two populations of fish may have evolved a particular colour pattern that is advantageous in the environments in which they live. If the two populations were to interbreed, they may produce an intermediate form of the pattern that is not advantageous in either of their environments, reducing the fitness of the overall population (Attiwill & Wilson 2003). Natural selection (evolution) causes changes in wild populations over many generations. This often results in many sub-species originating from the founder population. Domestication enhances and speeds up the process via artificial selection dictated by rapid change.

For the most part, aquarists maintain small populations of rainbowfish species. When a new species is obtained, the number of fish acquired is usually low. In many cases the fish that are acquired come from one or two spawnings, or more often, offspring that have been produced by a single



spawning. In these situations, it is more likely that related individuals will breed, simply due to the lack of alternative mates. Breeding closely related rainbowfishes in captivity increases the likelihood of the population suffering from inbreeding problems. In aquarium populations the effect of inbreeding may be severe and should be a concern to breeders. While inbreeding can be used to improve a population when it is planned and directed, unplanned and uncontrolled inbreeding can ruin a population through a process known as 'homozygosity' (inbreeding depression), i.e., less variation in genes. Greater homozygosity may be desirable in some cases, such as line-breeding, where an outstanding individual is mated with a descendant to increase that individual's characteristics (e.g., better colour, size, etc.) to the gene pool.

Tave (1986) reported that inbreeding populations can still produce good offspring even though inbreeding depression occurs. The depression has been found from various genotype and population means, but outstanding individuals are still produced. These outstanding specimens could be kept for selective breeding purposes because these individuals contain more desirable alleles and are free of degenerative alleles.

Inbreeding depression is probably the most serious consequence of small population size. Expression of a trait is determined at the gene level by information contributed by each parent, and a predictable percentage of offspring will display these traits. If one parent's gene is recessive, then the trait it codes for will be expressed by a predictably small number of the offspring. Others will possess the gene, but won't express it. The population is said to be heterozygous. However, in any strategy involving inbreeding it is necessary to take effective steps to insure against excessive fixation of deleterious alleles.

The problem with breeding related individuals is that over time you remove the heterozygosity from the population and create a population homozygous at all genes (i.e., both genes code for the same trait expression). This can increase the occurrence of traits which are detrimental to a species' fecundity, disease resistance, fertility, and growth. Inbreeding depression resulting from increased homozygosity is well documented in fish. The majority of inbreeding experiments on fish have been done in aquaculture and laboratory-type environments.

While the actual inbreeding depression varies widely between fish species and inbreeding levels, significant levels of inbreeding depression have been found in many aquacultured brood stocks after only one generation of brother-sister mating. Therefore, the high level of abnormal fish especially spinal malformation appearing in hatchery fish is a major problem in many aquaculture farms. Malformation often is associated with growth depression, leading to high mortality rates at early fry stage.

Inbreeding, along with selection and cross-breeding has been traditionally used to create new varieties and colour forms in many aquarium fish species. With rainbowfishes, however, I would hope that most enthusiasts are trying to breed a species as close as possible to its wild form. Unintentional domestication of rainbowfishes may be unavoidable, but it is possible that it can be minimised by the introduction of new broodstock. It is possible for rainbowfishes derived from the same source to have different genes when separated for any length of time. It's very important to properly select the breeding stock (related or not) and also to properly cull the fry for obvious defects.

These days most rainbowfishes are either obtained from commercial sources or bred by individual hobbyists using a limited number of broodstock fish. In both cases the genetic background and the degree of inbreeding of the fish is generally unknown. Aquarists must give careful consideration to the choice of brood stock if genetic 'pollution' of the aquarium stock is to be prevented. To avoid genetic problems it is best to start with as many fish as possible (minimum of five pairs) or get your fish from at least two different sources, or at different times. If you get all the fish from the same source or at the same time, there is a good chance that the fish will be related, especially if they have been bred in captivity.

Diversifying your sources for the fish and expanding their genetic base will help enhance their genetic variability, and may reduce problems resulting from inbreeding depression. In this strategy, individuals from another population (hopefully with greater genetic variability) are introduced to your population in an attempt to recover genetic diversity and reduce inbreeding. Breed every fish in the group, using random selection to determine pairing. Separate the fry from each pairing and select 4-6 fry from each spawn for breeding the next generation. With problem species it is advisable to raise the fry from each spawn separately until they sex out, then select a male and a female from each for breeding. In small populations it takes as little a one individual per generation to maintain genetic diversity. Ideally, a number of individuals should be sourced from the wild population every so often.

It is apparent that numerous New Guinea rainbowfish species may be in danger of being lost both in the wild and in captivity. There are only very small populations of some species now kept in captivity. A number of the early New Guinea species have all but disappeared from the hobby (I think some have disappeared). Therefore, saving the existing aquarium species and the integrity of each individual species becomes vital as there may in the future be no wild fish to restore captive stocks or genetic variability.

To maximise genetic diversity captive populations need to be perpetually managed so an adequate number of separate brood stocks are maintained with occasional intercrosses between them to reduce the probability of fixing deleterious genes. Zoos engaged in captive breeding programs are aware of this need to outcross their own stock to animals from other collections. Captive populations are at risk from inbreeding since relatively few mates are available to the animals, hence zoos must borrow animals from each other in order to maintain the genetic diversity of offspring. A practical solution for this problem is for rainbowfish enthusiasts to collaboratively maintain a much larger gene pool collectively, than they would as individuals.



A successful breeding program, in essence, depends on the successful manipulation of inbreeding. A well designed long term breeding program should be directed at preserving the basic genetic diversity of the population, and must be based on detailed information about the species characteristics and the origin and genetic history of the population. The main strategy for reducing inbreeding is to maintain a large population of broodstock fish, and ensure that a large proportion of them get a chance to breed and contribute to the next generation. At least a few fry from all of these broodstock should be retained and grown up for use as the next generation of broodstock, before the previous generation gets too old and is discarded.

This sometimes requires a lot of small tanks or raising tubs, and good record keeping to record breeding results, both successes and failures. Good record keeping is essential in solving problems with inbreeding and is paramount in improving survival rates. Records of fry growth and development should be made regularly. This activity is often carried out by the serious hobbyists.

In wild populations, inbreeding is avoided because individuals prevent themselves from mating with relatives. Evidence for this "behavioural avoidance of inbreeding" has been found in a wide variety of animals, including fish. A number of different species have demonstrated the ability to recognise and discriminate in favour of familiar conspecifics in laboratory trials. Further research has found that this preference for certain individuals may persist for relatively long periods, even after a 2-month period of separation. The duration of time over which it persists is likely to vary between species.

The ability to recognise related kin is based on visual and/or chemical cues. In other words, individuals are less likely to interact sexually with others with whom they were intimately familiar during development, namely siblings and parents. Females were found to avoid breeding with siblings and parents more actively than males. This may be why we have the occasional breeding failures with some species of rainbowfishes we maintain in our aquariums. Maybe it might just be that the females are reluctant to breed with their siblings. This could also explain male aggression toward uncooperative (possibly related) females.

One study (Arnold, 2000) found females rainbowfishes were able to differentiate between levels of relatedness at a relatively fine scale. Full-siblings could be distinguished from halfsiblings, and half-siblings from non-relatives. In terms of shoaling preferences, the sex of shoal-mates was also important. Females always preferred to associate with those individuals of the same sex with which they were most closely related. In contrast, when given the choice of associating with males of different levels of relatedness, females always spent significantly longer with non-relatives than with full- or halfbrothers. Thus, females appeared to avoid associating with potential mates that were close relatives.

However, other researchers have noted that females can still reduce the probability of inbreeding by seeking multiple mates, regardless of the ability to recognize kin, because by so doing they increase their chance of producing at least some outbred young. This would be especially important in situations where females cannot avoid breeding with close relatives as may be the case in captivity. The group spawning of rainbowfish species can also decrease the harmful effect of inbreeding due to the intensive mixing of gametes and high number of progenies.

In another study (Gleeson *et al.*, 2000) two species of rainbowfishes from three locations in Australia were experimentally infected with the parasite *lchthyophthirius multifiliis*. One of the species (*M. eachamensis*) was much more susceptible to the parasite than the other species (*M. splendida*). *M. splendida* served as a control for a follow-up hybridisation experiment which involved crossing *M. eachamensis* from the original population with another population of the same species located some distance away. The population hybrids had significantly higher resistance than the single-population fish. It was tentatively suggested that there may be a link between the heterozygosity of populations of rainbowfish and their initial ability to resist infection by *lchthyophthirius multifiliis*. Therefore inbreeding rainbowfishes in captivity may reduce their natural disease resistance.

A broad genetic basis has been suggested as a key element for parasite resistance, with heterozygous individuals assumed to detect and present a wider range of pathogen-derived antigens due to a larger number of different major histocompatability complex (MHC) molecules. MHC plays an important role in the immune system, autoimmunity, and reproductive success. Other studies have reported similar results and have found the most homozygous populations display greater infection and mortality compared to heterozygotes. Further, inbred populations showed lower survival and higher infection compared to outbred populations, leaving the authors to conclude that low genetic variation in general, or for the important MHC genes, and populations with a history of inbreeding are more likely to suffer detrimental effects from parasitic infection.

#### Visible symptoms of inbreeding:

- Deformed fins.
- Absence of abdominal fins.
- Thinning and deformation of hard rays of fins.
- Deformed gill cover operculum.
- Skeletal deformities.

DNA technologies have revolutionised the study of phylogeny (study of the pattern of ancestry and descent among species, populations, genes or alleles) and species-level questions that have application in wildlife management. Recently, these techniques were used to distinguish between endemic and introduced populations of the freshwater turtle (*Emydura macquarii*) in coastal NSW. A similar story emerged in the case of the rainbowfish (*Melanotaenia eachamensis*), once thought to be a rare species endemic to Lake Eacham, but where DNA sequence variation revealed a more complex relationship between it and the more regionally distributed *Melanotaenia splendida* (Zhu *et al.* 1998).



# Deformities

Deformities in rainbowfishes are occasionally found in wild population. However, such abnormalities are quite common among rainbowfishes kept in captivity. The cause of the deformities is often unknown, but often results from a wide range of causes, including genetic variance (hereditary factors), inbreeding depression, significant environmental changes, namely temperature, pH, disease, nutritional deficiencies, injury and environmental contamination. Inbreeding can also elicit such abnormalities in fish species but in the absence of sound evidence no single specific reason for deformities can be established. However, it is probably that most deformities are caused by inbreeding within a small gene pool.

Available evidence suggests that deformities in aquarium bred rainbowfishes usually develop very early during the larval/ juvenile stages. The most common deformities observed in rainbowfishes include spinal malformations (lordosis, scoliosis, coiled vertebral column), deformed operculum and fin malformations. Spinal malformations increased steadily with growth. I believe that most deformities in rainbowfishes we see today are the results of inbreeding. Genetic deformities become more likely with inbreeding over a span of several generations.

Deformities as a result of inbreeding have been observed in many hatchery reared fishes in aquaculture (Tave, 1986). The same trend has been observed in limited studies of aquarium fishes. There are cases of reproductive failure, growth reduction, bodily deformities, and behavioural changes in *Amatitlania nigrofasciata*, *Brachydanio rerio*, *Poecilia reticulata*, *Carassius auratus* and a number of other species due to inbreeding depression. In some species such as *Poecilia reticulata*, which have been exposed to sustained selective breeding for more than fifty years, a great many deleterious alleles have been eliminated. The rainbowfish breeder may like to establish 'pure lines' also, but in practice this is much more difficult to achieve.

The inbreeding depression observed in the convict cichlid (Amatitlania nigrofasciata) is comparable to the results seen from inbreeding rainbowfishes. Amatitlania nigrofasciata appears to be unable to survive extensive laboratory inbreeding without deleterious genetic effects (Winemiller & Taylor, 1982). No major deleterious inbreeding effects were noted until the F4 and F5 generations. Of the surviving (5 months) fry from nine F4 broods, 26.4% were moderately deformed and 58.1% were severely deformed. Moderately deformed fish exhibited abnormal fins (often shortened with an absence of dorsal spines), a pronounced vertical slope of the forehead, shortened, flared opercula (exposing the gills at all times) and a permanently depressed hyoid apparatus. The position of the spinous dorsal fin frequently deviated laterally from the dorsal midline of deformed fish. Severely deformed fish were characterised by the same abnormalities, but in a more pronounced form.



Severely deformed fish frequently exhibited moderate lordosis and abnormal swimming behaviour, wherein the head remained lowered and the lateral undulations of the body appeared grossly exaggerated. Of the surviving F5 fry (five broods), 17.6% were moderately deformed and 65.9% showed severe deformities. All deformed fish appeared to develop normally until the end of their most rapid growth phase (2–3 months). The variety of morphological and behavioural abnormalities observed in F4 and F5 inbred generations indicate that a number of genetic factors may act singularly or together.

The Zebra Danio, *Brachydanio rerio* inbred for four generations, exhibited vertebral abnormalities, a lack of swim bladders, protruding jaws, opercular deformities, oedema and behavioural irregularities (Piron, 1978). Larval deformity occurred in a study (Badger, 2004) of *Melanotaenia splendida* as well i.e., in the larvae hatched from eggs obtained from fish fed a 20% lipid content diet. It is surmised that the high lipid content in the diet was the likely cause of larval deformity as it was not seen in the larvae from the 12% or 9% lipid diet trials.

In one study, deformed operculum was the most common deformity (37.6%) observed in *Cyprinus carpio*. The deformities were first observed at two months of age and the degree of deformity was variable but in most cases operculum was usually shortened with involuted edges. In some fish it was minimally shortened, while others had an operculum that was so malformed that the posterior gill lamellae were exposed. The deformity usually occurred only on one side; however, some of the fish had a bilateral deformed operculum. Fish with this kind of deformity swam normally, but growth was very slow. In some studies it was reported that operculum deformity was found to be non-inheritable and was associated with ascorbic acid (Vitamin C) deficiency in the diet.

It is also reported that some deformities in rainbowfish larvae, particularly spinal deformities, are greatly affected by dietary ascorbic acid levels during early rearing. Feeding trials (Ako *et. al.*, 1999) using *Pseudomugil furcatus* found an unusual number of the fish had crooked spines at the end of the grow-out trials. The next batch of fry was fed brine shrimp enriched with a product containing ascorbic acid and the crooked spines largely disappeared. This points out the importance of early nutrition in fish rearing. Low doses (25–50 ppm) of ascorbic acid are reported to have a positive effect on growth and to reduce skeletal deformities. Duckweed can provide vital biopigments that are required for the development of improved colouration for captive bred rainbowfishes, and can also be a rich source of ascorbic acid (2.78–4.90 mg/100 grams dry weight).

Non-inflation of the swim bladder (belly-sliders) is another common problem often encounter in larval/juvenile stages of rainbowfishes. Swim bladder inflation is a fundamental developmental step during the early larval stage of most fish larvae. Initial inflation occurs when air, gulped by larvae from the surface, passes through the gut and is transferred via a pneumatic duct to the swim bladder lumen. Because the pneumatic duct exists for only a brief period of time in larval development before it regresses, inflation must occur during this narrow window of opportunity. Once the physical connection between the gut and swim bladder lumen has degenerated, swim bladder inflation cannot occur. This is one of several critical stages of larval development that may cause high mortality or swim bladder problems. Swim bladder inflation failure is common in intensively reared fish species. Noninflation of the swim bladder commonly appears to affect about 5% of rainbowfish larvae. The swim bladder of rainbowfishes usually inflates within the first 12 hours of hatching.

Although the exact causes of swim bladder non-inflation remain uncertain, factors such as genetics, nutritional status or water conditions, probably play an important role. The use of surface skimmers to prevent build-up of surface oils on the water has been found significant to optimise swim bladder inflation. In several fish species spinal malformation was found to be associated with the absence of a functional swim-bladder. Swim bladder problems can also be caused by bacterial infection.

#### Predisposing factors that can cause deformities:

- Disturbance of fertilised eggs during the first 12 hours of embryonic development.
- Elevated levels of dissolved gases (CO<sub>2</sub> etc.) during embryonic, larval and juvenile development.
- Accelerated water flows in the larval/juvenile tanks.
- Alterations in water quality.
- Fluctuations in water *p*H.
- Elevated levels of light intensity during larval development.
- Vitamin deficiency: There are numerous reports of deformities in aquarium fishes due to ascorbic acid and calcium deficiency. High levels of vitamin A have been observed to cause spinal effects in some species.
- Amino acid deficiency: Amino acids may be absent in foods due to improper formulation, extended storage or excess heat in processing.
- Hereditary: Mostly as a result of inbreeding most prominent in aquarium fish.
- Heavy metals (arsenic, copper, lead, mercury), have been recorded as causes of vertebral defects.
- Spinal deformities associated with organophosphate, organochlorine and carbamate pesticides.
- Antibiotic therapy: Spinal deformities and decreased growth rates have been frequently observed in fish fed with therapeutic doses of Oxytetracycline.
- Infectious disease (particularly of bacterial origin).

#### **Management and Prevention**

- If the proportion of deformed larvae exceeds 25–30%, reject the whole batch.
- Careful selection, nutrition and conditioning of brood-stock.
- Proper collection and incubation of fertilised eggs.
- Meticulous control and recording of water parameters and larval management.
- Avoid inbreeding by using two independent lines from the same fish species.



# Rainbowfishes Foods & Feeding



Photo: Gary Lange

Rainbowfishes are frequently described as meiophagous omnivores or opportunistic feeders, meaning that they will eat almost anything that is available. However, just because they will consume something does not necessarily mean that they will grow well on it. Rainbowfishes are found in a diverse range of habitats, therefore it is not surprising that this diversity is reflected in their feeding habits. The size and variety of their diet can differ widely from one species to another and from one habitat to another. In fact, quantitative data on the diets of most species of rainbowfishes are limited.

In their natural habitat rainbowfishes consume nearly 100% live foods. They can be seen around sub-surface aquatic vegetation feeding on a variety of terrestrial insects, small aquatic crustaceans, insect larvae, worms, phytoplankton, zooplankton, aquatic plant material, detritus (inanimate suspended matter principally of organic origin), and where the opportunity occurs, smaller fish will also be consumed.

Aquatic insects and their larvae are the most important food resource for most rainbowfish species, with microcrustaceans, algae and food from terrestrial origin (mainly terrestrial insects) also providing important food sources. A review of published dietary information for rainbowfishes shows that aquatic insects contribute almost 35% of the total mean diet. Other important food items were microcrustaceans (14.4%), algae (8.5%), pollen, seeds and terrestrial insects (8%).

Food sources of allochthonous origin (e.g., terrestrial insects) form a significant component of the diet of rainbowfishes. Rainbowfish consume such items once they have entered the aquatic environment. Terrestrially derived food form an important alternative food source during times of low aquatic food availability. For example, aquatic food items may not be available all year round due to natural seasonal declines in these resources and in such instances rainbowfishes may switch to other food sources. In large floodplain rivers, terrestrial food items increase in importance as water floods on to the floodplains. Planktonic invertebrates (mostly zooplankton) are importance in their early life history stages. The availability of appropriate zooplankton is an important determinant of mortality levels endured by larval rainbowfish populations and thus is an important determinant of recruitment into the adult population.

Studies in the Alligator Rivers of the Northern Territory (Bishop *et al.*, 1981), bearing in mind that this is probably by far the most thoroughly surveyed aquatic environment in Australia, with years of accumulated research available, found rainbowfishes were omnivorous, feeding opportunistically across substrates and in surface waters, with possibly less emphasis on mid-water areas. Traces of hydrophytes, oligochaetes, gastropods, arachnids, crustaceans, fish (larvae or juvenile), terrestrial plants, detritus and inorganic material were found in the stomach of rainbowfishes. Their diet was found to vary in relation to the habitat they occupy.

In the mainchannel waterbodies they eat mainly aquatic insects, with small amounts of terrestrial insects, plant material and algae. In perennial streams, algae and terrestrial plant material are less important, while aquatic insects and, to a lesser extent, oligochaetes and microcrustaceans, are consumed. The diet in the lowland sandy creekbeds had much larger algal and terrestrial insect components. Specimens examined from the floodplains feed mainly on aquatic arachnids and aquatic insects, and a small amount of algae. In the pools and riffles that enter the floodplain in the wet season they feed mainly on non-aquatic insect forms such as winged diptera and ants. A variety of aquatic insects are eaten; the main identifiable species being chironomid larvae and pupae, and coleopterans. The main terrestrial insects were formicids (ants), while the main microcrustaceans were cladocerans, ostracods and conchostracans. The algal component consists mainly of green filamentous species.

One study (Rayner, 2006) on the diet of *Pseudomugil signifer* and *Melanotaenia splendida* in the Mulgrave River in north Queensland found these species specialised on a combination of aerial and surface insects (28–92%), particularly emerging chironomid nymphs, and terrestrial invertebrates (up to 43%), principally green ants (*Oecophylla smaragdina*), which had presumably fallen on to the water surface from overhanging vegetation.

In another study (Balcombe *et al.*, 2005) it was found that terrestrial fauna was a major food group consumed by *Melanotaenia splendida tatei*. Their diet consisted of terrestrial insects (67.4%), other terrestrial invertebrates (10.2%), algae (16.8%), and aquatic insects (5.6%). In addition to the consumption of some aquatic insects and algae, this species fed chiefly upon terrestrial arthropods, many of which were flying insects (e.g., ants, wasps and dipterans). Other items included aquatic dipterans, coleopteran larvae and zooplankton. The aquatic dipterans were mostly chironomid larvae, while the zooplankton prey consisted chiefly of conchostracans and cladocerans. Terrestrial foods included isopods, scolopendridid centipedes, or a variety of alighting insects such as dipterans, hymenopterans and coleopterans. Up to 100% terrestrial insects can be consumed during the dry season.

Other studies have shown that rainbowfishes have a preference for foods such as cladocerans, ostracods, copepods, rotifers and other invertebrates. The stomach contents of specimens collected from the Noosa River in Queensland consisted of copepods, cladocerans and shrimp (*Caridina sp.*) and a variety of aquatic insects (especially chironomid larvae). A small percentage of algal material and other plant tissues plus a few terrestrial insects were also found. However, terrestrial sources of food were relatively unimportant in their diet in this study. The natural diet of *Rhadinocentrus ornatus* was reported to consist mainly of terrestrial insects. They feed on a variety of terrestrial and aquatic insects, insect larvae, and small aquatic crustaceans. Algae and pollen (mostly pollen) also appear to be a major alternative food.



The natural diet of Pseudomugil tenellus includes algae, microcrustaceans and aquatic insects. The identifiable algae were green filamentous and blue-green algae and dinoflagellates. The microcrustaceans were mainly cladocerans, ostracods and copepods. Chironomid larvae were the main aquatic insects eaten. Other food items found in the stomachs were terrestrial insects and miscellaneous organic matter. One study found that the diet in the late-dry season was mainly based on detritus (with associated unidentified organic material) and small quantities of chironomid larvae and pupae, and algae; no microcrustaceans were eaten. In the early-wet season microcrustaceans appeared in the diet and detritus decreased in importance; aquatic insects also appeared in the diet during this season. In the mid-wet season *Pseudomugil tenellus* ate mainly microcrustaceans (particularly cladocerans) with smaller amounts of terrestrial and aquatic insects. By the late-wetearly-dry season algae were the main component of the diet.

*Melanotaenia fluviatilis* consume high numbers of cladocerans throughout the year and copepods were consumed in high numbers in all seasons except winter. Cladoceran eggs were recorded in high numbers in spring and summer, and were likely to have been detached from adult cladocerans in the digestive tract of the fish, rather than having been consumed as individual food items. The highest numbers of ostracods and dipteran larvae were consumed during spring, while algae were only consumed in summer and autumn. Fish eggs were recorded in the stomach contents of some fish during spring and summer.

Other diet categories were either recorded in low numbers or abundance counts did not apply (such as detritus and plant material). In another study large numbers of copepods were consumed in spring and summer. Algae were recorded in high numbers in summer only. Dipteran larvae were recorded in stomach contents in spring and autumn, whilst ostracods were recorded in summer and winter. Zooplankton was recorded in the stomach of fish from summer and autumn. Unidentified material (organic and inorganic detritus) constituted a major proportion of the diet in both studies.

Nevertheless, dietary analysis can reveal contradictory information on the food resources available to rainbowfishes, particularly across the seasons - it really depends on the season and the type of habitat!

Nutrients in freshwater rivers are mostly derived from sources outside the stream itself, mainly in the form of organic material derived from the riparian vegetation. Some primary production occurs from algae and in-stream vegetation, although this is comparatively small, with upland streams too shaded or too cold, and lowland streams too turbid for effective photosynthesis.

Leaves falling into the stream are eaten directly by invertebrates known as "shredders", such as stonefly and caddisfly larvae, which shred or chew the softer parts of the plant material. Material not consumed by the shredders is colonised by microorganisms such as aquatic fungi and bacteria and broken down to progressively smaller sizes. Freshwater algae also colonise the leaves and twigs. Invertebrates known as "scrapers", such as some mayflies and snails feed directly on the fungi and algae. As the organic material is broken down, the resultant finer material then serves as a food source for other invertebrates, which filter (filterfeeders) material from the water, or collect deposited material on the stream bed (detritus feeders). These animals, in turn are preyed upon by other invertebrates and animals such as rainbowfishes. Rainbowfishes are then a food source for water birds and other fish-eating animals. Without the initial inputs of organic material or invertebrates, the entire food chain of the stream system is compromised. Hence, the importance of riparian vegetation to stream health cannot be underestimated.

Available evidence suggests that feeding rates for rainbowfish species are higher in spring and summer than in winter, possibly because of higher metabolic demands during the warmer months. This time also represents an important period for larval development and growth of juveniles. The season of greatest feeding activity in tropical rivers is the wet season. Feeding activity increases most dramatically between the late-dry season and the early-wet season. By the mid-wet season feeding activity has peaked, and then decreases slightly by the late-wet to early-dry season. An examination of variations in body condition indicated that most species obtained their best condition from the mid-wet to the mid-dry season, with a peak in the late-wet-earlydry season. During the dry season, when the water volume of their habitat is greatly reduced, the only food they get is whatever happens to fall into the water.

Adult fish can survive for weeks or even months on very little food, but without a plentiful food supply they will not flourish. If food is in short supply, growth will be reduced or nonexistent and fish may lose weight and become more susceptible to disease. During spawning egg numbers will be fewer. Juveniles and larvae in contrast are much more dependent upon a regular food supply and may starve within days or even hours.

Scientific data from stomach content analyses in wild populations can be helpful in selecting the right type of diet for your rainbowfishes. Few people, however, will be able to duplicate the natural combination of the many foods consumed by rainbowfishes in their natural environment. Therefore, it is necessary to provide favourable aquarium conditions and careful feeding with specially formulated diets in an attempt to satisfy all their nutritional requirements.

## Feeding & Reproduction

Rainbowfishes require a highly nutritious diet in order to maintain spawning condition and produce large numbers of eggs. Research studies have shown that during periods of low food availability, a decrease and then cessation of reproduction occurs. During these times rainbowfish must allocate resources to meet basic metabolic functions rather than reproduction. Research has also found that diet strongly affects not only fecundity but also the biochemical make-up of eggs and sperm as well as the growth rate and survival of larvae.



The breeding seasons of rainbowfishes must coincide with the conditions that offer the greatest amount of protection for the eggs, and food and shelter for the newly hatched larvae. The duration and timing of reproductive activity are thus two critical components of the rainbowfishes lifehistory strategy. Rainbowfishes are often aseasonal spawners, breeding continuously at intervals throughout the year. This strategy increases the chances of at least some eggs surviving during the year. Rainbowfish eggs are attached by adhesive threads or tendrils to aquatic plants and other objects in the water, which hide them from predators.

One study (Badger, 2004) covered a variety of topics on the nutrition of *Melanotaenia splendida* ranging from identifying what reproductive factors are affected by nutrition to how diet composition affected these factors. This study determined that egg number as opposed to egg diameter was affected by nutritional status, and that it is best to feed *M. splendida* daily to satiation in order to maximise reproduction. The best diet found in this study for maximising reproduction was one with a 43% protein and 12% lipid content, which was supplemented with essential fatty acids. The energy content in the diet effected egg number the most, while the protein content, lipid content, and fatty acid supplementation maximised unfed larval life.

Feeding trials found that rainbowfishes fed every day produced significantly more eggs then fish fed every second day. Average total number of eggs produced by fish fed every 2nd, 3rd, 4th and 5th day showed a clear trend of decline with increased feeding interval. It declined from 152 eggs in those fish fed every day, to 99 eggs in fish fed every 2nd day, 26 eggs for fish fed every 3rd day, 21 eggs for fish fed every 4th day and 19 eggs for fish fed every 5th day. The number of spawns was also affected by feeding, with the highest number of spawns occurring in fish fed daily. Hatching rate also declined from fish fed daily, to fish fed once every five days. All fish in the trials were fed to satiation on feeding days, with excess food siphoned out.

Egg quality also declined during periods of lower food availability. Eggs that had been laid after the onset of reduced feeding frequency or feeding ration had correspondingly lower fertilisation and hatching rates.

Lowered survival to eyed embryo may have been due to at least one of two factors:

- a. the males were affected and in so doing produced less sperm that was not as viable or with a lower motility rate and/or
- b. the viability of the eggs was affected, either the structure could have been effected making fertilisation difficult or impossible, or there were inadequate nutrients to support any embryonic development.

These two effects may have acted alone or in concert. The eggs that were fertilised also had a decrease in hatching success.

A very dramatic increase in reproductive success could be seen (Table 1) when comparing fish fed a 43% protein, 6% lipid diet modelled on the commercially available flake food (Nutrafin<sup>®</sup>) and fish fed a 43% protein, 12% lipid diet supplemented with the essential fatty acids arachidonic acid, docosahexaenoic acid and eicosapentaenoic acid. Egg number doubled increasing from  $121 \pm 11$  eggs to  $245 \pm 21$  eggs. The number of spawns significantly increased while average spawning interval decreased. There was no significant difference in survival to eyed embryo and hatching rate, however, fish fed the formulated diet achieved a 100% survival to eved embryo and hatching rate which is guite notable. The unfed larval life increased significantly as well, increasing by over two days from  $8.9 \pm 1.5$  to  $11.1 \pm 1.2$  days. The increase in egg number, unfed larval life and decrease in spawning interval achieved by replacing the control diet with the formulated diet showed that a very marked increase in reproductive success had been demonstrated between these two diets for Melanotaenia splendida.

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	Nutrafin	Formulated Diet
Egg Number	121 ± 11	245 ± 21
Total Spawns	19.3 ± 0.3	26.4 ± 0.8
Av. Spawning Interval (Days)	2.1 ± 0.1	1.5 ± 0.1
Survival to Eyed Embryo %	99.8 ± 1.2	100 ± 0.0
Hatching Rate %	98.6 ± 0.8	100 ± 0.1
Unfed Larval Life (Days)	8.9 ± 1.5	11.1 ± 1.2

Energy, protein and lipid contents in a diet are important. Whatever is consumed in feed is needed by the fish for a variety of metabolic functions, and must first be allocated to essential needs such as respiration, digestion and excretion. Any excess that is available after meeting these requirements can then be allocated to other processes such as growth and reproduction. It is therefore necessary to ensure that diets for rainbowfishes contain enough of these nutrients not only to meet essential metabolic functions but also to sustain egg and sperm development.

Both protein and lipid are very important constituents of fish eggs. They are the primary components of vitellogenin, the lipoprotein that is the main energy source in the yolk of eggs. They are also an energy source, therefore it is important to ensure that fish feeds contain enough of each so that female rainbowfish will have enough lipoproteins to allocate to egg production. Many studies have previously linked increased protein level with increased reproductive ability. It has also been shown that while lipid is important to fish reproduction, too much lipid in a diet can be detrimental to egg quality. Too little lipid or poor quality, however, can lead to a decrease in ovary size and lower egg survival to hatching.



Furthermore, too much lipid in a diet can cause deformities in eggs and reduced hatching success. Larval deformity occurred in the study of *Melanotaenia splendida* as well, i. e., in the larvae hatched from eggs obtained from fish fed a 20% lipid content diet. It is surmised that the high lipid content in the diet was the likely cause of larval deformity as it was not seen in the larvae from the 12% or 9% lipid diet trials. When fed a high lipid diet, rainbowfishes may develop lipidosis (fatty degeneration of the internal organs) and eventually die. A diet containing 43% protein and 12% lipid was deemed to be the more ideal diet. However, in outdoor ponds, natural foods contribute to the total amount of protein and energy available to fish, so results might differ from aquarium studies.

The total dietary protein requirement may be defined as the minimum amount of protein that produces best fish performance (e.g., growth, feed conversion) under a given set of conditions. In addition to total protein, the balance between protein and energy in a diet is critical. When more protein is added to a diet than is needed for growth and other bodily functions, the excess will be metabolised for energy or used to make energy-storage products (e.g., body fat). Excess energy in the diet can also reduce feed consumption and growth.

The major food nutrients (i.e. carbohydrates, proteins and lipids) are required by animals not only as essential materials for the construction of living tissues, but also as sources of stored chemical energy to fuel these processes. The ability of a food to supply energy is therefore of great importance in determining its nutritional value to animals. The nutrient composition of numerous feedstuffs can be found in the literature and on the Internet. One book that deals almost entirely with nutrient composition of feedstuffs is the "Standard Methods for the Nutrition and Feeding of Farmed Fish and Shrimp", which is available free on the Internet.

### **Essential Fatty Acids**

Essential fatty acids are fatty acids that are needed for growth and maintenance, however, the body is unable to synthesize them. In fish, fatty acids are the main component of egg membranes, maintaining structure and function. Fatty acids have an effect on the condition of female fish as well as a significant impact on spawning performance. Fatty acids are also essential to the development of larvae, and many aspects of development may be affected by the amount and type of fatty acids in a larval diet.

For herbivorous and omnivorous freshwater fish, many of the essential fatty acids are gained from freshwater algae. In most freshwater plants, many of the long chain n-3 and n-6 essential fatty acids are not found in abundant levels especially docosahexaenoic acid (22:6n-3, DHA). Arachidonic acid (20:4n-6, AA) and eicosapentaenoic acid (20:5n-3, EPA) are found in low levels in some freshwater invertebrates such as insect larvae, but are not in abundance, with the result that these fatty acids are still limiting.

The fatty acids in saltwater fish eggs differ from those found in freshwater fish eggs. Although eggs from both saltwater and freshwater fish are rich in DHA (docosahexaenoic acid) and EPA (eicosapentaenoic acid), AA (arachidonic acid) is more abundant in eggs of freshwater fish species. Because all three of these fatty acids are found in relatively high amounts in freshwater fish eggs, supplementing dry feed given to rainbowfish with these fatty acids should ensure proper egg formation and larval development.

*Melanotaenia splendida* fed a diet containing all three fatty acids produced eggs with a 100% survival to eyed embryo and hatching rate. They also had the highest number of eggs, highest number of spawns, and the shortest average spawning interval when compared to results from the other trials in the study (Badger, 2004).

### Feeding in Captivity

Although environmental factors may limit food availability, wild rainbowfishes generally have the opportunity in their natural environment to acquire adequate levels of all their nutritional requirements. The same cannot be said for captive rainbowfishes. Maintaining good health and growth in your rainbowfishes depends directly on the quality and quantity of the nutrients received through their daily diet.

Rainbowfishes that receive an inadequate diet grow poorly or not at all, and often exhibit deformities in the instance of vitamin/mineral deficiencies. Without the right amount of nutrients, rainbowfishes lose colour, stop breeding, and become susceptible to disease and, in certain circumstances, might even die. An undernourished animal cannot maintain its health and be productive, regardless of the quality of its environment. Obviously, no single food will meet their needs at all life stages, and the best way to ensure that they are getting a well-balanced diet is to feed them as wide a variety of food as possible.

Many old-time aquarists relied upon catching and culturing live foods to feed their rainbowfishes. This was mainly due to the fact that there was not the variety of commercially manufactured fish foods available then as there are today. Aquarists who were fortunate enough to discover pristine ponds inhabited by live foods generally keep them a close secret, and set out to collect them at dead of night, in order to keep them to themselves. The live foods that became most popular were those that could be easily collected or cultivated.

Up until the 1950's, keeping and breeding rainbowfishes in captivity generally required the routine collection of live food from streams and ponds or whatever the aquarist was able to find. Some of the items that were used included bloodworms, small garden worms, gentles (maggots), presoaked wheat, barley or millet grains, and now and then, coffee biscuit, vermicelli crushed small, occasionally varied with a little fresh food or gentles, and the various varieties of animalculae (*Aquarium and Terrarium Society of Queensland, 1928*).





The feeding of aquarium fishes was revolutionised with the invention of dried flake food by Dr. Ulrich Baensch, founder of Tetra Werke. Today, approximately 80% of aquarium hobbyists feed their fish exclusively with prepared foods, usually in flake or pellet formulations.

Flake food is the most common form and is consumed by a wide variety of aquarium fish. Any discussion that involves fish food deserves a brief explanation on these two types of commercially prepared foods. While flakes have been the most popular type of food for the past 50+ years, experienced hobbyists have learnt that pellets are the superior choice for feeding aquarium fishes. Pellets are preferred over flakes due to the fact that each pellet has uniform nutritional value for the fish. Not only can you feed much less on a volume basis, but pellets will also remain stable in the aquarium for an extended period of time. By their very design, flake foods are paper-thin and absorb water very quickly. Some studies suggest that once flakes are added to the aquarium, the majority of water-soluble vitamins (such as vitamin C) are leached out of a flake food within 60–90 seconds.

Most brands are designed to meet the nutritional requirements of multiple species. In this way, the manufacturer can satisfy a larger consumer base. A problem with this approach is that this "all purpose" feed may not be completely suitable for rainbowfishes. Since there are very few published reports on rainbowfish nutritional requirements, or an evaluation of formulated diets on growth and fecundity, it is not known whether the protein, lipid and carbohydrate levels in these diets provide the complete nutritional requirements of rainbowfishes.

Rainbowfishes can be maintained on a diet consisting totally of commercial feeds such as flake and pellet formulations. Such feeds however, need to be not only nutritious and palatable, but also capable of floating or sinking slowly. Rainbowfishes are primarily surface feeders so feeds that sink too quickly may become wasted in the gravel substrate and can cause water quality problems. Avoid feeding pellet feeds developed for the aquaculture industry, as they can cause health problems in rainbowfishes over the longer term. Rainbowfish keepers determined to keep rainbowfishes in foremost breeding condition and to raise and maintain the finest specimens should be aware that nutritional deficiencies may exist in commercially prepared foods. An ideal aquarium diet for rainbowfishes could include foods such as mosquito larvae, live and frozen daphnia (cladocerans), bloodworms and other aquatic invertebrates; chopped fish or shrimp, duckweed and blanched zucchini, spinach (*Spinacia oleracea*) or silverbeet (*Beta vulgaris ssp. cicla*).

In a well-established planted aquarium, rainbowfishes will always fine some green food among the aquatic plants, but a feed of duckweed, zucchini or spinach, at least once or twice a week is to be recommended. If duckweed is taken from natural waters it should be thoroughly cleansed before giving it to the fish. Failure to take this precaution may result in undesirable parasites being introduced into the aquarium.

Duckweed has higher concentrations of the essential amino acids, lysine and methionine, than most plant proteins and more closely resembles animal protein in that respect. Newly harvested duckweed contains up to 43% protein by dry weight with little to no indigestible material. Cultured duckweed also has high concentrations of trace minerals and pigments, particularly beta-carotene and xanthophyll that make duckweed an especially valuable supplement for rainbowfishes. The fairly high concentrations of these pigments deepen the overall body colouration of rainbowfishes. Duckweed can also be a rich source of ascorbic acid (2.78–4.90 mg/100 grams dry weight). The smaller species like *Wolffia* and *Lemna* are better for rainbowfishes and most will eat it avidly.

Freeze dried foods are another type of food that can be fed to rainbowfishes. The process of freeze-drying is to remove only the water content, leaving the nutrients content intact. If you feed freeze dried foods all the time then it would be best to pre-soak them in water before feeding to your fishes.

Frozen foods can be a more natural source of nutrients than dry manufactured foods. When feeding frozen foods, always thaw them to room temperature before feeding your fishes. Many frozen foods available from commercial suppliers often contain liquefied nutrients or processing water that may pollute the aquarium water when thawed. The best way to defrost frozen foods quickly is to place the frozen food into a strainer and run it under a cold water tap. This will also wash away the dissolved nutrients or any processing liquid that will remain uneaten if fed to your fishes. Although freezing does not significantly alter the nutritional content, nutrients do leach out rapidly into the water.

I formulated my own food mix which was a gelatine-based blend consisting of about 90% equal portions of fresh/frozen whitebait, shrimp, and mussel plus about 10% of beef liver and spinach. Small additions of other items such as dried mosquito larvae or similar dried foods, spirulina, fish roe, and a host of other gournet delights can be added to the mix. Food sources of marine origin are often utilised in fish foods because they are an excellent source of essential amino acids, fatty acids, vitamins and minerals. Variations of this formula could be tailored to suit nutritional needs of specific fish and according to available ingredients.



Vitamin supplementation is not necessary. Water-soluble vitamins such as B-complex vitamins and vitamin C are most vulnerable to nutrient leaching and must be replaced each day. Fat-soluble vitamins such as A, D, E and K dissolve in fat before they are absorbed in the blood stream to carry out their functions. Excesses of these vitamins are stored in the liver. Because they are stored, they are not needed every day in the diet and may cause problems if given in large doses. However, over supplementation with the aim to overcoming vitamin degradation and leaching should be avoided to limit the risk of hypervitaminosis. Although, overdose of most vitamins is unlikely to occur as excess vitamins are removed from the body via the kidneys. Brewers yeast, cereals, fish liver and various fish meats and oils contain most of the essential vitamins when fresh, and are key ingredients in most formulated foods.

The quality of ingredients in the food will have an effect on the adequate delivery of key nutrients to the fish especially in relation to all the water-soluble components of the diet. Care must be taken to overcome the leaching properties of certain foodstuffs in the aquarium. Once the nutrients are released from the diet into the water they are lost to the fish. Any vitamins or nutrients that are leached into the water are not known to be taken up through the skin or gills by freshwater fish species which, in a hypo-osmotic environment, do not drink.

The formulation of a nutritionally balanced diet for your rainbowfishes requires efforts in research and evaluation. Inadequate nutrition obviously impairs fish growth and reproduction, and can result in deterioration of health until recognisable diseases ensue. The borderlines between reduced growth and diminished health, on the one hand, and overt disease, on the other, are very difficult to define. There is no doubt that as our knowledge increases, the nature of the departures from normality will be more easily explained and corrected. However, the problem of recognising a deterioration of performance in its initial stages and taking corrective action will remain an essential part of the skill of the aquarium hobbyist.

The first consideration for the formulation of a successful diet is the quality of the feed ingredients. Diets produced with poor quality raw materials and under adverse processing conditions have inferior nutritive value and adverse effects on fish health. The composition of the ingredient obviously plays a determinant role in the quality. However, biological aspects, such as digestibility and utilisation of nutrients are most important and often overlooked.

The use of a good quality food will provide the fish with all the nutrients that they need to remain healthy and to grow. However, you should note that even good quality food will deteriorate if improperly stored or kept too long. Unfortunately, few fish food containers are stamped with an expiration date. Storage time for most commercial fish foods will vary depending upon environmental conditions; however, as a rule of thumb, 90 days is normally the maximum safe storage time for fish feed. Fish foods should be stored in a cool and dry place (refrigerator), and used within 30 days of opening, particularly in hot, humid climates.

Never feed mouldy, discoloured or clumped feed. Moulds on feed may produce aflatoxins, which can kill fish. Only buy large containers of food if you have a large number of fish. Frozen foods can probably be kept for 6 to 8 months. They should not however, be thawed and then refrozen.

Whatever choices you make regarding feeding your fishes, remember that variety is not only the "spice of life", but is the best way to provide them with the essential nutrients to give energy and build tissue. With proper feeding and aquarium management, most rainbowfishes will have the opportunity to live a much longer life in captivity than they would in the wild. A reasonably knowledge of the function of foods and feeding will therefore help in the choice of the most suitable food available for your rainbowfishes or in formulating your own mixture.

Sometimes rainbowfishes can be encouraged to experiment with unfamiliar food items by withholding other preferred food items. This works well, for example, when feeding blanched zucchini (a good source of B and C vitamins) for the first time. Simply withholding their regular food and offering the less familiar zucchini usually results in rapid acceptance (1–3 days). Similar strategies can be used to feed rainbowfishes homemade gelatine-based foods. Gelatine-based foods can serve as an excellent carrier for oral medications, but the fish should be use to eating a plain (non-medicated) homemade diet before there is a need to use it as a means of delivering medication. As with many sick animals, the appetite is often significantly depressed in sick fish, and that is not the time to try and introduce an unfamiliar food item.

Feeding live foods may not be necessary, but if you have access to them then this is by far the best food that you can feed to your rainbowfishes. However, the time required collecting live food from local ponds and streams and the risks of introducing pests, parasites and disease to a healthy tank are enough to make even the most avid aquarist question the desirability of such practice. What to watch out for are dragonfly larvae, hydra, leeches, planaria, snails, water beetle larvae, water tigers, and other carnivorous insect larvae. They may not bother larger rainbowfishes, but small fry have no defence against most of these pests. You can overcome all these problems by maintaining live cultures of cladocerans (daphnia, moina etc.), copepods, mosquito larvae, drosophila, whiteworm, etc., at home.

Rainbowfishes are well adapted to capturing live zooplankton and will show an active response to this type of food, indicating that zooplankton is a very attractive food for rainbowfishes. Upon adding zooplankton to the tank, it is quite often the case that rainbowfishes will exhibit an immediate response that somewhat resembles a feeding frenzy. Live zooplankton is preferred over inert food by rainbowfishes, suggesting a moving prey item may stimulate or influence feeding preference. In the presence of both live zooplankton and a pellet diet, rainbowfishes spend significantly more time feeding on zooplankton than on inert pellets. The natural diet of newly hatched rainbowfishes is dominated by zooplankton. Zooplankton is a rich source of protein and varies from 30–70% depending on life stage and nutrient availability of their phytoplankton diet.



### **Feeding Skills**

Although it is often postulated that feeding aquarium fish little and often throughout the day will result in more efficient feed utilisation, research has not yet been conducted to validate this hypothesis. Published literature on frequency of feeding aquarium fish is limited when compared with that available for aquacultured fish species. This has led to uncertainty in the feeding routines used by many rainbowfish keepers. Both over and under feeding can be detrimental to the health of rainbowfishes. Rainbowfishes have different dietary requirements based on size, age and water temperature, therefore the amount of food fed to the fish should vary accordingly. This generally means more frequent feeding for fish of smaller size such as larvae and juveniles.

Many problems can be encountered when feeding rainbowfishes in an aquarium. First, delivery of feed to fish in a water medium requires particular physical properties of feed together with special feeding techniques. It is not possible in the literal sense to feed rainbowfishes on an "ad libitum" basis. The nearest alternative is to feed to "near satiety" with a pre-determined number of feedings per day; however, this can be very difficult and subjective.

Rainbowfishes will, however, easily adapt to a feeding schedule. In their natural environment, rainbowfishes will adjust their food consumption to satisfy their requirements. If the amount of food available decreases they will increase their food intake during feeding. In captivity, however, rainbowfishes have to rely entirely on the aquarist on the amount, as well as when and how often, they feed. This means that a dominant fish may be able to consume more feed than other less-dominant or smaller fish, which are held in the same aquarium. Being attentive to changes in appetite of fish is, nevertheless, a very important skill that aquarists must acquire.

Feeding frequency and timing is another factor that has been suggested as affecting feed intake and utilisation by rainbowfishes. On a weekly basis, studies have suggested that feeding the equivalent of six days a week resulted in growth performance similar to feeding seven days a week. Feeding five days a week resulted, however, in significantly less growth. There is no dependable evidence that daily feeding frequency and timing affect feed utilisation. The most important factor is to insure frequent and spaced meals to insure that the fish can consume enough feed to meet their growth potential.

It is obvious that food availability and abundance have important effects on reproduction. However, it is not always the case that more food is better. Therefore determining exactly how much food and how often to feed the fish in order to maximise reproductive capacity is important for optimising reproductive output. Feeding too little will result in decreased reproduction, while feeding excessively may compromise fish health and increase the probability of having poor water quality problems.

Rainbowfish feeding research (Badger, 2004) found that *Melanotaenia splendida* coped much better with lowered

rations on a daily basis then infrequent feeding. From this study it may be deduced that it is not as important in this species to feed to 100% satiation every feeding as it is to feed every day. Although a 100% ration on a daily basis would still provide the best reproductive performance. These findings are important as it is necessary to know how often and how much to feed the fish in captivity.

Knowing how much to feed rainbowfishes without overfeeding them is a problem for most aquarists. Rainbowfishes always seem to be hungry; you can feed them, and five minutes later they will look at you as if they are starving. This is because the fish have learnt to associate you with food and get excited whenever they see you. It is necessary to understand about conditions in the wild to appreciate why rainbowfishes have this behaviour. Food is a limiting resource in nature and it is rarely available in excess quantities. The individuals that survive are those best able to acquire this limited resource. Rainbowfishes are opportunistic and feed whenever they can find food. It is neither necessary nor advantageous in the wild to control their appetite.

Water temperature also directly influences the desire of rainbowfishes to feed. Being poikilothermic animals, the metabolic rate, growth, energy expenditure, and feed intake are highly influenced by water temperature. In tropical waters, which have prevailing high temperatures, rainbowfish generally grow faster, mature younger, and have a shorter life span than rainbowfishes in temperate waters. It is, therefore, important to understand how water temperature affects these parameters. When temperatures drop below 20°C, rainbowfishes will consume less food so reduce the feeding rate accordingly. During cold weather conditions, it is best to feed late in the afternoon when the water temperatures have had a chance to elevate during the day.

Most rainbowfishes require a feeding ratio of about 5–10% of their body weight daily and ideally, should be fed this amount in four or five smaller feeding's during the day. However, knowing the weight of your fish is not practical, so feed sparingly, and supply enough to give three to five minutes of continuous feeding per meal. The actual amount of food required depends on the type of food, aquarium conditions, and individual fish.

Juvenile rainbowfishes consume a higher percent of their body weight per day than do adult fish. Newly hatched larvae need to feed continuously. Research has shown that juvenile rainbowfishes will grow faster if fed three or four times a day. Feeding several times a day can also reduce problems of feeding dominance among juveniles of different sizes. Multiple feeding's also spread the waste load on the biofilter.

Feeding is also the best opportunity for you to observe the overall vitality of your fish. Poor feeding response is a signal that something has gone wrong in the aquarium. Check all aspects of the system particularly water quality, and look for signs of disease or stress.



#### Feeding and Nutrition

At present there is little information available concerning the quantitative dietary nutrient requirements of rainbowfishes under aquarium conditions, or regarding the nutrient requirements of the first-feeding larvae and broodstock of rainbowfishes.

Data from aquaculture research is commonly extrapolated and applied to aquarium species, which often proves to be unsatisfactory because of the differences in fish species and variation in diet formulations which is aimed at maximum growth in a short time period. This might be of value for the commercial farming of ornamental species, but would be unsuitable for rainbowfishes kept in home aquaria. In addition, the majority of research has focused on feeding fish to satiation, measuring the food intake, and linking this to growth performance and utilisation.

Under aquarium conditions, rainbowfishes characteristically display a considerable range of growth rates. Despite this, and on the basis of 'laboratory based' nutritional feeding trials conducted to date, some generalised conclusions may be drawn regarding the recommended nutrient levels. Nevertheless, there are still many unanswered questions when it comes to the nutritional requirements of rainbowfish species.

The formulation of adequate diets for organisms in an aquatic environment gives a greater challenge to those involved in their care, compared to terrestrial species. In general the maintenance energy requirement of fish is less than 10% of the maintenance energy required by birds or mammals. The low maintenance energy requirement is partly due to the poikilothermic nature of fish. Fish also exert less energy on posture and have an energetic advantage over mammals in their nitrogenous waste management as they excrete mainly ammonia instead of urea or uric acid, thus losing less energy in protein catabolism and excretion of nitrogenous waste.

The provision of energy and other nutrients to poikilothermic animals can take many forms and is generally chosen by reference to their natural ecology. However, it is pertinent to note that feeding diets high in protein to herbivorous and omnivorous fish may result in increased tank pollution as excess protein is for the most part excreted as waste.

Feeding strategies and anatomical differences between fish species make formulation of one diet for a community of species quite difficult. Fish are unique in that only a small proportion of species use plant material as their primary food source. Those species consume nearly their body weight in vegetation daily.

Most fish eat a diet rich in protein and fat and digestion of plant materials by these species is poor. Therefore, most commercial fish diets contain protein levels above 30%, and diets for very young fish may contain nearly 60% protein. Commercial diets are partially cooked during production, which increases the digestibility of plant products. Lipids are also an important energy source for fish, and a source of essential fatty acids. The most desirable fats are unsaturated. Some research on the nutritional requirements of freshwater ornamental species in a commercial production environment has been conducted, mainly in Singapore. Protein requirements varied from around 30% dietary protein for growing omnivorous goldfish (*Carassius auratus*) to 50% for the carnivorous discus (*Symphysodon aequifasciata*). Whereas, mineral (phosphorus, iron, magnesium, zinc) requirements have received some attention in feeding *Poecilia reticulata*, few researches have concentrated on vitamin requirements of ornamental species.

Maintenance energy levels of ornamental fish varied from 0.068 kJ per day for *Paracheirodon innesi* to 0.51 kJ per day for *Trichogaster microlepis*, kept at a water temperature of 26°C. However, large commercial producers of aquarium fish in Singapore emphasise the importance of regular supplementation of formulated feeds with live feed, as the inclusion of live feed improves growth.

A study to determine the effect of increasing levels of dietary protein on the common swordtail (*Xiphophorus helleri*) was carried out by Chong *et al.*, 2004. Five semipurified diets having similar caloric values, containing 20%, 30%, 40%, 50% and 60% dietary protein were used. Results showed that while the 20% and 30% protein produced the lowest specific growth rate values, there was no significant difference between 40% and 60% dietary treatments. The 20% dietary treatment also displayed lowest protein content in both ovaries and muscle of female fish. Fry production was highest from females fed with 50% and 60% protein, followed by the 30% and 40% protein while the diet containing 20% protein produced lowest number of fry.

A significant correlation was also obtained between number of fry produced and the weight of female fish, indicating that size is a major factor influencing production. Relative fecundity was lowest for the 20% protein diet followed by the 30-40% and 40-60% protein diets. There were no significant differences in both weight and length of fry produced among the dietary treatments. Based on these results, they suggested that a minimum of 30% protein be included in the diet of female swordtail broodstock.

In another study of *Xiphophorus helleri* (Kruger *et al.*, 2001), three protein levels (30%, 38% and 45%) at three different dietary lipid concentrations (6%, 8% and 12%) were used to formulate nine different diets that were fed for 60 days to 6–8 weeks old juvenile *Xiphophorus helleri*. From that study it was suggested that a diet of at least 45% protein and 6% lipid concentration is needed for the best specific growth rate and feed conversion ratio.

Larval goldfish were found to grow best on prepared diets containing about 50% protein (Sales & Janssens 2003). Juvenile goldfish grew best on prepared diets containing about 40% protein. As they grow to adulthood, their nutritional requirements change. Adult goldfish require only 29% protein, and they will continue to grow when receiving only 1% body weight of a diet containing 36% protein.



### Feeding Larvae

The aquarium breeding and propagation of rainbowfishes requires not only specific aquarium conditions, but in most cases also the cultivation and use of live food organisms for feeding newly hatched larvae. The larvae require culture techniques which are normally different than juveniles or adults, especially with respect to feeding. The main reason for this is that the developing larvae are usually very small, extremely fragile, and generally not physiologically fully developed. For example, larval rainbowfishes have a small mouth size and are limited in the size of prey they can consume. Moreover, their ability to forage for prey is limited due to their small size and limited fin development. These are limiting factors in proper feed selection and use during the early first-feeding period. High mortality rates can often occur, especially during the early feeding stages. It is perhaps not surprising therefore that larval nutrition can be a major obstacle in the successful raising of rainbowfishes in captivity. However, mortality can be the result of several factors including inbreeding, inferior water conditions and improper hatching conditions.

Rainbowfish larvae in the wild feed on a wide range of live foods, but under aquarium conditions they are usually fed on a limited number of foods (two or three) which frequently are not part of their natural food and hence their nutritional composition may not always be the most suitable for maximum growth, development and survival of the larvae. Moreover, larvae undergo several morphological and physiological changes which in nature are simultaneous with changes in behaviour and even habitat and type of food consumed. All these changes will affect the nutrient availability and feed utilisation by the larvae in order to match their growing requirements. In theory, most of these problems can be simplified by the proper development of diets which are able to cover nutritional requirements at different times of larval development. In order to achieve those diets we need, among many other important things, is to have a complete knowledge of the nutrient requirements for rainbowfish species.

As the larvae grow, the amount of food they need increases and they prefer to eat larger-sized prey. It has been generally accepted that the optimal prey size for fish larvae is determined by their mouth size. Commercial food fish larvae feeds have been developed primarily based on mouth size data. This has resulted in successful improvement of larval survival and growth in many aquacultured fish species. Adjustment of the amount of feed depending on larval developmental stage has also resulted in better growth and survival.

The mouth size of first-feeding larvae restricts the size of the food particles which can be ingested. However, there are few, if any, published information on the mouth size of rainbowfish larvae. In general, mouth size is correlated with body size, which in turn is influenced by egg diameter and the period of endogenous feeding (i.e., yolk sac consumption period). Rainbowfishes are lecithotropic; they live off the yolk sac provided within the egg for a certain period of time after hatching (the yolk will be absorbed slower in cooler water and faster in warmer water). This means that there is a certain period of time where it is not necessary for a larval fish to obtain their nutrition from external sources.

Rainbowfish larvae can survive for as long as 8–10 days without external food sources providing that the yolk sac contained enough nutrients. However, if an egg does not hold adequate nutrients, larvae hatching from that egg would have a shorter period of unfed larval life than a larvae hatching from an egg with a good supply of nutrients. Both protein and lipid are very important constituents of fish eggs. They are the primary components of vitellogenin, the lipoprotein that is the main energy source in the yolk of eggs.

Live feeds are a convenient food source for the larvae of all rainbowfish species. In their natural environment rainbowfish larvae feed primarily on zooplankton. Zooplankton is importance to the early life history stages of rainbowfishes. However, these organisms are not normally available in captivity. Zooplankton is a broad categorisation spanning a range of organism sizes that includes both small protozoans and large metazoans. It includes holoplanktonic organisms whose complete life cycle lies within the plankton, and meroplanktonic organisms that spend part of their life cycle in the plankton before graduating to either the nekton or a sessile, benthic existence. An abundance of zooplankton is particularly important for larvae to develop into juveniles and for juveniles to develop into subadults.

Most rainbowfish larvae are not particular about the types of zooplankton they will eat, but the animals must be small enough for the larvae to ingest. Rainbowfish larvae are usually small in size (4–6 mm, total length), have poorly developed eyes, do not swim all that well but are mostly present in the water column, and require easily digested food. Having such tiny mouths, the size of the food is crucial to their surviving the most difficult period in their lives. Zooplankton swim slowly and stay suspended in the water column, thus being available for capture and consumption by larval rainbowfishes. Larger juveniles fed very tiny zooplankton may grow slowly because of the energy expended in catching the small prey, but usually they will not starve if enough zooplankton is available. However, feeding tiny larvae with zooplankton that are mostly too large for them to eat usually results in starvation.

Natural foods may also include phytoplankton and detritus. As plants decompose they become broken down into tiny fragments. The fragments become colonised by bacteria and fungi which feed off the decomposing material. These tiny fragments and the microscopic plants, animals, bacteria and fungi associated with them are known as 'detritus'. Detritus is a major component of a rainbowfishes diet at all stages of its life cycle. The tiny plant fragments themselves are not very nutritious but the microorganisms associated with them are a readily digestible, nutritious, protein rich food source. Phytoplankton is a particularly important food source for larval rainbowfishes. Filamentous and unicellular algae, especially epiphytic diatoms and desmids, also provide an important food source (> 10% of diet) for rainbowfish larvae.

In general, if you want your rainbowfishes to achieve maximum growth and have low mortality rates then you will need to provide them with at least some live foods. This often means that you will have to culture or collect live foods. They should also be fed several times a day. This is not always possibly as we all have other things in our life to do each day.



However, there are several kinds of live foods that can be fed to rainbowfish larvae that will remain alive in the aquarium until consumed. Live foods such as infusoria, zooplankton (rotifers, cladocerans, copepods, etc.), microalgae (greenwater), or other micro-organisms all fall into this category. Microalgal species can vary significantly in their nutritional value, and this may change under different culture conditions. Nevertheless, a carefully selected mixture of microalgae can offer an excellent nutritional package for larval rainbowfishes, either directly or indirectly (through enrichment of zooplankton). As the larvae develop they can be fed other live foods such as brine shrimp nauplii and microworms that will remain alive in the aquarium from two to four hours, providing food until you can feed them again. Brine shrimp nauplii are used primarily because of their ease of culture.

The numerous attempts to replace live food by artificial diets has had limited success so far, i.e., the highest growth rate in larval rainbowfishes is still obtained with live food. The reason why live food is better for growth has not yet been clearly defined. Perhaps proteins present in phytoplankton and zooplankton but not synthesised by the physiological system of the larvae are important. In such cases, live food organisms provide digestive enzymes that breakdown the food ingested by larvae. However, other evidence has led to contradictory views regarding the role of the live food contribution in the digestion process of fish larvae.

Live food organisms contain a 'package' of enzymes, gut neuropeptides and nutritional 'growth' factors that enhance digestion. These substances are frequently omitted in formulated diets. Moreover, particulate diets for fish larvae contain proteins and other ingredients that can be difficult to digest (especially since formulated diets contain 60–90% dry matter while zooplankton has only 10%). Inclusion of digestive enzymes, especially proteases, in the diets for fish larvae has been reported to significantly improve nutrient utilisation and performance of larvae, but still not as much as larvae fed on live food.

Additional support for this hypothesis is found in the different growth results when fish larvae are fed with either decapsulated cysts or nauplii of Artemia. On an individual weight basis, the decapsulated cysts and nauplii of Artemia have similar biochemical composition in all the major nutrients. Thus, with regard to the amount of nutrients, there is no difference in feeding Artemia cysts or nauplii to fish larvae. However, in some species higher growth rates have been achieved with nauplii. Also of importance are several essential biochemical compounds such as poly-unsaturated fatty acids. Primary producers of these fatty acids are phytoplankton and zooplankton.

From the practical viewpoint of the aquarist, a good diet should be readily available, simple to produce as well as versatile in application. The consistent availability of sufficient quantities of food organisms is of the utmost importance in successfully breeding and raising rainbowfishes. In this respect, the collection and feeding of wild plankton has proven unreliable and not always practical. In response, there has been increasing interest in developing alternative feeds for fish larvae, including alternative zooplankton species and formulated diets.

The major difficulty for the aquarist is providing organisms appropriate to the size of the larvae at the first feeding stage and then supplying the large numbers necessary to maintain them. The preferred size of prey for larval fish increases as mouth size and feeding competency increase and different types of live foods have to be cultured for the different stages in the larval rainbowfishes development. For example, different species of microalgae (phytoplankton) range from 2 to 100  $\mu$ m; rotifers from 50 to 200  $\mu$ m, copepods from 100 to 300  $\mu$ m and brine shrimp nauplii 400 to 800  $\mu$ m.

Apart from these main groups, a few other live feeds are used on a more limited scale including microworm (*Panagrellus redivivus*), vinegar eels (*Turbatrix aceti*), and cladoceran crustaceans. This group includes many species (*Daphnia*, *Moina* etc.). They can reach up to 4–6 mm but typically are much smaller than this; the smallest species is around 250  $\mu$ m. Larger juveniles and even adults of some species often selectively prey on these crustaceans.

### **Feeding Program**

The essential requirement for successfully raising rainbowfishes is the implementation of a suitable feeding regimen for the larvae. Using the following feeding program, the survival of rainbowfish larvae can be as high as 80–95%. Begin feeding with small amounts of infusoria. The term infusoria is a collective name for many micro-organisms and can include paramecium, unicellular algae, ciliates, bacteria, protozoans, desmids, rotifers, and a host of other small organisms.

Rainbowfish larvae will also benefit from the addition of small amounts of greenwater (phytoplankton). It is well known that phytoplankton (microalgae) contain an array of essential nutrients that help the growth of rainbowfish larvae. Providing natural greenwater with infusoria as food for the newly hatched rainbowfishes has several advantages. The larvae are easily able to switch to different sized prey, a feature not present in monocultures of organisms such as rotifers or brineshrimp. Greenwater also enables the "infusoria" to feed on resident algae and microbes, thus retaining their nutritional value for greater periods of time. Feed 5–10 ml (per 20–50 fry) of infusoria suspension three or four times per day. You can tell if they are feeding well as the larvae should have nice "swollen stomachs" after feeding.

Starting on day 7 to 14, depending on size, you can begin introducing small amounts of newly hatched brine shrimp nauplii and/or microworm. The feeding of rainbowfish larvae on the brine shrimp nauplii will be very inefficient at first; so add just enough such that the fry will encounter a small "cloud" of nauplii for a few minutes in the water column. Do this each time you feed brine shrimp nauplii to older fish. Observe fish closely after feeding.



Once you can see that the fish have been successfully taking the brine shrimp (their bellies will be orange), add more with each feeding. Never add so much brine shrimp that more than 10–20% of it dies in the tank. Brine shrimp will accelerate growth so it is important to commence feeding as soon as possible. The smallest larvae will continue to eat the infusoria while the larger ones will start feeding on the brine shrimp nauplii or microworm.

Gradually increase the proportion of brine shrimp nauplii or microworm and phase out the infusoria - this should be around day 14 to 21. Continue feeding the juveniles with brine shrimp nauplii and microworm three times a day. Do not overfeed, a good rule of thumb when feeding brine shrimp nauplii is that they should be mostly eaten after about 20 minutes, at which point the babies will have nice red stomachs. When weaning fish to a new food, introduce 10% of the new food daily while reducing the same percentage of the initial food until 100% of the new food is accepted. Commence feeding adult foods as soon as the juveniles are big enough to eat it and feed them often (at least twice daily).

In addition to microworm or brine shrimp nauplii feed them once each day with a sprinkle of powdered spirulina. Spirulina is an algae-derived food rich in protein, carbohydrates, amino acids, vitamins and minerals, and essential fatty acids. Spirulina powder is available from most health food stores. The powdered microalgae Chlorella and Dunaliella that are sold in most health food stores can also be used as aquarium food supplements.

The growth rate of the rainbowfish larvae is generally slow, with little variation until around 14 days. After that time growth rates increased. However, considerable variations in the growth rate of juvenile stages of rainbowfishes have frequently been observed. Lack of food during early development leads to appreciable variation in growth (length), if compared to larvae offered food ad libitum. Adequate food and suitable temperature generally results in higher growth rates. Beside effects on growth, a decline in mortality between fish receiving small amounts of food and well-fed fish has also been observed.

It should be noted that the hatching of eggs might vary, resulting in the presence of larvae at different stages of development. As the larvae increased in age, the variation in length between individuals also increased. Thus, different factors can increase the duration until sexual maturity is reached. If you have a batch that differs greatly in size, you will often find that the smaller ones are females. To raise an entire spawning, you may have to sort the growing fish by size, as the larger ones will eat their smaller siblings or repress their growth rate.

Size grading separates the faster and slower growing fish. When these smaller fish are transferred to another tank, their growth rate is no longer negatively impacted by the faster growing individuals. They should increase their growth rates to compensate for the initial retarded growth rates that developed during the nursery phase. In addition, a high fish load can retard the growth of juveniles, probably due to release of growth-inhibiting substances. For maximal growth keep the number of fish per aquarium low and avoid overcrowding.

Uneven larvae growth can also lead to cannibalism. Uneven larvae growth rates means that some will quickly reach the size where they are capable of ingesting other, smaller larvae. Rapidly growing larvae have a high food intake requirement. If that need for food is not satisfied by supplemental feeding they might start looking for extra food sources such as their smaller siblings in the tank. It is believed that larger larvae are also more aggressive and successful at feeding when supplementary food is added. This then gives the larger larvae a further growth advantage. The main way to control cannibalism is with regular size grading. This is the single most effective method for reducing cannibalism. Also, if the fish are fed regularly and sufficiently, it reduces the number of fish that are hungry and may decrease the cannibalism on their siblings.

General maintenance includes daily siphoning of the aquarium to remove any uneaten food and faeces, and 25–50 percent waterchange at least one a week. The continued growth and development of the fish will vary from one hobbyist to another and is largely conditional upon captive environmental conditions such as temperature, water quality, and feeding regime. Sexual differences begin to appear between 9 and 12 weeks after hatching with sexual mature at 6 to 12 months.

### Formulated (Dry) Diets

If you are unable to culture or feed live foods to your larval rainbowfishes, you can still get reasonable results rearing them entirely on a fine powder-based diet manufactured by a number of commercial suppliers. Advantages include 'offthe-shelf' availability, which is especially important in remote locations and for hobbyists with limited resources. However, artificial diets are ingested at a lower rate than live foods, and are negatively buoyant in water.

Generally, rainbowfish larvae fed only dry formulated diets often show lower survival and poorer growth compared to those fed live foods and can often lead to higher incidence of deformities. Non-living feeds do not yet have an advantage over live food organisms. During recent years intensive research has been conducted by a number of research groups around the world to develop micro diets that can partially or fully replace the use of live food. Substantial advances have been developed and commercial micro-diet formulations are becoming more and more successful as a partial replacement for live foods.

Two types of micro-particulate particles have been used for manufacturing formulated diets for feeding fish larvae; these are (1) micro-encapsulated diets and (2) micro-bound diets. Both have been used extensively in nutritional studies with fish larvae. The major difference between the two is that micro-encapsulated diets have a membrane or capsule wall which separates dietary materials from the surrounding medium. The capsule wall helps maintain integrity of the food particle until eaten and helps maintain water quality;



however, it may restrict leaching of water soluble dietary components and therefore reduce the attractability of the food particles. The capsule wall is also thought to impair digestion of the food particle and a number of studies have reported poor growth and survival of fish larvae fed microencapsulated diets.

Micro-bound diets consist of dietary components held within a gelled matrix or binder. They do not have a capsule and this has been suggested to facilitate greater digestibility and increased attractability through greater nutrient leaching. Many different binders have been used in micro-bound diets including polysaccharides from seaweed such as agar, carrageenan and alginate and proteins such as zein and gelatine. They vary considerably in their properties and nutritional value and choice of binder can significantly influence the rate of ingestion of formulated food particles and nutrient assimilation. Water stability of micro-bound diets is also influenced by the binder employed. Micro-bound diets made from agar and alginate were amongst the most stable in terms of integrity, while carrageenan was amongst the poorest.

Prepared diets should be of a very fine grain, nutritionally complete, palatable, be less than 100 microns ( $\mu$ m) in size, and remain floating or suspended in the water column. I have found OSI<sup>®</sup> Micro-Food an excellent first food for rainbowfish larvae. OSI Micro-Food is a microencapsulated diet rich in protein, fatty acids, and vitamins. OSI Micro-Food or artificial plankton (APR), as it is known in the aquaculture industry, was designed for feeding prawn larvae, but it works well on larval rainbowfishes too. Just use it dry and sprinkle it over the surface of the water. Other well-known products such as 'Sera<sup>®</sup> Micron and TetraMin<sup>®</sup> baby fish food are also suitable first food for rainbowfish larvae.

It is important that the feed size increases as the larvae grow until they are large enough to take regular flake and pellet feeds. It can be advantageous to mix the sizes for a few weeks, especially if there is size variation within the larvae. Care should be taken not to overfeed, as prepared diets tend to easily decompose and pollute the water if overfed. Just a small tip here, keep all your fish foods in the refrigerator, they last longer.

Another simple, less expensive but very efficient method to feed larval rainbowfish is hard-boiled egg yolk. Just wrap the yolk in a clean cloth (or old nylon stocking) and twist it down into a section of the cloth. Then all you have to do is swish it around in the water. The egg yolk will keep in a refrigerator for a few days. An even easier option is to purchase some powdered egg yolk from your local baker. Powdered yolk can be stored in the refrigerator for several months. When you want to feed the yolk just sprinkle it dry on the aquarium water surface.

The copepod, *Diaptomus connexus* (sold freeze-dried under the commercial name of Cyclop-eeze<sup>®</sup>) is considered to be another excellent food for juvenile rainbowfishes by many aquarists. Cyclop-eeze is a commercially available product marketed by Argent Chemical Laboratories. Its actual purpose is the replacement of brine shrimp, spirulina and artificial plankton (APR) as starter diets for larval fish and shrimp.

The manufacturer describes Cyclop-eeze as a superior natural product, suitable for almost any aquaculture purpose. The copepods the product consists of "are selectively bred, biologically engineered micro organisms which are cultured in a pristine arctic lake". The decapods harvested for the production of the product are described to be of "blood orange" coloration. The company explains this with the high level of astaxanthin. The importance of carotenoids in fish diets has been stressed by several researchers working in the field of fish nutrition. In addition to the high content of carotenoids, high levels of highly unsaturated fatty acids (HUFA) are also found in the copepods. All polyunsaturated fats are increasingly recognised as important to animal health. High levels of omega-3 HUFA have been described as favourable for larval aquaculture applications and typically are obtained by food supplements.

Cyclop-eeze is also seen an interesting product because it occurs to be rich in the attractant betaine as well as other palatability factors. Betaine as well as carotenoids and phospholipids are widely used as attractants and stimulants. Unfortunately, not a lot of research on Cyclop-eeze has been published. The information available on nutritional values and compositional aspects of Cyclop-eeze are therefore limited to the manufacturer's information and little independent research has been conducted evaluating the product in fish feeding.

However, there are now numerous commercial products available for feeding fish larvae and they are all certainly worth trying. Recent developments have included the use of a newly formulated larval micro-diet (Gemma Micro<sup>®</sup>) that has helped reduce brineshrimp usage by more than 95 per cent. This diet has proven to be more attractive and beneficial to the fish. You need to try these products yourself and see if they are suitable for your situation. Evaluating and combining different food products can provide suitable diets for any of the fishes we maintain in captivity and can closely resemble the wild diets with which they have evolved. This should minimise nutritionally related health issues and improve your success as well as the general wellbeing of the fishes.

### Frozen Food

Frozen food can usually be fed approximately 14 to 21 days post hatching. Instead of hatching fresh brine shrimp nauplii for your baby and juvenile rainbowfishes every day, you might consider making frozen brine shrimp blocks and feeding these instead. First, hatch a large batch of brine shrimp. After hatching, collect the shrimp in the normal way and rinse with fresh water. Place the normal ration of brine shrimp for a tank into the well of an ice cube tray (plastic ones that make small cubes are best). Add fresh conditioned water to fill the trays and freeze. When it is time to feed the fish, just pop out a block and add to the tank. The ice will slowly melt and release the brine shrimp, feeding your fish. You can do the same thing with other types of live food.



A suitable homemade food can be made up with fish, shrimp, and/or squid flesh, chicken eggs, beef liver, and cod liver oil. The fry should be eating brine shrimp nauplii before feeding this food.

Percent weight: Fish, Shrimp, etc. – 85% Cod Liver Oil – 2% Chicken Eggs – 10% Beef Liver – 3%

#### A recommended procedure for preparation is as follows:

Thaw shrimp, squid or fish (if frozen) and blanch the liver in boiling water for at least 10 minutes. Homogenise with fish, shrimp, or squid in a food processor until well blended (smooth texture with no chunks). Mix chicken eggs, cod liver oil, and then add to food processor. Add a binding ingredient (gelatine or agar) gradually, and continue mixing slowly until a paste is formed. Take the paste and place in a plastic ice cube trays and store frozen. Later, the frozen food can be finely grated with a cheese grater.

This procedure will result in a mixture of particle sizes ranging from 250 to 1,000  $\mu$ m. However, depending on the size of the fish, you may require a certain particle size. To obtain the specific particle size, take a frozen cube out of the freezer, thaw and blend with water in an electric blender and manually press the material through a sieve having a specified size. Drain the sieved diet to remove dissolved particles that can foul the water and contribute to bacterial growth within the nursery tank.

### A selection of larvae food and sizes

Brine Shrimp: 400–800 micron (depending on origin) Decapsulated brineshrimp eggs: 200–250 micron OSI Micro-food (APR): 50–150 micron Cyclop-eeze: 750–800 micron Active Spheres Golden Pearls: 5–50 micron Golden Pearls Rotifer Size I: 50–100 micron Golden Pearls Rotifer Size II: 100–200 micron Golden Pearls Artemia Size II: 200–300 micron Golden Pearls Artemia Size II: 300–500 micron Golden Pearls Weaning Diet: 500–800 micron Golden Pearls Juvenile Diet: 800–1000 micron

#### Hatchfry Encapsulon

Hatchfry encapsulon is formulated to resemble artemia nauplii. Sized matrix bound, micro-particulate larval feed are excellent food for shrimps and fish. Argent has manufactured and distributed Hatchfry Encapsulon for more than two decades. All grades of Hatchfry Encapsulon are chemically composed so that their protein content, as expressed in the amino acids profile, closely resembles that of artemia nauplii. Larval of freshwater shrimp as well as most marine and freshwater fish fry will thrive on this diet.

Grade 0: 30 micron Grade I: 50–150 micron Grade II: 150–250 micron Grade III: 250–450 micron





## Foods & Feeding

### Infusoria

The term infusion animals (animalculæ) was introduced by the German naturalist, Martin Frobenius Ledermüller in 1763, to include all those microscopic animals that appeared in water in which hay had been steeped for several days. This was soon formalised to infusoria by Heinrich August Wrisberg in 1765. Wrisberg described what he took to be the process by which the decaying animal and vegetable matter in infusions produced infusoria. Many of these organisms would originally have been present as spores and cysts, so the infusoria that appeared typically included rotifers and other protozoa, as well as algae.

At the beginning of the nineteenth century infusoria comprised a vast array of organisms spanning in size and complexity from bacteria to small invertebrates including worms and crustacea found in lakes, ponds, and streams, as well as in infusions of decaying organic matter exposed to the air. Today the term 'infusoria' is mainly an informal aquarium hobby name that covers a wide diversity of microorganisms, including singlecelled, colonial and multicellular forms.

By far most 'infusoria' are found in freshwater free from actively-decomposing organic matter, rather than in those that contain organic substances in a putrescent state. They may be "benthic" (living in bottom sediments) or "epiphytic" (living on aquatic plants). They are an important link in detritus-based food webs in aquatic ecosystems and are consumed by other small animals, which are in turn preyed upon by larger organisms. Their principal importance is as consumers of bacteria. Infusoria feed on bacteria and organic detritus, hence when they present themselves in vegetable infusions, it is usually after decomposition. The decomposing organic matter feed the bacteria, which are even smaller than infusoria. They are not, however, absolutely confined to water; for there are a number of species which can maintain their existence in damp mud.

Many microorganisms can be collected in the wild and cultured as infusoria. Organic materials such as aquatic plants, leaf litter or pond sediment can be collected and added to a container that is then filled with water. After standing for a few days, the plant material decays and the infusoria will have come to the surface, where they can be siphoned off. A tendency of most infusoria is towards the light, and also to the surface. Such a culture will produce infusoria for a number of weeks, without any further treatment. When the infusoria begin to decline, they can be caused to increase again by the addition of small quantities of vegetable matter into the water to induce bacterial multiplication.

There are problems in collecting from the wild however, most notably the risk of introducing disease or pests into your culture. In addition to collecting organisms in the wild they may also be purchased as pure cultures from biological supply companies. Obtaining a starter culture from a fellow hobbyist is probably the simplest solution.



Infusoria Culture

Most aquarium related cultures of infusoria are a collection of many microorganisms and can include microscopic algae, bacteria, protozoans, rotifers, and a host of other small organisms. Unless you are maintaining pure cultures in sterile conditions, your culture will have a mixture of protozoans and algae in greater or lesser degree. Variety in composition is one of the outstanding characteristics of infusions in which infusoria are to be cultured. There appears to be no single method of successful culturing. In general, small quantities of materials, usually organic, must be introduced into water to induce bacterial multiplication. These bacteria are the chief food supply of the infusoria, at least in the earlier stages of the culture.

Although infusoria have been cultured and fed to fish larvae for more than a hundred and fifty years, and many fish breeding articles advise infusoria as a first food for fish larvae, there have been very few articles published in the aquarium literature on how to successfully cultivate them. The procedure adopted by old time aquarists for obtaining a culture of infusoria was by boiling hay, lettuce, spinach or other vegetable matter and allowing the resultant infusion to decompose in the air for a while in the hope that various living beings developed therein. At best this was a hit and miss method and in many cases it simply didn't result in a successful culture.



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"Now the water in an aquarium which has been kept for any length of time necessarily becomes more or less charged with the effete matter of its inhabitants, which if allowed to accumulate, would soon render the fluid poisonous to every living thing within it. This result is happily averted by Infusoria, which feed upon the decaying substance in solution, while they themselves become in their turn the food of larger animals. Thus the presence of Infusoria in the tank may be considered a sign of its healthy condition, although their increase to such an extent as to give a milky appearance to the water is apt to endanger the well-being of the larger, though delicate creatures.

The peculiar phenomenon allured to arises from decaying matter, which should be sought after and removed with all possible speed. The whereabouts of such objectionable remains will be generally indicated by a dense cloud of Infusoria hovering over the spot. The milkiness, however, although it may look for the time unsightly, is ofttimes the saving of the aquarium 'stock'. When these tiny but industrious scavengers have completed their task of purification, they will cease to multiply, and mostly disappear, leaving the water crystal clear.

I believe it is the absence or deficient supply of Infusoria that sometimes so tantalizingly defeats the attempts on many persons to establish an aquarium." (John Harper, 1860)

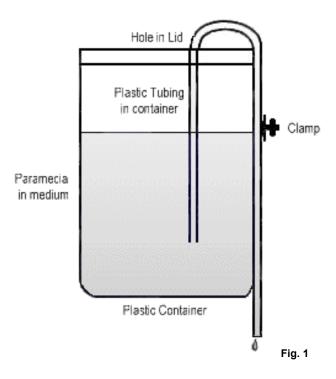
The best way to bring about a successful culture is to make a vegetable based infusion similar to the above (slices of potato or banana skin works well) using water from a well-established aquarium or outdoor pond (that hasn't been chemically treated). Cultures should be maintained indoors with sufficient daylight (but not sunlight) and kept warm (28°C). Cultures should not be covered if populations of maximum concentration are desired.

Aeration is helpful as the bacterial growth will consume most of the oxygen in the culture, inhibiting the growth of the infusoria. Aeration will also help to reduce any unpleasant odours produced by the decaying organic matter. Within a couple of days the water should turn cloudy with bacteria. Once the culture begins to clear a pale, ever changing cloud of tiny specks will be seen when a clear glass jar containing the infusion is held to the light. Although just visible to the naked eye, to see infusoria properly, a microscope is needed. At this stage you can begin feeding them to your fry.

When the culture is thriving well add a few pieces of vegetable matter, which will provide organic matter when the original culture is nearing exhaustion. Beef extract powder, lettuce leaves, cereal grains (rice, wheat, barley, oats, etc.), banana skin, hay, grass, skim milk powder, decaying aquatic plants and leaves, cabbage leaves, and mud are some of the materials which are reported to have been used.

Such diverse organic materials may be boiled in water, or they may be allowed to macerate. Slight variations of the above techniques require the introduction of algae, yeast, or bacteria into infusions such as the above. The culture will decline rapidly if the nutrients are completely used up. However, care should be taken with the amount added as too much could result in the water becoming depleted of oxygen and cause your infusoria to perish. Culture water that is too rich in decomposing organic matter will simply encourage a bacterial bloom and an unpleasant odour may develop. Generally the culture will settle down again once the bacteria do their work. You could also use one of the commercial solutions devised expressly for feeding paramecia.

A healthy culture should be clear and somewhat odourless. A thriving culture can be maintained for months, but it's always a good idea to start fresh cultures occasionally, because nothing crashes more unexpectedly than populations of infusoria. Once you have a good culture going it is a simple matter to prepare a series of cultures from the original one.



### Feeding Infusoria

Infusoria are an excellent primary food source for newly hatched rainbowfish larvae. You can collect the infusoria with a syringe and feed part of the culture solution directly to the larval fish. Feed the larvae, according to their number, as often as possible in small quantities so as not to pollute the water. You should be able to tell when they are getting enough to eat because their stomachs will be distended.

The best way to collect infusoria and avoid adding any of the culture solution into the raising tank is to use a laboratory grade filter paper placed over a funnel and into an empty collection container. Pour the contents of the culture through the filter paper. The filtered medium will collect in the container and the infusoria will be concentrated on the filter paper. The infusoria are then washed off the filter paper with fresh water and fed to the fry. Return the filtered medium back to your culture container, as the culture will continue to reproduce.

If it is desired to feed the culture to the fry over a predetermined period of time the following method can be used (**Fig. 1**): Drill a small hole in the screw-on lid of a clear plastic food container, the size of some small diameter plastic tubing. Push the tubing through the lid until it is about a centimetre off the bottom of



the container. Fasten the tubing to the outside with some sticky tape. The container is then filled with infusoria culture and placed on the lid of the aquarium with one end of the tubing dangling over the surface of the aquarium. The culture is transferred into the aquarium by syphoning and the flow is regulated with an aquarium clamp. By leaving the end of the tubing, on the inside of the container, just above the base less detritus is picked up.

You can add some water-soluble vitamins to enhance the nutritional value of the infusoria a few hours before feeding. They will absorb them directly from the water. Most vitamin solutions designed for human babies generally contain all the necessary vitamins fish require.

As an alternative feeding system you could use one of the commercially available AquaDose<sup>®</sup> systems, or medical intravenous infusion bag. These can also be used for feeding greenwater infusions, medications or any other aquarium additives.

AquaDose and Intravenous Infusion Bag ▼(photo: Gary Lange)



### Phytoplankton

Phytoplankton (green-water) is composed mainly of microscopic free floating or suspended algae and is generally referred to as greenwater by aquarium hobbyists. The word phytoplankton is derived from the Greek language (phyto = plant; plankton = wanderer). It is a term used to describe plants that are so small that their movement is primarily controlled by the motion of the water. These plants called algae (alga = singular) include a number of microscopic, single and multiple cell forms. In freshwater, large numbers of phytoplankton often occur in lakes, rivers and ponds. Various phytoplankton species bloom in response to certain conditions such as changes in temperature, photoperiod, light intensity and nutrients.

The majority of phytoplankton is made up of holoplankton, organisms that spend most of their life cycle in the planktonic community. However, many phytoplankton species are capable of producing resting spores, which can to be found in deeper water or in the bottom sediment of freshwater habitats. These 'resting stage' spores are generally what cause the algal blooms that we often see in freshwater environments. Phytoplankton is usually rich in green algae. However, it also includes diatoms, flagellates and blue-green algae are actually bacteria and not algae.

The diet of most rainbowfish larvae consists of a wide diversity of phytoplankton and zooplankton organisms that are found in great abundance in their natural habitats. This abundance and diversity of food organisms of different sizes and nutritional composition usually provide all the dietary requirements of the feeding larvae. Phytoplankton contains proteins, starches, fatty acids and oils and forms the base of the food chain in aquatic environments. As such, it is an excellent primary food source for newly-hatched rainbowfish larvae.

Although several alternatives for feeding rainbowfish larvae exist, such as commercially available fine powder-based feeds, live greenwater is still the best and the preferred food source. Most phytoplankton is very small, often-minute organisms ranging in size from 2 to 100  $\mu$ m and stay suspended in the water column. Phytoplankton also serves as food for zooplankton, which in turn are fed upon by the fish larvae. Besides, rearing rainbowfish larvae using the "greenwater technique" directly in the larval tanks is believed to play a role in stabilising the water quality. It would appear that phytoplankton can use up a lot of nutrients, and, instead of being over supplied with nutrients; an aquarium with greenwater is more often low in nutrients, at least in nitrogenous substances.

Phytoplankton species can vary significantly in their nutritional value, and this may also change under different growth conditions. Most phytoplankton contain 20–25% protein, 5–30% carbohydrate and 5–25% lipid. Phytoplankton is rich in essential amino acids and polyunsaturated fatty acids. Phytoplankton also provides vital bio-pigments that are required for the development of improved colouration for captive bred rainbowfishes, and can also be a rich source of ascorbic acid (0.11–1.62% of dry weight).

Cultivation of phytoplankton for feeding rainbowfish larvae is very simple. All that is required is water, light and nutrients. Natural sunlight is best, if not mandatory, as without strong light the phytoplankton will simply not grow. Phytoplankton use energy from the sun to make their own food through photosynthesis. In the process of photosynthesis, plants use carbon dioxide, water, nutrients, and sunlight to produce oxygen, sugar and energy. The sugar molecules then combine to form starch and cellulose, energy-rich organic molecules that are food for the plants. One of the critical chemicals for photosynthesis is chlorophyll, which is bright green.

Phytoplankton does not appear overnight, it may take a week or two to appear. However, once you have a culture established, they will bloom much quicker when you use some old greenwater to start a new culture, often within 3 days. Under favourable conditions, phytoplankton grows continuously by a process known as cell division. Each cell enlarges and divides into two daughter cells that subsequently grow and divide yielding a culture that increases exponentially (e.g., 8, 16, 32, 64...etc.). Growth slows as the algal population becomes more crowded. Nutrients are depleted, metabolites build, and light penetration decreases because of self-shading. The culture will then go into a stationary phase for the current conditions and will not increase in density. Phytoplankton contain their best nutritional value when their growth is still within the "exponential growth" phase.

Perhaps the simplest way to culture phytoplankton is just to place some old aquarium water (that hasn't been chemically treated) from one of your waterchanges into a suitable container and store it outdoors. When it turns green you can then collect a portion of the culture each day to feed to your fishes. Outdoor cultures should be protected from heavy rain and screened to prevent entry of predacious aquatic insects. Filamentous algae and predators of fish larvae can be troublesome in outdoor cultures.

One of the problems with outdoor cultures is that you have no control over the sunlight. Generally, the more light the greater the phytoplankton density. While phytoplankton will grow all year round in one region, they may not grow in all regions. Their light requirement remains the same even when the days are shorter and the air temperature is colder. Another requirement for successfully growing phytoplankton outdoors is the shape, size and colour of the culture container.

Deep containers need more light than shallower ones. The deeper containers get less light penetration and therefore fewer algae will grow. Shallow white or clear translucent plastic containers are the best. They allow the sunlight to penetrate most of the water column. Decreasing the depth of the container can have the same effect as higher light intensity. Shallower containers require much lower light levels initially. You could use a standard all-glass aquarium. Water circulation can be also be used to keep the phytoplankton in suspension, thus exposing as much surface area as possible to the sunlight.

If on the other hand you want to culture your phytoplankton indoors, then the best way is to use a bare 50-litre aquarium placed near a window that receives several hours of natural sunlight each day. Fill the tank with old aquarium water and it should turn green within a few days (depending on light and nutrients). Mechanical circulation can be used to keep the algal cells in suspension, thus exposing as much surface area as possible to the light for photosynthesis, while making sure they don't remain in the same spot too long for photo-inhibition to become a factor. Cultures are generally maintained at a temperature range of  $20-24^{\circ}$ C, *p*H 7.5–8.5. High-density cultures may require a *p*H buffer to prevent *p*H drift.

Unless you are maintaining pure cultures in sterile conditions, your culture will have both phytoplankton and zooplankton in greater or lesser degree. Cultures maintained indoors without sufficient light will probably contain more zooplankton than phytoplankton but outdoors the situation is usually reversed. Introducing more light will encourage the existing phytoplankton to grow.

Providing phytoplankton with zooplankton as food for the newly hatched rainbowfishes has several advantages. The larvae are easily able to switch to different sized prey, a feature not present in monocultures of organisms. Phytoplankton also enables the resident zooplankton to feed on the algae and microbes, thus retaining their nutritional value for greater periods of time. Studies performed in aquaculture have shown that fish larvae fed on diets composed of multiple phytoplankton species have higher survival rates and quicker growth rates than those fed with a single type of phytoplankton.

Feed 5–10 ml (per 20–50 fry) of phytoplankton suspension three or four times per day. You can tell if they are feeding well as the larvae should have nice "swollen stomachs" after feeding. As the fish larvae grow, the amount of food they need increases and they prefer to eat larger-sized prey. Larvae older than 14 days can start to be fed live brine shrimp nauplii and/or microworm. Brine shrimp and microworm will accelerate growth so it is important to commence feeding as soon as possible. The smallest larvae will continue to eat the greenwater while the larger ones will start feeding on the brine shrimp nauplii or microworm. Gradually increase the proportion of brine shrimp nauplii or microworm and phase out the phytoplankton - this should be around day 14 to 21. This feeding regime should generally result in greater than 90% larval survival.

Alternatively, commercially available phytoplankton products such as Phytoplan<sup>®</sup> can be incorporated into their diets. Phytoplan is a spray dried blend of several strains of phytoplankton. However, research has shown clear difference in nutritional value between non-living and living diets, and among commercial diets advertised as containing live algae. Overall, results showed that fresh cultures displayed the best growth and lowest mortality rates. Also, a mixed algal diet promotes faster growth and higher survival than single algal diets. Results also suggest that phytoplankton present in some commercial diets may lose their nutritional value during processing or refrigerated storage.



### Semi-intensive Green-water System

If you have a large breeding program you could set up a bank of 50 litre aquariums as raising tanks for rainbowfish larvae and simply place the spawning mops containing eggs into the phytoplankton tanks for hatching. The configuration of the system is simple and maintenance needs are minimal. The culture needs to be very thick whereby the bottom of the tank cannot be seen through the green water when looked at from above. When the eggs hatch the larvae will survive and grow without any additional help from you. As the fry grow you can start feeding brine shrimp nauplii and other foods until they are large enough to be moved to a more suitable environment.

Culture of phytoplankton indoors allows control over illumination, temperature and nutrient level. The most crucial part is getting the correct amount of light and nutrients. Strong light to simulate the outdoor brightness is required. The duration of artificial illumination should be a minimum 18 hours of light per day. Make sure the lamp's output is "daylight", (6500 Kelvin). A compact florescence lamp (55 watts, 7000 Kelvin) should give you sufficient light if the tank is not too large or deep (due to light penetration issue). Use the following rule: 0.7 watts per litre of water at a minimum of 6500 Kelvin full spectrum white light for 8 hours per day. The minimum wattage required is 55 watt. Lights can be purchased according to the size of the aquariums that are to be used for culturing.

Tanks are filled with dechlorinated water then fertilised and allowed to stand until the water turns green. Organic fertilisers (manure) are usually preferable to chemical fertilisers. Chemical fertilisers may be used and usually work better in earthen ponds than in tanks or tubs. Organic matter will produce bacterial microbes and detritus as well as phytoplankton as food for the larvae.

The fertiliser can be added to your culture in several ways. One is to soak the dry material in water until it breaks down into a thin mixture, then pour the resulting concoction into the tank, allowing it to slowly deteriorate. Another is to place the dry material in a mesh bag and suspend the bag inside the tank. Nylon stockings work well for this purpose. The use of a bag controls the organic matter and prevents suspended particles from being a problem. It also allows greater control of fertilisation. If fungus occurs on the bag containing the organic material it should be removed from the culture.

During the first 21 days of the larval culture period, water within the culture tank should be static with light aeration, just enough to gently keep the phytoplankton moving within the water column. Water exchange is not required with the greenwater technique and water only needs to be replaced due to losses from evaporation.

The presence of phytoplankton in the tank will maintain good water quality by taking-up the excess nutrients produced in the system. Phytoplankton helps the maintenance of proper water quality through their ability to cycle nutrients (such as nitrates and phosphates). Phytoplankton are also able to utilise the wastes produced by the young larvae (ammonia, urea), converting them into non-toxic forms, and also play an important role in regulating the *p*H of the water through their removal of excess carbon dioxide.

In a green-water system most of the nutrients are recycled back to the fish. With light aeration using an airstone, detritus, faeces and plankton can be kept in constant circulation. Nitrifying bacteria will colonise this floating "substrate" creating a "suspended growth treatment process". These bacteria oxidise toxic ammonia nitrogen into relatively harmless nitrate and heterotrophic bacteria (bacteria that consume organic matter) proliferate. Rainbowfish larvae will feed on these and receive supplementary nutrition.

The suspended growth treatment process maintains adequate water quality for the fish while recycling waste nutrients into phytoplankton and bacteria that encourages continued algae and bacterial population growth, which further improves water quality. An added advantage of this process is the elimination of the need for a fixed-film biofilter.

Once the larvae are added the whole system can be managed according to the densities of phytoplankton and water quality. Water quality within the green-water system tank should be monitored on a regular basis, with water replacement performed if ammonia or nitrite levels approach 1-ppm levels. Tank bottom cleaning using narrow-gauge siphon tubes should be conducted on an as-needed basis to remove fungused eggs, dead larvae, etc.

Larval stocking density should approximate 20–30 animals per litres of culture water. Stocking densities higher than this will require more attention to water quality parameters and will likely result in higher larval mortality or wide size variation among juveniles. Stocking densities below 20 larvae per litre may result in better overall growth and survival. Larvae will grow more quickly and show fewer deformities at lower stocking densities than at higher densities. However, stocking density is dependent upon the number of fry required, and the quantity and quality of the cultured phytoplankton.

The only problem you are likely to face raising rainbowfishes in green-water system tanks is that there is the risk of the culture suddenly 'crashing' (failing), dying off as suddenly as it appeared. The phytoplankton simply consumes all of the nutrients in the water and then collapses and the larvae are left with nothing to eat. If this happens, it is important to vacuum out all of the dead algae from the bottom of the tank as soon as possible and start feeding an alternative diet. Otherwise, this organic material will consume substantial amounts of dissolved oxygen from the water as it decays and in doing so can kill all the fry. Therefore, careful observation of the tank is required at all times. The whole concept of the "green-water technique" is to mainly get the larvae through their early life stages until they can start feeding on larger foods, which is easier to provide.

#### Artificial Green-water

Mix a suspension of 1 level tablespoon spirulina powder per litre of distilled water and fed at a rate of about 10–50 ml per 20 litres - but only if the water has cleared from any previous feeding. The amount of the powder is not so important that you need exact measures. It should not be so much that it won't go into solution and should be enough to make the mixture a dark green colour.



### Microalgae Feed Supplements

Microalgae are an important food source and feed additive in the commercial rearing of many aquatic animals. Microalgae such as *Dunaliella salina*, *Chlorella spp.*, *Haematococcus pluvialis*, *Spirulina platensis* and *Spirulina maxima* are used as a source of natural pigments for the culture of prawns, commercial fish species and ornamental fish. The use of algae in aquaculture is not surprising as algae are the natural food source of these animals. Microalgae are furthermore used to produce mass quantities of zooplankton (rotifers, copepods, and brine shrimp) which serve in turn as food for larval and early-juvenile stages of fish.

Although several alternatives for algae exist such as yeasts and microencapsulated feeds, live algae are still the best and the preferred food source. However, all algal species are not equally successful in supporting the growth and survival of rainbowfish larvae. Mixed algal diets promoted faster growth and higher survival than single algal diets.

Other studies have confirmed the feasibility of using dried algae, and *Spirulina* appears to be the preferred species. Longer-term trials have shown that *Spirulina* was nutritionally superior to all of the diets tested. Spirulina powder is an excellent source of food for fish and shrimp larvae. In some cases, the addition of relatively small amounts of algae to prepared diets has been observed to result in significant improvements in growth and pigmentation. Thus, microalgae have a clear role in dietary supplements to fulfil specific nutritional requirements.

Rainbowfishes are recognised for their bright, brilliant and beautiful colouration. The body colours of rainbowfishes are predominantly dependent on the presence of special cells located in the dermis of the skin, above or below the scales, called chromatophores. Colour changes result from chromatosomes concentrating in the centre of the chromatophore or dispersing throughout the cell. Iridiophores contain highly reflective guanine crystals. The crystals act as mirrors, which reflect the colours of the outside environment. Iridiophores are often responsible for the bluish or silvery appearance of many fishes.

The colouration of rainbowfishes often fades when maintained under aquarium conditions. Therefore, one of the greatest challenges for the aquarium hobbyist is to accurately replicate the natural colouration of rainbowfishes that are maintained in captivity. Most of the naturally occurring colour pigments are derived from phytoplankton, zooplankton and photosynthetic bacteria consumed in their natural diet, and these are not normally available in captivity, as they are mainly dependent on artificially prepared diets. Colour intensity can also be related to the environment and will often decrease when the fish are stressed, or from the nature of captivity itself.

Keeping and raising rainbowfishes in outdoor ponds with or without supplementary feed will enhance their colour development, most likely due to the natural food items in the pond. Many aquatic food organisms ingest astaxanthincontaining algae and plankton as a major part of their diets. Carotenoids are the most common natural pigments responsible for many of the bright colours found in fish and crustaceans in their natural environment, as well as a variety of biological functions. More than 650 different naturally occurring carotenoids are known. Carotenoids commonly occurring in freshwater include beta-carotene (orange), lutein (greenish-yellow), astaxanthin (red), tunaxanthin (yellow), doradexanthin (yellow), and zeaxanthin (yelloworange).

The importance of carotenoids in fish diets has been stressed by several researchers working in the field of fish nutrition. Carotenoids are a group of naturally occurring lipid soluble pigments that are produced primarily in phytoplankton. These pigments are responsible for the great diversity of colour seen in nature. While plants and algae can synthesise these pigments, animals are unable to produce them naturally and must obtain them from food.

Rainbowfishes are not able to synthesise carotenoids and depend completely on the presence of the necessary carotenoids in their diet. In many cases, the level of colouration is dependent on an individual's foraging success. The level of colouration can also indicate their quality as a potential mate. Therefore, rainbowfishes maintained in captivity should be fed a complete diet that includes a colour enhancing agent, such as astaxanthin, to supplement natural feeds that might be limited in captivity. Typical commercial diets used for aquarium fish are very low in total carotenoids.

It has been recommended that feeds containing colour enhancers or pigments such as astaxanthin should be fed to rainbowfishes to bring out their full colouration. Duckweed has high concentrations of carotenoid pigments, particularly beta-carotene and xanthophyll that make duckweed an especially valuable supplement for rainbowfishes.

Feeding *Chlorella vulgaris* and *Spirulina spp.* also produces positive pigmentation results in rainbowfishes, and the microalgae *Chlorococcum* seems to be a promising source of astaxanthin, canthaxanthin and adonixanthin. These algae have strong concentration in particular carotenoid pigments.

It is worth noting that aside from giving an enhanced colouration, microalgae sources could provide better growth performance as has been shown in a number of studies.

Carotenoids are vital nutrients for healthy growth, metabolism, and reproduction. Male rainbowfishes use colouration to produce sexual signals associated with breeding. It is well known that the most highly coloured males have the best chances of finding a mate.

One reason why females are not usually highly coloured is that they need to pass on their carotenoids to their offspring. At the time of sexual maturation, they mobilise stored carotenoids to the ovaries and finally, on to the progeny. This active transfer of carotenoids from the female to the eggs has led to the hypothesis that carotenoids are vital for larval survival and growth.



Experiments adding algae to the diet of *Pseudomugil furcatus* over a period of three weeks resulted in enhanced colouration. They became significantly more intensely coloured when fed a diet containing 1.5–2.0% of a carotenoid-rich strain of *Spirulina platensis* and 1.0% of a specially grown *Haematococcus pluvialis*. Colour enhancement was apparent after only one week, when the fish consumed these doses of algae; lower doses (0.5% and 0.4%, respectively) were not significantly different. Both treatments were significantly more effective than control treatments with no added algae. It appears, however, that *Haematococcus pluvialis* spores do not perform as well as commercially available synthetic forms of astaxanthin.

Research has also shown that the addition of astaxanthin can enhance the colouration of all rainbowfishes. Several species responded positively, and maintenance of the bright colouration was achieved with incorporation of 25 mg/kg astaxanthin into various diets on a continual basis. Some species also had faster growth rates. However, more research is required to establish the optimal level of astaxanthin to use in rainbowfish diets.

Commercially available alga products (e.g., NatuRose<sup>®</sup>) can be incorporated into their diets. When these alga products are incorporated into the diet at  $1\sim2\%$ , an enhancement of coloration in the treated fish may be noticeable within two weeks. NatuRose algae meal is a natural source of astaxanthin derived from the microalgae, *Haematococcus pluvialis*. It is a spray-dried, dark red powder, and is currently used worldwide as a coloration and nutrition source for numerous species of animals. The flakes of *Haematococcus pluvialis* contain the largest concentrations of astaxanthin found in nature (some 40,000 parts per million).

*H. pluvialis* is a freshwater alga which normally grows in temporary water bodies. The widespread occurrence of *Haematococcus* in temporary rather than permanent bodies of water is due, at lease in part; to the fact that such pools are usually free of other competing algae. The astaxanthin is produced in a thick walled resting stage, the aplanospore, whereas maximum growth occurs in the green thin-walled flagellated stage. This necessitates a two-stage culture process, one optimised for biomass production and the other for astaxanthin production.

Commercial production of astaxanthin from the freshwater microalgae *H. pluvialis* is a growing business worldwide, primarily due to its high astaxanthin content. However, this alga exhibits some unfavourable characteristics like its slow growth rate and complex life cycle, when compared to other microalgae successfully cultivated on a commercial scale like *Dunaliella* and *Spirulina* species.

Other commercial ventures for natural astaxanthin production utilise fermentation of the yeast *Xanthophyllomyces dendrorhous* or extraction of the pigment from by-products of crustacea such as the Antarctic krill (*Euphausia superba*). In addition to production from natural sources, astaxanthin may be chemically synthesised, and synthetic astaxanthin is the major form currently being used in commercial fish feeds.



#### **Culturing Algae**

Algae culture begins with a pure stock or starter culture of the algal species desired. These can be obtained from a number of sources. Commercial biological supply houses often sell algal cultures, but the species may not be suitable for raising rainbowfish larvae. Aquacultural suppliers are perhaps the best method of obtaining algae cultures.

Once obtained, starter cultures (usually transported in test tubes) are used to inoculate several new cultures. Some of these are kept as stocks for when an old culture dies, is harvested, or otherwise lost and must be restarted. The rest are used to inoculate progressively larger vessels until there is enough culture to start mass production tanks. The culturist must supply light, aeration, relatively stable temperature control, and sterile water with nutrients (media) to produce successful algal cultures. By avoiding major contamination from unwanted algal species and microscopic predators, a continuous, dependable supply of high quality algae will be available. However, pure cultures can become contamination by unwanted algae, bacteria, and predatory protozoans, and is a problem that cannot always be controlled successfully. Cultures are easily contaminated from non-sterile containers or splashed water from buckets, hoses, or hands.

Production of pure algae cultures is accomplished by providing a favourable environment for the species being cultured. As with all plants, microalgae photosynthesise, i.e. they assimilate inorganic carbon for conversion into organic matter. Light is the source of energy which drives this reaction and in this regard intensity, spectral quality and photoperiod need to be considered. Light intensity plays an important role, but the requirements vary greatly with the culture depth and the density of the algal culture: at higher depths and cell concentrations the light intensity must be increased to penetrate through the culture (e.g. 1,000 lux is suitable for Erlenmeyer flasks; 5,000–10,000 is required for larger volumes).

Light may be natural or supplied by fluorescent tubes. Too high light intensity (e.g. direct sunlight, small container close to artificial light) may result in photo-inhibition. Also, overheating due to both natural and artificial illumination should be avoided.



Fluorescent tubes emitting either in the blue or the red light spectrum should be preferred as these are the most active portions of the light spectrum for photosynthesis. The duration of artificial illumination should be minimum 18 hours of light per day, although cultivated phytoplankton develops normally under constant illumination.

The optimal temperature for microalgae cultures is generally between 20 and 24°C, although this may vary with the composition of the culture medium, the species and strain cultured. Most commonly cultured species of microalgae tolerate temperatures between 16 and 27°C. Temperatures lower than 16°C will slow down growth, whereas those higher than 35°C are lethal for a number of species. If necessary, algal cultures can be cooled by a flow of cold water over the surface of the culture vessel or by controlling the air temperature with refrigerated airconditioning units.

"Abundant in nature, green signifies growth, renewal, health, and environment."

### Copepods

Copepods are small crustaceans that occur naturally in all aquatic habitats and are one the major food items of larval rainbowfishes in their natural habitat. Copepods are aquatic crustaceans, smaller relatives of the crabs and lobsters. They have colonised virtually every habitat from 10,000 metres down in the deep sea to lakes 5,000 metres up in the Himalayas, and every temperature regime from sub-zero polar waters in the Antarctica to hot springs. Copepods are found in marine, estuarine and freshwater environments and also inhabit the terrestrial realm occurring in mosses and leaf litter. The majority are free-living and benthic, although there are a few pelagic and symbiotic species. This freeliving form is only part of the copepod success story, since they have also become parasites of almost every major animal group from sponges and corals to fish and mammals. Copepod's that parasitise fish skin and gills, for example, are serious pests of commercial fish farms.

Copepods exhibit a diverse range of body shapes, but in general they are linear and range in length from 0.2 to 28 mm, but most planktonic forms are between 0.5 mm and 2.5 mm. They represent a significant component of the meiofauna in freshwater habitats. The word 'meiofauna' is used to define an assemblage of benthic invertebrates smaller than the macrobenthic fauna. Prior to the introduction of the word meiofauna, researchers had referred to small invertebrates as microfauna; however this term now refers largely to Protozoa. Meiofauna are defined as animals that pass through a 500  $\mu$ m mesh sieve but are retained on 45  $\mu$ m mesh.

Copepods feed on diatoms, bacteria and protozoans. Food availability and water temperature are thought to be the prime factors influencing their reproduction and growth. Reproduction takes place through copulation and involves the transfer of sperm from the male's spermatophore to the female oviduct. Prior to reproduction a male copepod will grasp a female with his modified antennules. Coupling arrangements tend to be species specific.

Males may grasp the third or fourth legs of the females, the caudal rami, the caudal setae, and the posterolateral margin of the cephalothorax or around the genital double-somite. Precocious coupling of males and juvenile females is also common. In these situations, a male attaches himself to a juvenile female and stays with her until her final moult, when she is ready to mate. Males have been observed to release their hold on moulted exoskeletons of copepods, and then reclasp the body of the newly matured female. This behaviour ensures that the 'trailing' male is the first to mate with the female when she reaches maturity. Post-copulatory coupling has also been observed in some copepods and this is thought to prevent subsequent matings of the female with other males. Copepods carry their eggs in either a single egg sac or a pair of sacs, but many pelagic species release their eggs directly into the water column, since the carrying of egg sacs renders the adult more liable to visual predation. After an incubation period of one to eight days eggs hatch, and nauplii are released.

In general, copepods go through six naupliar stages and six copepodite (post-naupliar) stages, the last one being the adult form. The time this cycle takes varies and developmental rates are influenced by temperature, food supply and salinity. Some copepods reach maturity in just six days after hatching; while others can take up to 62 days. The early life stages are very tiny and can be as small as 0.05 mm. Males are smaller than females.







### Rotifers

Rotifers are valuable live food for feeding the larvae of most fish species because of their very small size. In addition, rotifers swim slowly and stay suspended in the water column, thus being available for easy capture and consumption by larval rainbowfishes and blue-eyes. They can tolerate temperatures of between 15 and 31°C and *p*H 6.0–8.0. Rotifers can also be used as a conditioning food to induce smaller adult species, such as *Iriatherina werneri*, to spawn.

The mouth size of first-feeding larvae usually restricts the size of the food particles which can be ingested. In general, mouth size is correlated with body size, which in turn is influenced by egg diameter and the period of endogenous feeding (i.e., yolk sac consumption period). There are few, if any, published information on the mouth size of rainbowfish larvae. Rainbowfishes larvae typically have a small mouth size and limited yolk reserves and therefore are dependent on the presence of abundant microinvertebrates or algal food at the time of first feeding.

The major difficulty for the aquarist is providing organisms appropriate to the size of the larvae at the first feeding stage and then supplying the large numbers necessary to maintain them. The preferred size of prey for larval fish increases as mouth size and feeding competency increase and different types of live foods need to be cultured for the different stages in the larval development. For example, different species of microalgae range from  $2\sim100 \ \mu\text{m}$ ; rotifers from  $50\sim200 \ \mu\text{m}$ , copepods from  $100 \sim 300 \ \mu\text{m}$  and brine shrimp nauplii  $400 \sim 800 \ \mu\text{m}$ . Apart from these main groups, a few other live feeds are used on a more limited scale including microworm (*Panagrellus redivivus*), vinegar eels (*Turbatrix aceti*), and small cladocerans. This group includes many species (*Daphnia, Moina* etc.). They can reach up to 4–6 mm but typically are much smaller than this; the smallest species is around 250  $\ \mu\text{m}$ . Larger juveniles and even adults, will feed on these crustaceans.

Most rainbowfish larvae are not particular about the types of live food they will eat, but the animals must be small enough for the larvae to ingest. Rainbowfish larvae are usually small in size (2–5 mm, total length), have poorly developed eyes, do not swim well but are mostly present in the water column, and require easily digested food. Having such tiny mouths, the size of the food is crucial to their surviving the most difficult period in their lives.

Rotifers are microscopic animals ranging in size from 50~ 2500  $\mu$ m and found in aquatic habitats worldwide. They inhabit freshwater streams, lakes and ponds, brackish water



and, to a lesser extent, salt water but they are predominantly freshwater inhabitants. They can be recognised by the presence of a crown of cilia at the anterior end, which is used for feeding and/or locomotion. However many species spend the majority of their lives attached to a host such as plants, rocks or even other cladocerans. In some forms the beating of the cilia, which are arranged around the edge of one or more disc-shaped lobes, give the appearance of a revolving wheel - hence the name Rotifer from Rota, Latin for wheel.

Rotifers are tremendously varied and the cilia rotifers use for feeding and locomotion can vary enormously. Some species don't use it for locomotion but have developed very special capturing devices. The genus *Collotheca* live attached to a substrate and capture bacteria with their elongated cilia. For feeding rainbowfish larvae, the freeswimming kind is the only useful ones.

Rotifers are filter-feeders and feed by moving food particles into the mouth through the action of the coronal cilia. Size of the particles varies, but most are small ( $<25 \mu$ m). Feeding is related to food size and shape and consists mostly of algae. Some species seize and ingest whole prey or puncture the cell or body wall and suck out the contents. *Asplancha*, the largest rotifer, preys on algae, other rotifers, and small planktonic crustaceans and has the ability to alter its size in response to changes in size of food particles.

Most natural populations of rotifers are composed primarily of females that produce eggs, which are capable of development without fertilisation. This development is called 'parthenogenesis' and occurs in other freshwater crustaceans, such as daphnia. Parthenogenetic amictic females are diploid and produce amictic<sup>1</sup> eggs that develop further into amictic females. There may be up to 20–40 amictic generations before sexual reproduction occurs. Egg development time is about 1 day under warm optimal conditions, so populations of amictic females can develop rapidly in 2–5 days under good growing conditions. This seems to be the main advantage of asexual reproduction.

As habitat conditions alter, some of the females lay eggs, which are smaller than and differ in other ways from the usual female-producing eggs. Factors that can trigger this are not clear. Stimuli appear to be species-specific and include high population of amictic females, food availability, accumulation of wastes, temperature changes, etc. Research has shown that when populations are fed paramecia, mostly amictic females are found. However, low densities of algae lead to a high proportion of mictic<sup>2</sup> females.

If not fertilised, these smaller eggs hatch into males. Males are capable of copulating within an hour of hatching. The males then fertilise the females, after which fertilised eggs are laid. These are distinguished from the parthenogenetic ones by a hard thick shell. These 'ephippia' contain embryos in a state of arrested development (diapause) that can withstand drying, freezing, and other unfavourable conditions. They remain in this state and will not complete their development and hatch until favourable conditions return, which may be several years. Because of the capacity of these eggs to resist drought, rotifers can live in places that are only temporarily wet. As soon as moisture appears they hatch into females and swim about and feed actively. Resting (diapause) eggs always produce parthenogenetic amictic females. Reproduction rates are related to the quality and quantity of food and temperature. Rates are generally lower if food volume is insufficient or of poor quality. Temperature affects the rate of egg development, metabolism, feeding, movement, longevity, as well as reproduction.

Most species are adapted to specific temperatures at which they exist best. Some are stenothermal (tolerate only a narrow range of temperatures) others are eurythermal (tolerate wide range of temperatures). Perennial species commonly exhibit maximum densities in early summer in temperate region. However, some species are seasonal, with some that develop greatest population densities in winter and early spring, and others that reach a peak in summer, often with two or more peaks, especially in late summer and often in conjunction with the development of blue-green algal species.

### Culture

Rotifer eggs have been commercially available for a number of years and their use would eliminate the need to maintain culture stocks. Both the saltwater rotifer (*Brachionus plicatilis*) and the freshwater rotifer (*Brachionus calyciflorus*) are available commercially and sold as Resting Rotifers<sup>®</sup> from aquaculture or aquarium suppliers. Resting Rotifers are actually lab-cultured resting eggs, and are ideal for either direct feeding after hatching, or setting up starter cultures. *Brachionus plicatilis* will survive for several hours after transfer to fresh water, and are comparable to brine shrimp. *Brachionus calyciflorus*, in contrast to *Brachionus plicatilis* will stay alive in freshwater and do not sink to the bottom but stay in the water column until eaten. Any excess can be stored in the refrigerator for up to a week.

When placed in a suitable culturing environment, hatching commences over a period of 24 to 36 hours. Rotifers begin reproducing 18 hours after hatching and will continue producing eggs every 4–6 hours if properly maintained and fed. The diet of rotifers most commonly consists of dead or decomposing organic materials, as well as unicellular algae and bacteria. Cultures should be aerated and generally maintained on algae concentrate. Feed with a small amount of green-water from either concentrated algae (e.g., Instant Algae<sup>®</sup>) or live algae. They can be harvested from 7 to 10 days after hatching with a fine net (50  $\mu$ m) and fed to the fishes. Keep about a quarter to start a new culture.

Most rotifers are indiscriminate filter feeders and will feed on algae, yeast, bacteria and micro-particles up to approximately 25  $\mu$ m in size. There are many different methods and adaptations used to mass culture rotifers. In general, rotifer production is based on either (a) a diet of marine microalgae and bakers yeast, or (b) a single diet of commercially produced fortified yeasts. Within each of these feed types, rotifers are cultured using either batch, semicontinuous or continuous methods.



In commercial fish hatcheries, rotifers are usually fed bakers yeast (Saccharomyces cerevisiae), or proprietary products as a food source. The amount of baker's yeast fed on a daily basis is about 1 gram per million rotifers. Although baker's yeast has a small particle size  $(5-7 \mu m)$ and a high protein content and acceptable as diet for Brachionus calyciflorus, it is not advisable to be used alone for rotifer culture. Rotifers raised on yeast alone often lack the essential fatty acids and vitamins required by larval fish species. Furthermore, rotifer cultures fed only baker's yeast are characterised by varying success and the occurrence of sudden collapses of cultures. Most probably the reason for these crashes was explained by the poor digestibility of the yeast, which requires the presence of bacteria for digestion. Commercially available diets including Culture Selco, Rotimac, DHA Selco, Protein Selco and Algamac. Ground shrimp meal, flour and rice bran are some other food sources that have been used for the cultivation of rotifers. The food should be run through a 100 µm sieve to obtain a suspended feed suitable for feeding the rotifers.

More rotifers can be produced from the same volume of water with a continuous culture system. However, continuous culture systems suffer from a build-up of waste products in the form of faeces and uneaten food, contributing to contamination problems with filamentous algae and fungal infections. It is imperative that continuous culture systems have some form of bio-filtration or organic waste removal system. The use of bakers yeast causes much of the water quality problems associated with continuous culture systems. It is important to note that feed rates should be based on the actual density of rotifers in the system and care should be taken not to overfeed. The culture container should be clear of algae before the next feeding to avoid excess algae accumulation. Any algae or food that is not consumed within 48 hours will degrade, increasing the level of ammonia and this reduces the dissolved oxygen level in the water. Optimal temperature of culture is usually 20-30° C with a pH of 7.5. To get a bloom, an entirely new culture can be started with a reasonable amount of feed/algae and subsequently harvested when it becomes concentrated with rotifers.

#### Brachionus plicatilis

Brachionus plicatilis is a euryhaline species being found in both brackish and saltwater environments. It is about one third the size of a newly hatched brine shrimp. They have been found in saline lakes in Australia with salinity levels almost twice that of seawater and also at a salinity of less than 1% of seawater. Geographic strains range in size from 90 to 320 µm in length. Hatching rates run at about 80% after 24 to 36 hours when hatched at a salinity level of 20‰ and a temperature of 28°C. Brachionus plicatilis is one of the rotifers which have become very popular for aquaculture. They can reproduce either sexually or as is more common, asexually. This organism is an excellent first feed for larval fish because of its small size and slow swimming speed and habit of staying suspended in the water column, thus being available for capture and consumption by fish larvae. Also they can be cultured in large numbers due to their high reproductive rate.

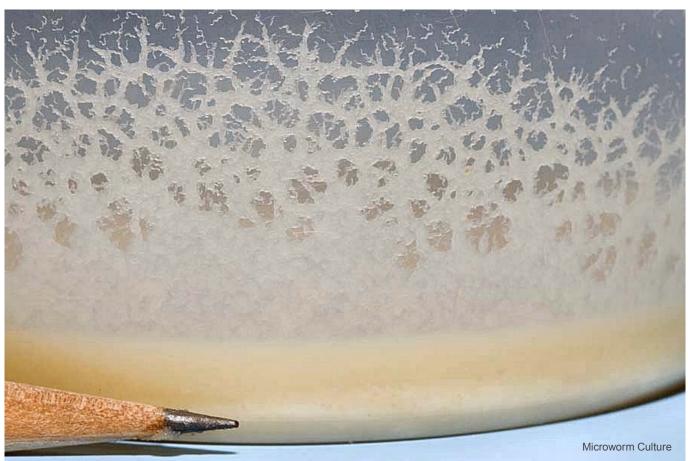
#### Brachionus calyciflorus

*Brachionus calyciflorus* is the most commonly cultured freshwater rotifer for both freshwater fish species and shrimps. There are several strains of different sizes of this rotifer, thus making them suitable for fry of a variety of sizes. With the increasing developments in larval rearing technology of freshwater fish species, demand for Brachionus calyciflorus is further increasing. Discus larvae are dependent on the body slime of their parents as a nutrient during the first two weeks of exogenous feeding. One study found that Discus larvae could be raised in the absence of the parent fish through feeding with *Brachionus calyciflorus* for 4 days (Days 4–7), followed by Artemia nauplii for a week (Days 8–14). Their growth and survival rates were comparable with those that rely on parental feeding.

<sup>1</sup> Amictic egg: A thin shelled diploid egg which cannot be fertilised. Instead, it develops by parthenogenesis. It is produced by rotifers when living conditions are optimal, and will develop to produce amictic females.

<sup>2</sup> Mictic egg: Pertaining to the haploid eggs of rotifers. If it isn't fertilised, the egg develops parthenogenetically into a male; if fertilised, mictic eggs secrete a heavy shell and become dormant, hatching in the spring into amictic females.





### Microworms

Microworms are well known to be an excellent food source for first feeding fish larvae. The species most commonly cultured in the aquarium hobby is believed to be *Panagrellus redivivus*, a member of the nematode family, Panagrolaimidae. The Panagrolaimidae family include opportunistic bacterial-feeders and some notable specialists of fermenting liquids or decomposing wood. I suspect, however, that there are probably a number of different nematode species being cultured in the aquarium hobby as "microworms".

The first Panagrellus species to be described is currently known as *Panagrellus redivivus*. It was described by Linnaeus (1767) as Chaos redivivus. This species was more commonly known as the 'sour paste nematode' in reference to its isolation from book-binding glue, or as described by Linnaeus in 1767, "habitat in aceto and glutine bibliopegorum". The generic name Panagrellus was not established until 1938, when Gerald R. Thorne described a nematode isolated from wounds of a cottonwood tree in Utah. Based on observation of new diagnostic morphological features, Thorne erected the genus Panagrellus, describing Panagrellus pycnus, as a new species. Controversy over the acceptance of the generic name Panagrellus over Chaos prevailed for many years. However, because Chaos redivivus had a rather vague description and no known type specimens, the name Panagrellus was eventually accepted based on modern taxonomic standards.

Panagrellus has a worldwide distribution, with species described from almost every continent except Australia and Antarctica. Currently 12 species are recognised, with *P. pycnus* as the type species and eleven other named species: *P. ludwigi*, *P. nepenthicola*, *P. silusioides*, *P. redivivus*, *P. redivivoides*, *P. ventrodentatus*, *P. dorsibidentatus*, *P. dubius*, *P. filiformis*, *P. ceylonensis* and *P. leperisini*. These species have been delimited and described using Linnaean or phenetic species concepts, based on morphological or morphometric data. Only the morphology of the spicules and occasionally the structure of the vulva in females are useful for diagnosis and identification of species. This lack of distinctive morphological features makes diagnosis of *Panagrellus* species rather problematic (Stock & Nadler, 2006).

*Panagrellus redivivus* is a small bacteriophagous, ovoviviparous, free-living (non-pathogenic) nematode found living naturally in soil. It moves using four longitudinal bands of muscle with alternate flexing and relaxation generating dorsal-ventral waves along the body that propel the animal along. The adult essentially comprises a tube, the exterior cuticle that contains two smaller tubes, the pharynx and gut, and by a reproductive system that takes up most of the animal. Neural structures include an array of sense organs in the head region which co-ordinate responses to smell, taste, temperature and touch. *Panagrellus redivivus* does not possess eyes, but will respond to light; of the 530 somatic cells of the female approximately 250 of them are neurons.





It is one of the few that do not lay eggs, but hatch juveniles internally. It has four larval stages before becoming adults. The first larval stage is intrauterine, but the remaining stages are free-living. It is gonochoristic, producing equal numbers of males and females (Stock & Nadler, 2006). Bacteriophagous nematodes are already known as a potential food source for fish larvae. Panagrellus redivivus is a nematode which is easy to rear in large quantities in culture. The worms feed on bacteria which are precultured. They have a short life cycle and a high fecundity. Panagrellus redivivus are a tiny nematode about 0.5 to 2.0 mm in length and 0.05 mm in diameter. They reproduce sexually and are livebearers; releasing 10-40 young every 5-7 days for a 26-36 day life span. The young reach sexually maturity in approximately three days. Their size increases by three times during the first day and five to six times during the next three days.

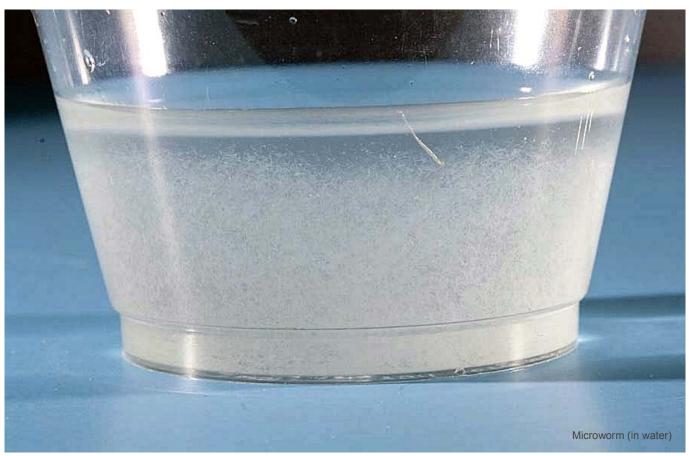
Microworms have been cultured by aquarists since the early 1930's as a live food for a variety of fish species. Their small size and ease of culture has received renewed attention in recent years with rising costs and declining hatch rates of brineshrimp eggs sold in the aquarium hobby. Microworm has as good if not better nutritional profile to that of brineshrimp, containing 48% protein, 21% lipids, 7% glycogen, 1% organic acids, and 1% nucleic acids. Approximately 70% of the lipids are fatty acids and the remainder is phospholipids.

Microworms are one of the simplest live foods to culture. When cultured under the right conditions they will multiply in vast numbers. They are a valuable live food and tolerant of environmental variables. Microworms like it warm and a temperature range of  $20-25^{\circ}$ C is about right. As the temperature begins to rise or fall below this range; their production rate will decline. However, they can maintain their life cycle at temperatures from 5°C up to and including 37°C. They have the added advantage of staying alive for six to eight hours in freshwater, by which time they should all have been eaten.

I have cultured microworm for many years and have tried several different culture mediums; bread soaked in beer, yeast blends, and a host of other foods. There are almost as many different culture methods for microworm as there are aquarists, each having their own successful anecdote. I will outline some of the more successful methods I have used. What you have to do is find one that suits your particular requirements. Starter cultures are available from biological supply companies or fellow hobbyists.

Microworm can be cultured in almost any shallow, flat, watertight container with a snug-fitting lid. This prevents contamination by insects and other bugs, and also prevents the culture from dehydrating. Small holes should be punctured in the lid for air circulation and the containers stored in a well ventilated room. Microworm should be able to be harvested daily for about 28–56 days using the same culture medium. However, it largely depends on the cultures running at the same time. Start your second culture about two weeks after the first. You may find that a culture will sometimes rapidly decline in production of worms. Having a second culture in production will ensure that you have worms available at all times.





Culture medium can be prepared from almost any grain flour, yeast, and water. However, research has shown that the type of culture medium used has a dramatic influence on worm yields. One such trial was conducted using three mediums - wheat flour, oatmeal, and cornmeal. Yield of worms in wheat flour was significantly greater than in oatmeal or cornmeal. Production of worms stopped after day 20 in cornmeal, day 33 in oatmeal, and day 53 in wheat flour.

The addition of yeast during initial media preparation was found to have no effect on worm yields. However, the addition of yeast on a weekly basis to the wheat flour medium gave a significant greater yield of worms than did untreated wheat flour. The wheat flour was mixed with water to form a smooth paste and placed in the culture container. After inoculation with live worms, the addition of 5 ml of a yeast solution, consisting of 7 gm bakers or brewers yeast (*Saccharomyces cerevisiae*) dissolved in 70 ml water; was lightly sprayed over the medium every 7 days. The addition of yeast should also inhibit the growth of nematophage fungi.

One method I have used is oatmeal (porridge). Use one part oats with one part of water. Place the mixture into the culture container and spread to a thickness of 15–25 mm and microwave on high setting for three minutes. The mixture is then allowed to cool to room temperature. Any media on the sides of the container should be removed with a damp cloth. After the mixture has cooled, place the starter culture on top of the porridge. Within 3–6 days you should see the surface moving. If you use a magnifying glass, you will observe hundreds of tiny worms.

You can increase the production of worms by sprinkling dry yeast powder over the surface of the mixture. You do not have to add the yeast until after about two weeks, then once a week should be sufficient. If the culture medium becomes very watery, you can add a slice of bread to the container to soak up the moisture. The addition of bread has a similar effect as does the bakers yeast.

Yet another method requires only a slice of white bread and brewers yeast. This culture method produced the best results for me in terms of the number of worms produced. Firstly, cut the crusts off the slice of bread and place it squarely on the bottom of the container. Mix 5 grams of brewers' yeast with <sup>1</sup>/<sub>4</sub> cup of water and pour the mixture onto the bread, making sure that the bread is completely saturated. It is important that there should be very little excess fluid in the container when the container is tilted.

Next add the starter-culture of microworm, by spreading it over the surface of the bread. Replace the container lid securely, and place the container in a warm area. Within three to four days, the culture should be thriving with worms migrating up the side of the container. As the bread is consumed another slice can be added to keep the culture active. After the addition of around 3 or 4 slices of bread the culture will need to be replaced. If the culture becomes too wet, more bread should be added to absorb the excess moisture. Remember the wetter the culture, the lower the production of worms.

During the warmer months of the year I often found another small worm in the culture as well. This is because the odour of



the culture will attract the common housefly, which lays its eggs through the small holes of the container lid. The eggs then hatch and the larvae develop and grow on the culture medium. This may seem a little unpleasant to some people, but these worms are ideal for larger rainbowfishes, which love them. Their development doesn't appear to have any detrimental effects on microworm production. Nuov (1995) reported very excellent growth of African catfish fry after feeding the high protein and lipid content house fly larvae as live maggot.

Ricci et al. (2003) developed a system for the mass production of *Panagrellus redivivus*. It consisted of autoclavable plastic bags filled with sponges soaked with medium. This system enlarges the usable surface inside the growing system by using crumbled polyether polyurethane sponges to create an interstitial space. This space allowed optimal reproduction conditions and served as a living habitat for the nematodes. It also guaranteed sufficient aeration. The bags were inoculated with *Saccharomyces cerevisiae* to guarantee a monoxenic culture. The system was aerated and kept humid during the 11– 13 days of incubation at 25°C. Ricci et al. (2003) also experimented with different potato/water mixtures and found that a one-part instant potato flakes to two-parts water was the best mix for the worm-frosting technique presented above.

Worm harvesting is a very simple procedure. Wait until the worms are climbing the container walls and you will be able to collect them by running your finger around the walls. If you find this method a little unpleasant, then you can use a small stiff brush. The worms can then be fed directly to the larvae by swishing your finger or the brush in the aquarium water. Do not dip your finger or brush into the culture medium to collect worms, as any culture media residues should be minimised in order to avoid pollution of the aquarium water. Another method of harvesting is to lay wooden ice-block sticks (or similar objects) on the surface of the culture. The worms will crawl onto the sticks and you can then simply swish the stick in the aquarium water. Yet another method is to use a damp (thick) paper towel cut to fit over about half of the culture surface. To harvest the worms just use a spoon or spatula to gently scrape the worms right off of the paper towel, making sure you don't tear the towel (Wedekind, 2008).

Do not forget uneaten worms will die and pollute the aquarium water, particularly in a small aquarium. If left unattended, it can decimate an entire batch of fish larvae in a matter of hours. To prevent this problem, try feeding the larvae three or four times per day in small amounts rather than one or two large ones. In all, microworm offers a cheap, simple and nutritious food for feeding the larvae of most freshwater fish species. It is also suitable for feeding juveniles and adults of some of the smaller fish species such as *Iriatherina werneri* and *Pseudomugil* species.

Microworm can be fed alone or in combination with other foods such as brineshrimp nauplii, rotifers, zooplankton, egg yolk, dry diet, etc. Studies of fish larvae fed microworm are not significantly different from those fed brineshrimp. A feeding program utilising a combination of food items is better able to meet the nutritional requirements of all freshwater fish larvae. The use of different culture media to produce microworms affects the reproduction of the nematodes as well as their nutritional composition. Studies on enriched media for microworm have shown encouraging results. The nutritional quality of microworm can be enhanced by the use of the direct enrichment technique. Enrichment is simply carried out by adding the product to the culture medium. In one report (de Lara et al. 2007) the microworm were cultured in two media: one with oatmeal and the other with spirulina enriched oatmeal, in 15x15x5 cm plastic containers with 200g oatmeal and 300 millilitres (ml) purified water. Five grams of spirulina was used in the medium. The results show that growth of the microworm population in the spirulina-enriched medium presented the highest abundance of individuals on the second week of culture, whereas the population grown in the oatmeal medium showed the highest abundance on the fifth week of culture but did not reach the number of organisms attained by the population cultured in the spirulina-enriched medium.

The amino acids content of the populations from both media were compared to those reported for brineshrimp fed with spirulina, observing that the amounts were higher for most amino acids in microworm cultured in the spirulina-enriched medium. The composition of fatty acids in the microworm cultures in both media depicted significant differences for the linoleic, arachidonic, and eicosapentaenoic fatty acids, which were found in a higher percentage than reported for microworm cultures in oatmeal supplemented with sunflower oil. This information shows that Spirulina accelerates growth of microworm populations and allows the presence of amino and fatty acids.

Rouse et al. (1992) used a culture medium which was fortified with a 10% fish oil emulsion, obtaining nematodes that had significantly higher total lipid content and elevated levels of highly unsaturated fatty acids (HUFA). Additional investigations concerning the effect of an added oil source (fish oil or sunflower oil) on body composition, average yields and multiplication factors of the nematodes were conducted by Schlechtriem *et al.* (2004). Corn oil and yeast were added in varying combinations to culture media of *Panagrellus redivivus* to effect a change in growth and fatty acid content. The addition of corn oil or yeast to cultures increased nematode growth over standard media. Combinations of corn oil plus yeast increased nematode growth by 68%.

Other nematodes currently being cultured in the aquarium hobby that are similar to microworms are Walter worms and Banana worms. Both can be cultured in the same manner as microworm.

Walter worms were reportedly isolated from a grindal worm culture in 2002 and cultured by Helmut Walter of Germany. They are smaller but thicker than microworms. They are reported to live up to 35 days, producing 60 or more young from about day-4 onwards. Walter worms can stay alive in the aquarium water for 24 hours or more and stay suspended in the water column longer.

Banana worms are an endoparasitic soil nematode found in the banana-growing areas of Australia, Central and South America, Africa and the Pacific and Caribbean Islands, causing what is variously called root rot, blackhead or toppling disease, and predisposing trees to fungal infection.



# Gary Lange

### Vinegar Eelworms

*Turbatrix aceti*, formerly known as *Anguillula aceti* and colloquially as the vinegar eel, wine eel or vinegar worm, are small (1-2 mm) free-living, non-parasitic roundworms that feed on bacteria and yeast that cause fermentation in vinegar. They are adapted to living in a low *p*H (acidic) medium and are an excellent live food for freshwater fish larvae. Although vinegar eelworms live best in an environment of weak vinegar, they can be cultured in a variety of different media such as apple cider and 4% sugar in water. It is the sugar rather than the acetic acid which appears to be the essential element of the medium. *Turbatrix aceti* is often found in great numbers in vinegars made of apples or other fruits, or in other fermenting substances. The nematode is free-swimming in the liquid, reaching high individual numbers at the surface, where the oxygen concentration is higher, and are constantly in motion.

Starter cultures can be obtained from biological supply companies, aquarium stores, or fellow hobbyists. Vinegar eelworms are readily cultured in large numbers provided certain simple procedures are followed. They must be grown in natural cider vinegar or Balsamic vinegar that has not been chemically treated to inhibit growth of bacteria and yeast upon which the worms feed. The vinegar can be used either pure or diluted with 25 to 50% water. The periodic addition of a small amount of apple juice seems to add something that causes a greater population of worms, but is not absolutely necessary.

Another successful method for culturing vinegar eels was reported by inoculating commercial clear apple juice diluted 1: 1 with water with a dense suspension of eels in vinegar (about 20 millilitre per litre of diluted apple juice). Other fruit juices, e.g., pear, peach, apricot, may also be used although the sediment interferes with observation and cleaning of the worms. The inoculated juice was poured into five litre glass containers to a depth of about 25 mm. The bottles were covered loosely and kept at room temperature. After 7-10 days the fermenting cultures were dense with vinegar eels. At this point the bottles were half-filled with vinegar. These cultures can be kept for long periods of time with no attention. If the fermenting juice was kept too long without adding vinegar, putrefaction with loss of the culture often occurred. With a little experience it was possible to judge the right time for the addition of the vinegar.

Add your eelworm culture to approximately 500 ml of culture medium in a wide-mouth glass jar covered to reduce evaporation. Punch small holes in the lid (cover) for aeration. Cultures should be maintained at 20–30°C and subcultured every 6–8 weeks to fresh medium. Vinegar eelworms can rapidly increase in number and females with developing embryos can be found within one week of starting a new culture. Embryonic development takes about 7–10 days from time of fertilisation of the egg to time of birth of the young. The larvae become sexually mature in about 28 days. Median life span of vinegar eelworms varies with temperature, from 55 days (25°C) to 40 days (30°C). The maximum life span has been reported as 10 months.



The advantage of culturing and feeding vinegar eels is:

- Vinegar eels will live for a long time in the aquarium water.
- Vinegar eels swim in the water column and stay towards the surface where rainbowfish fry feed.
- Vinegar eels are just a little smaller than micro-worms, a great size for most baby fish.
- Vinegar eel cultures require little attention (indeed they can be ignored for weeks at a time). I had two stock cultures (ca. 500 ml each) in my fishroom that had living populations for more than two years without the addition of either fresh vinegar or apple.
- Vinegar eel cultures don't "go off" leaving an unpleasant smell.

Harvesting of the worms may test your patience until you have developed a procedure to collect them, as it is very important not to get any culture medium in the fish larvae tank. Several methods of concentrating the worms and washing them free of the culture medium have been reported. The most common method to harvest the worms is to pour or siphon the culture medium through a laboratory (1.2  $\mu$ m) or coffee filter paper and in so doing collect most of the worms. Filters with larger pore sizes would probably be better as a small pore size





captures more debris. Return the medium back to your culture container. The filter paper with the collected worms is then rinsed into a jar of clean freshwater and can then be poured into the larvae tank. The eelworms will live for a long time in the tank but care should be taken to prevent supplying too many worms at one time.

Some culturists separate the worms from the vinegar in small test tubes. Culture medium on the bottom; some filter floss, and clear water on top. The worms seeking oxygen move up through the filter floss to be near the surface. Very effective, but not enough worms to feed many fry.

A modified technique using the same theme but productive enough to be useful is to use longneck bottles for culturing. Keep the culture medium level well below the neck to have adequate surface area. To harvest, remove the floss plug and add enough spare culture medium to reach above the bottom of the narrow neck. Push the polyester filter floss down to the surface. Add fresh water up to the top of the neck. In a few hours (or overnight), there will be a rich collection of eels in the fresh water, but no noticeable mixing from the vinegar below.

Collect the worms with a bulb baster or dropper. Remove the floss and squeeze dry. Pour enough vinegar back into a spare bottle to get good surface area again in the main culture bottle and loosely plug the top of the neck with the damp floss.

Another method depends on the negative geotropism of the vinegar eels. The culture is concentration in a separatory funnel. Most of the organisms will, within a few minutes, aggregate at the surface of the liquid in a dense layer 5-8 mm or so deep. The lower liquid is drained off rapidly. The concentrated suspension remaining is poured into burettes (50 or 100 ml.). After a few minutes the lower portion of the burette will contain few worms and the organisms will have begun to aggregate at the surface. The stopcock is opened slightly and the liquid allowed to drain out drop wise at a rate which will leave the surface-aggregated organisms behind, adhering to the wall of the burette. If the outflow rate is properly adjusted, most of the vinegar eels will remain on the walls of the burette when the liquid has drained out. The worms are rinsed from the burette with a small amount of a suitable solution.

When a sufficient number of worms have been collected they may be washed any desired number of times by repetition of this procedure.

An even less complicated method (P. J. Unmack, *pers. comm.*) is to hang something porous in the culture such as a plastic coffee filter. In the absence of a plastic coffee filter an abrasive pot scrubbing sponge used for cleaning the dishes can be used. The reason for the abrasive sponge rather than a regular sponge is that the culture medium will drain out of it fairly thoroughly without having to squeeze it. The sponge only needs to be inserted in the culture medium about 2–3 cm as most of the vinegars eels are at the surface.

To feed your fish just lift the sponge out and allow it to drain. Vigorously swash the sponge in some water and you have your vinegar eels ready to feed to your fry. Hang the sponge back in the culture and you are ready for next time. One thing to be wary of is that you will transfer very small amounts of vinegar to your aquarium which may lower the pH. To counter this, add some sodium bicarbonate to the water that is used to swash the sponge in containing the vinegar eels once every few days. As the water-worm mixture is pour into the fry tanks it buffers the tank against acidic conditions. Just try the different methods until you find one that works best for you.

### Whiteworms

Whiteworms, *Enchytraeus albidus* (Henle, 1837) are probably one of the most popular forms of live food cultured by aquarists. They are an excellent, easily produced form of live food that is highly nutritious and especially valuable for conditioning rainbowfishes before spawning, or for young fast growing fish. Fed two or three times a week; they will give your fish a nutritional boost. The actions of your fish will change dramatically when they see the movement of live struggling worms in the aquarium.

Some aquarists feel that fish fed exclusively on whiteworms become obese due to the fat content of the worms. However, the problem may lie more with overfeeding with the worms, rather that the fat content of the worms.

The secret for successfully raising whiteworms is to understand their particular needs and supply them. Their successful cultivation is largely dependent upon constant care and attention to small details in the condition of the culture. The principal things to be considered are the medium; moisture, and food. Whiteworms required soils containing relatively high organic matter content and a soil *p*H of about 6.8 to 7.2 for optimal conditions. Whiteworms will usually not survive in acid soils (i.e. < pH 5).

Experience has shown that shallow wooden boxes work best. Typical worm boxes are 15 to 60 cm long, 15 to 30 cm wide and no more than 10 to 15 cm deep. In any case the use of several small cultures rather than a large one is advisable. Plastic containers with drainage holes punched in the bottom have been used with success.





However, simple boxes made of pine and plywood is generally preferable to plastic, Styrofoam, or other materials because the joints allow better drainage and aeration of the soil.

The culture must be covered to block out light and keep out predators. Ants, beetles, and other creepy crawlies will feed either on the worms or the food. A secure lid and careful placement of the culture box will prevent such pests. An inner soil cover is recommended to keep the soil surface from drying out. Any flat material that can be pressed lightly onto the surface of the soil will serve as a cover.

I use a thin piece of scrap glass cut smaller than the surface area of the soil. Leave a border of about 15-mm of soil exposed to the air. The collection of moisture at the cover attracts the worms, making it an ideal place to feed them. By feeding and attracting the worms to the surface, it will be easier to collect them to feed to your fishes.

Whiteworms will grow in any kind of light loam soil of such a character that it does not easily harden when dry, while on the other hand it should not be sandy. Potting mix obtainable from most garden supply stores should be adequate. Choose a good quality mix as I have found some potting soils contain a lot of coarse material in them. Leaf mould and humus are excellent additives that will improve the soil significantly. Some of the best mixes are those that have been designed for growing seedlings. These are generally very fine and hold moisture well, but remain loose. Whichever soil you choose, ensure that it doesn't contain any chemical fertilisers, sterilisers, fungicides, pesticides and other man made chemicals or contaminants, as these additives will kill the worms.

Fill the box about two-third full with your chosen soil mix. The surface of the soil should be level and pressed down, not too firmly, to leave no lumps above to dry or mould. Wet the soil until it's reasonably damp, allowing any excess water to drain. There should be sufficient moisture to allow free movement of the worms but not enough to bring them to the surface except as they may congregate on the under side of the piece of glass resting on the surface of the soil. The next step is to get your starter culture. These are often available from aquarium stores, live foods suppliers or from a fellow hobbyist.

Once you have your starter culture, empty the contents on top of the media. Sprinkle a small amount of food over the surface of the soil and spray with water. Place the soil cover on the surface, put the lid on and move the box to an area that will stay between 15–21°C. For best results, keep the culture in a cool dark area. Allow the culture to stand undisturbed for several days to allow your whiteworms to propagate.

The current trend for culturing whiteworms is to use small individual flat plastic food containers stored in an electronic wine cooler, which can be maintained at the desired temperature. This not only provides the ideal temperature, but also keeps unwanted pests out of the culture.





Whiteworms reproduce normally at and above  $8-10^{\circ}$ C, with optimum growth and reproduction occurring between 15–21°C. As the temperature begins to rise or fall below this range; their production rate will decline. Growth has been reported to occur more rapidly at the higher end of the range with maturity in about 28 days at 20°C, the clitellum (The clitellum is responsible for producing the cocoon in which the eggs are deposited) forming when the worms were about 13–14 mm. The maturation period at 8°C was at least twice that at 20°C. Experience has shown that at temperatures above 30°C or below 0°C, whiteworms will die.

Whiteworms will eat just about anything organic. Aquarists feed their worms vegetable based foods such as plant material, oatmeal, bread soaked in milk, wheat flour, cereal, mashed potato and dozens of other similar foods. They will even eat flake and pelleted fish foods, dry dog and cat food, if they are pre-soaked beforehand. One feeding trial reported that the best single food for whiteworms was breadcrumbs. In another study (Memi et al., 2004), whiteworms were fed five different diets, and after 90 days the numerical increase in their population was calculated. Four of the five diets were composed of carbohydrates, vegetables, fruits, and commercial trout feed pellets, and the fifth was composed of a combination of all four of these. At the end of the study, the greatest numerical increase and best reproduction was found to have occurred with the commercial trout feed pellets, which contained 45-47% protein and 12% fat. The least increase in number of individuals was observed in the vegetable-based group. In yet another study with whiteworms that were fed vegetable-based and cereal powder-based diets containing casein attained higher levels of weight and reproductively than those that were fed vegetable and cereal powder-based diets not containing casein.

A more recent study reported that the best results in whiteworm production were obtained by implementing different diets in alternation. However, we are what we eat, so the nutritionists tell us. Well, worms are no different and I found that Heinz<sup>®</sup> high protein baby cereal (blended with water), provides excellent results. The cereal also provides higher protein levels than many of the other foods. This higher protein increases the nutritional value of the whiteworms,

which is then passed on to your fishes. Similar procedures are used in commercial fisheries with brine shrimp. Young brine shrimp are fed an enhanced diet, which is passed on to the fish when they eat the shrimp.

Whiteworms should not be fed too heavily at first because surplus food tends to attract mites, fungal growth, and bacterial contamination. You will have to regulate the amount of food offered during the first month until the culture stabilises. Replenish the food supply, as needed (ideally every three to four days). If the food supply is not entirely consumed between feedings, you are adding too much food for the worm population.

Whiteworms are one of the largest species of the genus Enchytraeus (adults reach 15-40 mm in length and from 0.5 to 1.0 mm in diameter). They are hermaphroditic, with each individual having both male and female reproductive organs. One worm mates with another individual and each fertilise the other. The worms exchange sperm cells during copulation and eggs are laid in transparent cocoons. Each cocoon produced by young adults contains 9-10 eggs; cocoons from mature adults produce 20-25 eggs. As the culture density increases, the reproductive rate levels off and old worms will only produce around 2-3 eggs per cocoon. The highest egg production reported was in the vicinity of 35 eggs per cocoon. Average per total population in culture is 10 eggs per cocoon. The eggs hatch in 12 days, and worms begin reproducing in 20-28 days depending on temperature. Each individual can produce as many as 1000 eggs over its life span.

If the culture is maintained properly, the worms will gather in mass on the surface of the soil. The worms will often congregate on the glass cover where they can be scraped off and fed to your fish. Do not harvest worms before the first month of growth. Let the culture grow and you will be able to make new cultures and collect all the worms you need. You will need to inspect the culture for food and moisture levels two or three times a week. If the food is gone, then increase the amount of food given. If food remains, then remove the excess and reduce the amount provided.

You may find that the moisture level of the culture will drop and that the surface of the soil will begin to dry out. If this condition is allowed to continue the worms will start to go deeper in the soil seeking moisture. When they do this, they are also moving away from the food you place on the surface of the soil. Spray with water to maintain a damp, but not soggy. look and feel. A plant sprayer or mister can be used for this purpose. The regulation of moisture may be aided by removing the cover for a time as necessary. In laboratory testing, reproduction and body length was reduced with soil humidity (moisture) content of 15% and lower. On the other hand, higher soil humidity did not necessarily coincide with higher densities. During the test, it was found that low soil humidity inhibited not only reproduction, but had also a negative effect on the growth of the parent generation. Soil humidity is best maintained at around 22-26%.

Mould may often be present but does not seem to interfere with worm production if the food masses are not large. Removal of surface growth and taking the cover off to allow short drying



periods will help keep it in check. However a souring culture is to be strictly avoided. Whiteworm cultures are often infested with mites. These small spiders like creatures are harmless and will not do any damage other than eating the whiteworms' food. If you keep your culture in a refrigerator, then mites will not be a problem. After a period of six to nine months, the soil texture will begin to break down due to the activity of the worms, and the soil will become very acidic. This inhibits the production of worms, leaving you with only adult worms. To maintain your culture, the old soil should be removed and fresh soil placed in the box. The culture can be divided into several boxes at this time, as it is a good idea to have more than one culture in operation. You can transfer most of the worms by collecting from the old box and placing them in the new box. Another simple method to replace an old culture is to scoop away the top 2–3 cm of soil with most of the worms and gently mix it into fresh, moist soil in a new box.



### **Grindal Worms**

Grindal worms, *Enchytraeus buchholzi* (Vejdovsky, 1879) are a smaller relative of the whiteworm, that usually only grow to about 10-mm and thus are an ideal size for most rainbowfishes including both adults and larger fry. Mrs. Morten Grindal, of Sweden, who was prominent in the development of culturing techniques for whiteworms, was apparently the first person to isolate this smaller species. Grindal worms can be cultured exactly as whiteworms but are a much more adaptable species and have a greater tolerance for warmer temperatures. Maturity has been reported to occur around 16 days at 20°C, the clitellum (see above) forming when the worms are about 3–4 mm. The generation period (cocoon to cocoon) is about a month at 20°C.

In laboratory testing, *Enchytraeus buchholzi* were kept in an incubator at  $15 \pm 2$ °C. Water loss and food were replenished if necessary during the test period. After 21 days the offspring and the surviving adults were counted. For two moisture levels (5% and 20% water content) the segment number of the surviving adults was counted. From 20% up to 40 % water content *Enchytraeus buchholzi* showed no significant difference in reproduction. Below 20% and above 40%, the number of offspring was reduced. No juveniles were found at 5% water content, although adult survival was equal to higher moisture levels. Reproduction was decreased at 30% water content compared to 25% and 35% water content. Low soil moisture inhibited not only reproduction, but had also a negative effect on the growth of the parent generation.





### Wonderful Worms

Any number of worm species may be readily cultured or purchased for use as a live food source for your rainbowfishes. All will provide excellent nutrition for your fishes, especially if they have been cultured under clean conditions and fed on a nutritious diet. One can also find different types of worms in streams and ponds all of which are good for rainbowfishes. Nearly all rainbowfishes go into a feeding frenzy when presented with live worms. The smaller they are the better because rainbowfishes do not masticate their food, they swallow it whole, but you can always chop or squash the worms according to the size of your fish. Chopped worms of all kinds make an excellent food for small juveniles and fry. However, although worms can be chopped into pieces so as to be readily swallowed by the smaller rainbowfishes, a whole, small worm will provide more complete nutrition than will a small piece of a larger worm. Therefore, you may wish to purchase worms in various sizes or maintain a live culture at home.



### Earthworms

Many aquarists have had excellent results when using earthworms as food for the larger rainbowfishes. They may also be chopped into smaller pieces for feeding the smaller rainbowfishes. Earthworms may be purchased at some pet stores, or ordered in large quantities from commercial suppliers. Earthworms that are purchased and stored for later use do best when kept in a refrigerator. They usually keep for weeks in the refrigerator; simply rinse them off with tap water before feeding. Some people take the extra step of stripping the worms before feeding them to their fish. Worms usually have their stomach filled with dirt or undigested matter; to remove this, hold the worm by the thick end, pinch its body, and slide your hand down its length. The dirt will be pushed out the other end and can then be rinsed off before you feed the worm to your fish. Uneaten earthworms will remain alive in freshwater aquariums for up to eight hours, but they decompose rapidly upon death.

Earthworms can be easily cultured in any suitable container filled with alternating layers of good-quality soil and dead leaves. The earthworms will consume the leaves as well as fish food flakes, or any vegetable matter. Collection of the worms can be simplified by feeding them on the surface of the soil, below a layer of damp burlap.



#### Blackworms

Blackworms (Lumbriculus variegatus) are small, aquatic relatives of earthworms. They are found in sediments and submerged organic debris - especially along the shallow margins - in ponds, marshes, and lakes throughout North America and Europe. This family of some 18 genera and about 170 described species is represented in Australia by only two species: Lumbriculus variegatus are usually small (4-6 cm in length), green anteriorly with the remainder red to black and Stylodrilus heringianus, (3-4 cm in length), and pale to white in colour. It is very likely that both these species have been introduced in modern times. Lumbriculus variegatus is commonly cultured and sold in Australia as a live food for aquarium fishes. Although, some blackworms sold in aquarium stores in Australia are actually Limnodrilus udekemianus, a species of tubifex. Blackworms are readily accepted by all rainbowfishes.

*Lumbriculus variegatus* is often readily identifiable in the field by its vigorous thrashing when handled and the greenish-brown anterior end and reddish posterior end, the latter due to the branched blood vessels in each posterior segment. This species is commonly found in disturbed habitats and is particularly common near urban centres or in catchments associated with major cities and towns of eastern and southern Australia. *Stylodrilus heringianus*, with its pair of permanently exposed penes on mature specimens, is distinctive but less common.

Blackworms will live well in freshwater aquariums, where they burrow into the substrate and provide foraging opportunities for bottom dwellers such as catfish and even rainbowfishes. It is possible that they will develop their own self-sustaining population. At normal aquarium temperature populations can double in about 3–4 weeks or less. The worms are also excellent scavengers, burrowing under rocks and deep into the substrate to consume uneaten food and wastes.



### Tubifex

The type species of this family, *Tubifex tubifex* was one of the first aquatic oligochaetes described and is well known to aquarists as an excellent live food. It is cosmopolitan in distribution, as are several of these species in Australia. The Australian tubificid fauna appears to have biogeographic affinities with northern hemisphere tubificids. The Australian fauna is relatively small, with only about 29 described freshwater species. *Tubifex tubifex* has been the subject of intensive research because of its role as the intermediate host of the myxozoan parasite (*Myxobolus cerebralis*) that causes whirling disease in salmonid fish. The presence of this species in Australia is thus of interest to the aquaculture industry, although the parasite itself has not yet been recorded here.

Freshwater tubificids are often called sludge worms for the propensity of some widespread species, such as *Limnodrilus hoffmeisteri*, to occur in organically polluted waters, even sewage sludge. This has sometimes led to all tubificids being labelled as 'pollution tolerant' whereas in fact many species are sensitive to organic enrichment and tubificids show a wide range of sensitivities to other pollutants such as pesticides and heavy metals.

Tubificids occupy a wide range of aquatic habitats, from deep lakes to small streams and ponds and have recently been found in Australian groundwater. In its natural habitat the tubifex has a bi-annual life cycle although the period of breeding is influenced by several environmental factors. Most adult tubifex die after breeding, others are able to reabsorb their reproductive organs and return to the immature condition. These individuals are then able to reproduce more than once in their life time.

Tubificids are small burrowing worms often about 2–5 cm long and roughly 1-mm in diameter when fully mature. They typically feed by placing their heads down into the sediment and leaving their tails protruding into the overlying water. Although, the worms can also remain totally submerged during feeding. Particles of sediment ingested at depth then pass through the worm and are egested onto the sediment surface in small mounds of faecal pellets. The worms use the dissolved oxygen present in the ingested water to breathe. Once isolated outside their protective sediment the worms come together and form a protective ball.

Many aquarium books advise aquarists to avoid feeding these worms. Years ago, these worms were collected from the mud of polluted waters and fed to aquarium fishes. If they were not thoroughly rinsed and purged, they would often cause disease problems. However, because tubificids are directly exposed to contaminated sediments and are at the bottom of the food chain, the greatest threat I would think would be from the bio-accumulation of toxic substances that can cause a wide range of health problems.

Today, however, tubifex worms such as *Limnodrilus udekemianus* and *Limnodrilus hoffmeisteri* are commercially cultured as a food source for aquarium and food fishes.



However, live worms commonly sold as 'tubifex' may comprise a mixture of species including *Tubifex tubifex*, *Limnodrilus hoffmeisteri*, *L. udekemianus*, *Rhyacodrilus coccineus*, *Dero digitata* or *Lumbriculus variegatus*.

#### **Feeding Worms**

Blackworms and tubifex are not usually cultured by the home aquarist as they are readily available in the aquarium trade. However, with a little research, successful cultures can be easily maintained. Generally they are just purchased as required and stored in a pan of shallow water in the refrigerator, where they should live for at least two weeks.

Optimum water temperature is 18–20°C with an upper lethal temperature of 35°C; at 5°C the worms become inactive. Tubifex and blackworms are very sensitive to chlorine or chloramine, so treat the water with a suitable chlorine neutraliser. Change the water after 24 hours and thereafter once a week should be adequate. An airstone can be used to help circulate and aerate the water. However, keep the airstone off the bottom of the container; otherwise the tubifex will clump tightly around the airstone cutting off the air supply. A small piece of boiled potato can be added to water if the worms are to be kept for any length of time.

When feeding the worms to your rainbowfishes, a plastic cone worm feeder will help retain the worms up at the top of the aquarium until the fish have had time to eat them. If the worms are simply added directly to the tank, many will fall to the bottom and bury themselves in the gravel where the fish will be unable to find them.





### **Brine Shrimp**

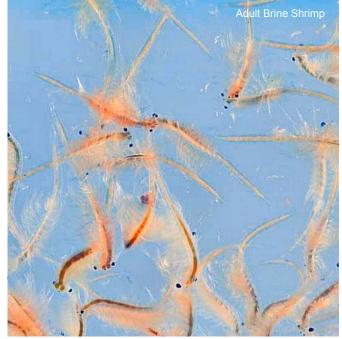
Brine shrimp nauplii are widely used in aquariculture for feeding fish larvae. The nauplii exhibit two essential qualities for this purpose: they are of an appropriate size to be ingestible, and they move actively in the water column, establishing themselves as an active target for young fishes. Brine shrimp can produce cysts (eggs) under certain conditions and these, since they float, are easily harvestable. The eggs are collected and placed into cold storage for at least three months. This process is called 'Diapause Inactivation' - a process that is similar to hibernation.

Following the cold storage period, the eggs are cleaned, washed, and separated. The partially hydrated eggs are disinfected, dried in rotary ovens to about 6% residual moisture, and then vacuum packed. The finished product can then be stored for long periods. When the eggs are placed into saltwater they are re-hydrated and hatch. The hatching rate of cysts varies according to storage time and conditions as well as geographical origin and commercial brand. Generally, 150,000 hatched Artemia nauplii can be obtained from 1 gram of cysts.

Brine shrimp eggs can last for several years as long as they are maintained in a dry condition at all times. Sealed cans can be stored for years at room temperature, but once opened, should be used up within two months. Store opened eggs in an airtight container in the refrigerator or in a cool dry place. If the entire contents of a can will not be used up in two months, it is recommended that the portion that is expected to be unused be placed in a tightly closed container and frozen until needed.

Alvin Seale, the Superintendent of the Steinhart Aquarium, USA, first reported the suitability of brine shrimp nauplii as a source for larval fish in 1933. However, it wasn't until the late 1970s that a continuous supply of eggs was available. At that time, the major supply of eggs occurred in the United States, mainly from San Francisco Bay and the Great Salt Lakes, Utah. Commercial brine shrimp farms have now been established in many parts of the world, and are commonly introduced into evaporation ponds used for the commercial production of salt. However, except for limited, small-scale production, the bulk of the eggs used today emanate from a wild-capture harvest susceptible to over-fishing and reduced production due to uncontrollable climatic influences. The net result is an unreliable supply of a product with concomitant fluctuations in price. There is also a large variation in nutritional quality, hatching quality, and size of nauplii among commercial sources of brine shrimp eggs.

The ease of hatching brine shrimp eggs and the commercial availability of the adult stage has made them a popular food source. Freshly hatched brine shrimp nauplii have a lipid-rich yolk, high in unsaturated fatty acids. Due to this nutritious yolk and small size, brine shrimp nauplii have become the standard food for larval fish in the aquaculture industry. Brine shrimp nauplii emerging from their protective shells are extremely small, mostly less than 500  $\mu$ m but can differ (400 to 800  $\mu$ m) according to origin. The smallest is believed to be the San Francisco Bay variety.



Another overlooked fact is that fish larvae are thought to take advantage of the nauplius' digestive enzymes, as most fish larvae have a very weak digestive system when being young. Additional support for this hypothesis is found in the different growth results when fish larvae are fed with either decapsulated cysts or nauplii. On an individual weight basis, the decapsulated cysts and nauplii of Artemia have similar biochemical composition in all the major nutrients. Thus, with regard to the amount of nutrients, there is no difference in feeding brine shrimp cysts or nauplii to fish larvae. However, in some fish species higher growth rates have been achieved with nauplii. Protein is the major component of the dry matter in brine shrimp. Because the interaction of proteins with water has an effect on the functional properties of the protein, the protein structure might differ between cysts and nauplii due to the high water content in the latter.

Brine shrimp nauplii are an excellent live food, not only for larvae but also for adults of the smaller species of rainbowfishes. They can live in freshwater for around 2–4 hours before they die, making them an ideal live food for small rainbowfish larvae. However, they are not suitable as a first food for all rainbowfish larvae; some larvae are so small that they will require micro-organisms.

Initially sold as a frozen product to the ornamental fish hobby, adult Artemia are now marketed live, and in several forms such as decapsulated cysts, newly hatched nauplii, meta nauplii, juveniles and adult stages as well as processed forms like frozen, freeze dried, dried and flakes.

Brine shrimp are a relatively primitive form of aquatic crustacean that occurs naturally in saline waterbodies worldwide. The original species first described by Schlosser from the salterns of Lymington, England (1755) and named by Linnaeus (1758) as *Artemia salina* is now considered extinct, and several sibling species or sub-species are recognised today. They belong to the subclass Branchiopoda, which is characterised by many pairs of flattened appendages on the thorax, in contrast to other members of the Crustacea that have no more than six pairs.



The environmental conditions under which brine shrimp live are highly variable. The salinity can exceed 300‰, (parts per thousand) where most other life cannot survive. Advantaged by the absence of predators and food competitors in such places, brine shrimp develop very dense populations. Although not a marine species, they sometimes occur in bays and lagoons. They are more commonly found in highly saline lakes, such as the Great Salt Lake, Utah where the shoreline may become ringed with brown layers of accumulated brine shrimp eggs.

The development of brine shrimp is influenced by many factors and the tolerance of these factors is strain dependent. Optimum temperature for most strains ranges between 25 and 35°C but strains have been reported thriving at 40°C. Most geographical strains do not survive temperatures below 6°C except as eggs. These eggs are tolerant of temperatures from far below 0°C to near the boiling point of water.

Although brine shrimp can survive and reproduce under a wide range of salinity, they are seldom found in nature in salinities below 45% or above 200‰. The *p*H tolerance varies from neutral to highly alkaline but the eggs will hatch best at a *p*H of 7.5 to 8.5. Many predators including zooplankton that populate natural salt waters, fish, several insect groups (odonates, hemipterans and beetles), and birds feed on brine shrimp in situations where they can tolerate the conditions.

Copulation is initiated when the male grasps the female with its modified antennae. At low salinities (<85‰) and optimal food levels, fertilised females usually produce free swimming nauplii (ovoviviparous reproduction) at a rate of up to 75 nauplii per day. They may produce 10–11 broods over an average life cycle of 50 days. Under ideal conditions adult brine shrimp survive for several months and produce up to 300 nauplii every 4 days. Cyst production (oviparous reproduction) is considered to be induced by high salinity, under conditions of high eutrophication (large  $O_2$  fluctuations between day and night) and chronic food shortages.

At high salinities (>150‰) and low oxygen concentrations, the embryos develop to the gastrula stage. They then become surrounded by a thick shell and enter dormancy (diapause). Females can release up to 75 cysts per day which float in the highly saline water (eggs from Mono Lake in California sink). The floating cysts are eventually blown ashore where they accumulate in large masses and dry.

Development is resumed when the cysts are re-hydrated and the life cycle is begun again. After several hours the outer membrane bursts and the embryo emerges still encased in the hatching membrane. Soon the hatching membrane is ruptured and the free-swimming nauplius is born. The first instar is brownish-orange coloured and has three pairs of appendages. The larva grows through about 15 moults and becomes differentiated into male or female after the tenth moult.

Brine shrimp are typically filter feeders that consume organic detritus, microscopic algae, and bacteria. Blooms of microscopic algae are favourite habitats, and large populations develop in such areas where they feed on the algae and heterotrophic bacteria that are produced by these blooms.

Australian saline lakes harbour a rich diversity (with eight described species) of endemic brine shrimps (*Parartemia*), which, like Artemia, can produce cysts from which nauplii hatch, but under quite different limnological conditions. Their economic and scientific values remain almost totally unexplored. Australia also has a freshwater cousin of the brine shrimp (*Branchinella*), with nineteen described species that occur in temporary fresh waters (pools, ditches, rock-pools, and ponds).

Artemia (*Artemia francisiana*) have been introduced into a number of coastal salt-works in Australia for a number of years. Artemia are now found in habitats occupied by *Parartemia*, facilitated both by increasing degradation of saline lakes and by human involvement. The net result could be the replacement of many local species of *Parartemia* with a cosmopolitan species.

### Hatching Brine Shrimp

The standard procedure for hatching brine shrimp nauplii is to incubate the eggs for 24–48 hours in a saltwater solution and then separate the nauplii from the unhatched eggs and shells. I use a 2-litre wide-mouthed glass jar filled with tap water. To this I add 10 to 20 grams (2 to 4 level measured teaspoons) of cooking salt and a pinch (<sup>1</sup>/<sub>4</sub> level teaspoon) of Sodium bicarbonate.

Brine shrimp eggs have been shown to hatch out at salinities ranging from 5 to 35‰ (parts per thousand). However, research has shown that better hatching results have been achieved at the lower range. During the hatching process, the eggs absorb water through the shell by osmosis. When the osmotic pressure within the egg is great enough, the shell then bursts, freeing the larval brine shrimp. At higher salinities, the osmotic pressure outside the egg is higher than within the egg, lengthening the time for hatching. However, if you are having problems with poor hatch rates then experiment with different sali levels as I have found that you can get better results using different salinity levels.

The *p*H for the hatching solution may range from 7.5 to 8.5. A *p*H above 9.5 tends to be too alkaline, while a pH below 6.5 results in a dramatic decrease in the hatching results. Hatching time varies with incubation temperature and the geographic strain of brine shrimp used. The temperature for optimal hatching rate and high hatching efficiency is considered to be  $27-30^{\circ}$  Celsius. However, at least 90% of premium grade eggs should hatch within an 18-hour period in a temperature range of  $25-32^{\circ}$  Celsius. Lower temperatures will cause the eggs to hatch at a slower rate. The air temperature of my fish room governs the temperature in my case. During summer, I harvest the shrimp after 24 hours, wintertime 48 hours, and spring/ autumn 36 hours. I live in a sub-tropical climate and my fishroom rarely drops below  $20^{\circ}$ C during winter, but in summer it can reach 30 to  $35^{\circ}$ C.

The recommended hatching density of eggs should not exceed 5 grams per litre of water. Constant aeration should be provided by an airline, without an airstone, inserted to reach the bottom of the jar. Aeration is essential for two reasons - the maintenance of dissolved oxygen levels, and keeping the eggs suspended in solution. Adjust the air supply so that moderate aeration occurs.





Too little aeration will result in low levels of dissolved oxygen while too much aeration will cause the cysts to stick to the upper jar out of the water. These conditions will significantly affect the hatch rate.

After the incubation period, turn off the aeration and allow the contents to settle for about 5 to 10 minutes. A distinct separation will occur; the unhatched cysts and egg shells will rise to the surface and be dark brown. Brineshrimp nauplii are bright orange and are located near the bottom of the hatching container or within the water column. Most of the newly hatched nauplii will accumulate just above the bottom. Siphon the shrimp into a fine mesh net ( $<120 \,\mu$ m) through a length of airline tubing, which has a short rigid extension (the depth of the jar) on the intake end. This makes it possible to position and siphon very accurately. This separation step is necessary, however, because small fry cannot digest unhatched cysts and shells, which can cause mortality if consumed. After rinsing the nauplii in a gentle stream of freshwater, which will remove any waste or salt residue, the nauplii can be fed to the fish. It is important to collect the nauplii quickly, because after 10 minutes or so, the oxygen levels of the water begin to drop quickly and the nauplii will begin to show signs of distress and die.

Discard the remaining contents of the hatching jar and wash with hot soapy water, rinsing well before use. The net should also be rinsed. Prepare fresh salt water for each new hatch. To have a fresh supply of brine shrimp daily, at least two hatching containers should be used; so that newly-hatched shrimp can be harvested daily.



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Artemia nauplii are most nutritious while they contain the yolk sac and should be fed as soon as possible after hatching. Artemia nauplii in their first stage of development can not take up food and thus consumes its own food reserves. At 28°C, the freshly-hatched Artemia nauplii develop into the second larval stage within 12 hours. It is important to feed first-instar nauplii to the fish rather than second-instar meta-nauplii which will have already consumed 25 to 30% of their food reserves within the first 24 hours after hatching. Moreover, instar II Artemia are less visible as they are transparent, are larger and swim faster than first instar larvae, and as a result consequently are less accessible as a prey. Furthermore they contain lower amounts of free amino acids.

Moulting of the Artemia nauplii to the second instar stage may be avoided by storing the freshly-hatched nauplii at a temperature between 5 below 10°C. This procedure decreases the metabolism of the nauplii, thereby preserving a higher nutritional value. Only slight aeration is needed in order to prevent the nauplii from accumulating at the bottom of the container where they would suffocate. In this way nauplii can be stored for periods up to more than 24 hours without significant mortalities. This technique allows not only a constant supply of high quality nauplii but also more frequent feeding for freshwater fish larvae.

#### Decapsulated Brine Shrimp Eggs

Brine shrimp eggs (cysts) consist of dormant embryos covered with a three-layered shell. Under optimal hatching conditions the embryos break out of the shell and hatch into nauplii, which are then used for feeding. However, the hard outer shell of the brine shrimp egg, the alveolar layer can be completely removed through a chemical process known as decapsulation. Consequently, the task of separating the nauplii from the unhatched eggs and shells is eliminated because 100% of the decapsulated eggs (both hatched and unhatched) can be fed. An important application of the decapsulated cysts is that even low-hatch or no-hatch cysts can be used for feeding.

The decapsulated cysts can be used immediately or dehydrated in brine solution for storage (brine cysts), or further subjected to a drying process for longer term storage (dried cysts). The advantages of decapsulation include disinfection of the cysts, improved hatching and no risk of fish larvae suffering from gut obstruction due to the ingestion of empty egg casings.

Many studies have demonstrated that the growth and survival of fish fed decapsulated cysts is better than or comparable with those fed newly-hatched nauplii. In one study (Lim *et al.* 2002), it was found that the fry of five common ornamental fish species tested (*Poecilia reticulata*, *P. sphenops*, *Xiphophorus maculatus*, *X. helleri* and *Hyphessobrycon herbertaxelrodi*) could readily feed on the decapsulated cysts, and their performances in terms of growth and survival was comparable to or better than those fed on *Artemia* nauplii or *Moina*.



Decapsulated eggs offer a number of advantages compared to the non-decapsulated ones:

- Empty egg casings are not introduced into the fry tanks.
- Nauplii that are hatched out of decapsulated eggs have a higher individual weight (30–55 % depending on strain) than regular instar I nauplii.
- In some cases, hatchability is improved by decapsulation.
- Decapsulation results in disinfection of the eggs.

Decapsulated eggs can be used as a direct energy-rich food source for rainbowfishes. The cysts contain an average of 30% more nutrients than newly hatched nauplii. The cysts have the appearance and the practical advantages of a dry feed and, in contrast to nauplii (470–550  $\mu$ m); their small particle size (200–250  $\mu$ m) is more suitable for smaller fish. If they have been dried before use, they have a high floating capacity, and sink only slowly to the bottom of the culture tank. Dried decapsulated cysts, when treated appropriately, can be stored for years, and hence may be fed off the shelf without the need for hatching.

#### Decapsulation [Method 1]

Decapsulation involves soaking the eggs prior to hatching in a solution of chlorine bleach (Sodium hypochlorite). Chlorine bleach chemically removes the shell without affecting the viability of the unhatched embryo.

1. First, hydrate the eggs by soaking 5 grams of eggs per 150 millilitres tap water for about 1 hour at 25° Celsius. Hydration is helped with low aeration and by the eggs being kept in suspension.

2. After hydration, add an equal measure of Sodium hypochlorite to the decapsulating container. The decapsulation process is allowed to proceed under low aeration and constant stirring for 5-10 minutes (depending on hypochlorite strength) or until the eggs are completely decapsulated. As the eggs decapsulate, they change colour from dark brown to grey to orange, and they lose their buoyancy. Decapsulated eggs are completely orange and no longer float.

It is crucial not to leave the embryos in the decapsulation solution longer than strictly necessary, since this will affect their viability. The time it takes for all the eggs to be decapsulated will vary with the type of eggs being used, so it is more important to observe the colour change than to watch the clock.

3. The decapsulated eggs are then placed in a fine mesh net  $(100-150 \ \mu m)$ , where they are rinsed thoroughly with a steady stream of tap water for 1–2 minutes, or until the chlorine smell is no longer detectable. (Sodium thiosulphate could also be used to neutralise the chlorine residue)

4. The eggs are now ready for hatching, or can be stored in a refrigerator ( $0-4^{\circ}$  Celsius) for a few days without any decrease in hatchability.

#### Decapsulation [Method 2] (Schumann, 2000)

Prepare a buffer solution by dissolving 40 grams of 40% sodium hydroxide in 60 ml of freshwater. Then add seawater to yield a total amount of 0.33 ml of sodium hydroxide and 4.67 ml of seawater per gram of cysts. Cool the buffer solution to 4°C. It should be about *p*H 10. Add the cysts. Then add 10 ml of liquid bleach to the buffer solution. Use a thermometer to watch the temperature during the chemical reaction and keep the solution between 20° and 30°C. Starting with pre-cooled buffered seawater makes it easier to keep the reaction in the right temperature range. If needed, an ice cube or "ice pack" can be added to help drop the temperature.

Powdered pool chlorine can be substituted for household liquid bleach at a rate of 0.7 g of dry chlorine powder per gram of cysts. If pool chlorine is used, substitute sodium carbonate for NaOH as a buffer, adding 0.68 g sodium carbonate to 13.5 ml filtered seawater per gram of cysts. It is easier to split the water in two equal parts, adding the chlorine to the first part and the sodium carbonate to the second. Allow them to dissolve and react, which will cause a precipitate. Pre-cool the two solutions; mix them together then add the hydrated cysts. After everything is placed together, note the colour of the solution. It will change from a dark brown, to gray, to white, and then to a bright orange. This reaction usually takes 2 to 4 minutes. With the liquid bleach the cysts will change only to gray or light orange, and the reaction takes about 6 minutes.

The cysts must be filtered from the solution quickly and immediately after the membranes have dissolved (as indicated by no more colour change or the final colour-bright orange or gray); otherwise you will dissolve the whole cyst instead of only the outer shell. Washing cysts and deactivating the residual chlorine is the next step after decapsulation. The chlorine should be washed off the cysts with freshwater or saltwater until there is no more chlorine smell. The residual chlorine attaches itself to the decapsulated eggs and must be neutralized. Do this by washing the cysts in a 0.1% sodium thiosulfate (0.1 g sodium thiosulfate in 99.9 ml water) solution for 1 minute. An alternative method uses acetic acid (one part 5% vinegar to seven parts water). The first method works better, but the second method is easier because the materials are more readily available.

Obviously, the time and effort involved in decapsulating brine shrimp eggs may not suit every one. However, if you have the time and patience, it could be worth trying.

For long term storage (several months) decapsulated eggs need to be dehydrated in a saturated brine solution. A saturated brine solution is salt dissolved in water until no more can be dissolved and salt remains in the bottom of the container. The brine solution dehydrates the eggs, effectively stopping the hatching process, but the eggs hatch normally when placed in a hatching solution of lower salinity water later. (For storage, 50 ml brine should be mixed with every 100 grams of cysts.)

Since they lose their hatchability when exposed to UV light it is advised to store them protected from direct sunlight. For hatching the decapsulated eggs use a 2-litre wide-mouth glass jar, tilted at 45°, with aeration provided by a 3-mm rigid plastic airline that extends to the lowest point of the jar.



Decapsulated eggs hatch better at higher salinities (above 20‰) and are maximised at a salinity level of 28‰. You may find that decapsulated eggs require increased aeration to keep the eggs in suspension than non-decapsulated eggs, even at the higher salt level. When newly hatched nauplii are to be fed, the contents of the hatching jar are poured into a fine-meshed net. The nauplii and unhatched embryos are rinsed with fresh water and all eggs are thus utilised as food.

Decapsulated cysts can be dried by spreading eggs over a 100  $\mu$ m screen in a layer < 5 mm thick and oven dried at 35–40°C for 24 hours. Heat treatment at 40°C used in cysts preparation does not affect the protein quality of the cysts. Freeze dried decapsulated artemia cysts are available commercially and can be used for direct feeding to small fish and fry as a dietary supplement and transition from live food to pelleted diets. The cysts are not hatched and so they retain all of the nutrition inside the cyst that would normally be used by the nauplii at hatching. 100g of freeze dried decapsulated artemia is equal to 1 kg of unhatched artemia cysts.

#### **Growing Brine Shrimp**

When feeding young rainbowfishes, adult brine shrimp may be preferred over nauplii. Nauplii grow rapidly and may double in size in 20 hours or less. They continue to grow through several moults or instar stages and become adults in about 3 weeks. All that is required is a small (50-litre) aquarium filled with natural or synthetic salt water. For about 2 days after the shrimp hatch they live on the food from their yolk sac so no feeding is required until the yolk sac is depleted. Feed the brine shrimp nauplii on a micro encapsulated diet, which has a particle size of 5–50 microns. Adult brine shrimp can be fed a homemade food formula containing the following ingredients:

- 20% Brewers Yeast (Saccharomyces cerevisiae)
- 35% Soy Flour
- 35% Wheat Flour
- 10% Powdered Milk

Feeding Rate: 1 level teaspoon per 40 litres - feed again when the water has cleared.

Another suitable food for adult brine shrimp is spirulina alga powder: Pre mix a suspension of 1 level tablespoon spirulina powder per litre of distilled water. Allowed the mixture to settle for 5–10 minutes then strain through a fine brine shrimp net to remove larger particles and any detritus. The suspension can be fed to the shrimp at a rate of about 10–50 ml per 20 litres - but only if the water has cleared from any previous feeding.

When feeding the shrimp, the water needs to be a light green colour while still being able to see the bottom of the tank. Small amounts are much better than overfeeding. A bright green detritus (dust) sitting on the bottom indicates that you're putting more algae than you need into the tank and it's settling out. In that case, use a little less by aiming for a lighter shade of green water at feeding time. Light feedings will allow the shrimp to consume most of what's in the water before it can settle out. As they feed, the water will become clear again. Brine Shrimp are continuous, non-selective filter feeders and should be fed around the clock or several times a day at the very least. The food should be continuously added to the water column in the grow-out tank. The trick is to prepare the food as a solution (with light aeration) and drip the solution into the grow-out tank. Provide plenty of aeration to keep the food in suspension and maintain maximum oxygen levels. Brine shrimp populations have done well in cultures when fed algae, rice bran, soybean meal or whey powder.

Brine Shrimp are sensitive to poor water quality, such as high levels of ammonia and nitrite. The ideal on-growing tank should contain only a sponge filter. In this way the tank is easily kept clean and helps prevent any disease or water quality problems. Controlling water quality can be accomplished through a combination of biological filtration and frequent water changes. When doing a water exchange, simply catch the brine shrimp in a fine mesh net as you are siphoning out the water and immediately return the shrimp to the grow-out tank. Maintain *p*H between 8.0 and 8.5 and temperature 25 to 30° Celsius.

Harvesting of shrimp for feeding the fish is done with a standard aquarium net. The larger netting will allow immature shrimp to remain in the culture. Cultures, which are not over-harvested, can become self-sustaining in 4–6 weeks.

#### **Bioenriched Brine Shrimp**

The nutritional value of brine shrimp nauplii can decrease as much as 25–30% within 24 hours after hatching when kept at 28°C. This has meant that the nauplii must be fed to the fish fry as soon as possible after hatching or be stowed at low temperatures to decrease their rate of metabolism. Another drawback is that they have been found nutritionally deficient, especially in the long chain polyunsaturated fatty acids. However, what makes the nauplii so attractive to fish fry is their wriggling way of swimming, which acts as a powerful feeding response for fish fry.

In recent years, several enhancement techniques have been developed by the aquaculture industry to improve the nutritional value of brine shrimp by feeding them a nutrient-rich medium. Obviously, the time and effort involved in nutritionally enhancing brine shrimp may not be worthwhile for the average aquarist. However for the serious aquarist with an extensive breeding program, nutritionally enhanced brine shrimp may well be worth the effort. It is more convenient and easier to use a commercial enhancing supplement rather than make up your own concoction.

A feeding trial using *Selco*<sup>®</sup> at 300 ppm found that their fatty acid profiles changed according to the duration of the enrichment period. Newly hatched brine shrimp had 7.0 mg total fatty acids/100 mg dry weight. After enrichment for 12 hours the total fatty acids increased significantly to 10.3 mg/100 mg. After 24 hours of enrichment the total fatty acids increased to 12.6 mg/100 mg dry weight. The results indicated that the length of the enrichment process should be considered when preparing brine shrimp nauplii as a food for the larvae of ornamental fish. However, enriching nauplii will result in larger individuals than newly hatched nauplii, and may not be a suitable size for all fish larvae.



Bio-enrichment of brine shrimp began several years ago using emulsified fish oils containing highly unsaturated fatty acids (HUFAs) for marine finfish and crustacean larvae. This enabled many previously difficult species of marine food fishes to be cultured. Marine fish require Docosahexaenoic acid (DHA), a fatty acid common in marine fish oils, in their diet. DHA is virtually absent in brine shrimp although a strain from China were found to contain DHA, but they are extremely difficult to obtain.

The cause for this variability in DHA and other fatty acids is unknown, but it may well be that the types of fatty acids found in certain strains of brine shrimp are influenced by their natural diet. Freshwater fish have a limited ability to synthesise DHA from one of the omega-3 fatty acids (linolenic acid), but they too will benefit from a diet that includes DHA.

Enriching brine shrimp is essential for culturing some species of marine fish, but is not critical for most freshwater species including rainbowfishes. However, improved consistency and higher survival in larval discus, improved growth and survival of goldfish fry, and increases in fecundity of angelfishes and guppies have been reported by aquarists who are using enrichment procedures. The implications of these results are that the general health and well being of the fish fry have been significantly improved by the feeding of enriched brine shrimp nauplii. These observations still needs to be verified however, under controlled laboratory testing. Fry mortality can be the result of several factors including inbreeding, inferior water conditions and improper incubation methods.

Another role for enriched brine shrimp is feeding adult female rainbowfish for several weeks prior to spawning them. In other species, fish eggs with low levels of DHA generally have poorer survival rates to the first feeding stage than eggs that are rich in DHA. Giving females a diet high in DHA allows them to carryover excess DHA into their eggs. Essential fatty acid deficiency is not a problem with most rainbowfishes fed a varied diet. It is possible; however, that supplementation with enriched brine shrimp may increase growth rates, fecundity, and fry survival. Therefore, if you are having problems raising a particular species, it may well be worth the effort.

#### **Enriching Nauplii**

The use of brine shrimp nauplii as a delivery system for dietary enrichment or chemotherapeutics for fish larvae is developing rapidly. Commercial enrichment preparations have also been tested with other live feeds used in freshwater ornamental fish culture and have been found to have similar results.

1. Prepare and hatch brine shrimp nauplii as normal.

2. Add bio-enrichment about 12 hours after hatching. The shrimp will either ingest the supplement or it will just adhere to their body. Either way the fish will benefit from the enrichment when they are fed.

3. Feed enriched shrimp within 12–16 hours or they will have digested the enhancement formula and you will need to repeat the enrichment.

Brine shrimp nauplii are filter feeders but will not consume any supplements until after the second instar II stage (moult) begins, at about 12 hours after the nauplii hatch. The first instar nauplii do not feed. Their value as food decreases from birth until they begin feeding. To enrich live adults, just add the supplement about 12–16 hours before feeding the fish.

Bio-enriching brine shrimp with hormones has been used successfully to induce ovulation in *Paracheirodon axelrodi*. Exposure of the brine shrimp to the hormones for between 30 and 60 minutes was optimal for inducing ovulation while shorter and longer periods showed a trend to a decreasing percentage of ovulating fish.

Because brine shrimp are non-selective and continuous filter feeders, almost anything will be consumed, as long as the particle size is between 5–50 microns. Any supplements must be in a non-soluble form as brine shrimp do not "drink" soluble components.







# **Mosquito Larvae**

Mosquito larvae grow in aquatic habitats and provide a good natural live food for fish and other aquatic predators. Their nutritional value has been fairly well recognised. Due to presence of high micronutrients and all the essential amino acids, the larvae (and pupae) may be used as a live or processed (frozen) food for feeding rainbowfishes. Furthermore, mosquito larvae have a high protein content (~50%).

Rainbowfishes are well adapted to capturing live mosquito larvae, and will show an active response to this type of food, indicating that mosquito larvae are a very attractive food for rainbowfishes. Upon adding mosquito larvae to the tank, it is quite often the case that rainbowfishes will exhibit an immediate response that somewhat resembles a feeding frenzy. Live food is preferred over inert food by rainbowfishes, suggesting a moving prey item may stimulate or influence feeding preference. Rainbowfishes seem to prefer mosquito larvae to all other live foods, and their effortless method of culturing or collection makes them an ideal live food.

Mosquitoes have four aquatic instars, and a final or fifth stage being a non-feeding pupal stage. Adults will emerge after 2 to 3 days from pupae. The eggs may be laid singly or in rafts, deposited in water, on the sides of containers where water will soon cover, or on damp soil where they can hatch when flooded by rainwater or high tides - even small water bodies such as the bottom trays of pot plants are used. Australia has about 350 species of mosquitoes. It is only the females that seek animals out for their blood, which they need in order to reproduce. Females live for about a month while males often only live for a week, during which they feed on nectar.

Mosquitos have been cultured for many years by aquarists in numerous parts of the world. There are numerous reports on this subject, and many aquarium hobbyists have developed their own technique. The mosquito usually lays its eggs in water containing decomposing organic matter. Often the adult mosquitoes are attracted by the odour of decomposed material. The larvae can easily grow even without the presence of dissolved oxygen, due to the presence of air sacs in which larvae can store air containing oxygen for a long period and use the oxygen for respiration.

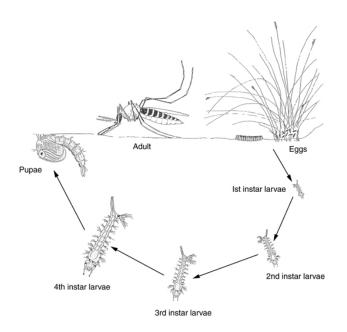
Culturing mosquito larvae is fairly straightforward. First established a tub or container outside where it will obtain partial shade. Shallow containers with a large surface area seem to be preferred to deep containers. Fill the tub with preconditioned freshwater. The best source of water is filtered stream or pond water; collected rainwater, or use the water from your aquarium water changes.

Next, add some animal manure or organic fertiliser. Fresh material is preferred over old because they are richer in microbes and organic matter. This especially applies to manure, which is usually dried before use. The fertiliser can be added to your culture in several ways. One is to soak the dry material for several hours, then distribute the wet material over the bottom, allowing it to slowly decompose. Another is to place the dry material in a mesh bag and suspend the bag inside the tub. Other substances such as crushed dry dog food or fish food pellets can also be used.

Sometimes a scum will form on the surface of the culture container, which can affect the earlier instar larvae as well as the pupae. The nature of such scum formation is undoubtedly influenced by various conditions of the culturing container, for instance water temperature, nature and amount of organic matter, and number of larvae. Generally speaking, high temperature, fatty or large-sized particles, excess of added organic matter, and high larval density may result in scum formation.

After a few days when the water has cleared, add a handful of duckweed to afford the mosquitoes a place upon which to rest while laying their eggs in the water. The elongated eggs look like little black rafts and are about 6 mm wide. Each raft contains from 50 to 500 eggs, depending partly on how much blood the female has fed on, and one female may lay





several batches. If the female doesn't get blood she is still able to produce eggs, but the eggs will be smaller and have less chance of survival. In warm water, the eggs of most species will hatch in two to three days and the larvae or "wrigglers" feed mainly on algae, protozoans, and organic matter.

Once you observe egg-floats or rafts, spoon them into small glass jars containing about 5 cm water and cover with a fine mesh. Later, when the eggs hatch into larvae, merely pour the contents of the jar into the aquarium. Then again, if you require larger larvae or pupae, place the eggs-rafts in a larger jar until the desired size is reached. The larvae are tiny and must moult five times before reaching the adult stage of life. The larvae hang upside down on the underside of the water surface. The tail of the larvae has feather like structures, which allow it to hang onto the surface using the water surface tension. The mosquito larvae have an interesting way of breathing. They have a breathing tube that reaches from the tail to the water surface. This tube is used in much the same way humans use a snorkel to breathe while remaining underwater.

Sometimes the culture will be so successful that if all the larvae were fed at once to the fish not all would be eaten and the mosquitos will hatch causing at the least some nuisance. In this event you can either place some of the larvae into containers in the refrigerator where their metamorphosis will be slowed down or preferably frozen and fed to the fish during the winter months. Other species of insects produce different larvae such as bloodworms and glassworms, which may be found in the container and can also be fed to the fish.

It is far preferable to breed mosquitos in the manner outlined above than to permit the eggs to hatch in the outside tub. It is inevitable, that some will manage to complete their metamorphosis and escape to plague you and your neighbours. Some mosquitos are capable of transmitting diseases such as encephalitis, malaria, and fever.

# **Bloodworms**

Bloodworms are larvae of the mosquito-like midge family Chironomidae. Almost all chironomids have aquatic larval and pupal stages. They have a world-wide distribution and occur in a wide range of aquatic habitats from fast flowing to completely still and stagnant, and in waters that range from fresh to saline. There may be more than 2000 species but only a small number have been formally identified. There is little difficulty in recognising the larvae; they are small, distinctly segmented worm-like animals. Although, they are not a true worm due to their exoskeleton and small clawed legs. Their colour is variable; some common ones are white, green, yellow, or deep red. The last is due to the presence in the larval blood of a red pigment, erythrocruorin, the presence of which is of respiratory advantage in waters with low levels of dissolved oxygen (stagnant pools). Some are also transparent and are commonly known as Glassworms. However, only those that contain erythrocruorin are red and hence the common name "bloodworm". In some countries they are also known as red mosquito larvae.

Chironomidae larvae and pupae are highly nutritious and nourishing and constitute one of the staple food items of rainbowfishes in their natural environment. Chemical analysis (% of dry weight) shows that bloodworms contain 71–93.6% moisture, 47.7–62.5% protein, 4.9–28.6% fat, 2.3–21% ash, and 4–23% carbohydrates. They are also a good source of iron for the fish since they contain haemoglobin.

Bloodworms are a commonly used live or frozen food source for aquarium fish culture. Almost all fishes will greedily devour them when they are offered. Research has found that most fishes when provided with bloodworms as a supplementary food item have better growth and spawning rates. Frozen bloodworms could be used as a substitute for live tubificids as they have a comparable protein level.

Chironomidae go through a complete metamorphosis in their life cycle, egg, larva, pupa, and winged adult midge. Each stage having different characteristics. After mating in flight the female releases the eggs while skimming the water surface. Egg numbers can range from 50 to 700. The eggs sink to the bottom where, under tropical conditions, they hatch in 24–48 hours into the next stage - the larva or aquatic stage. The newly hatched larvae are not more than 1 mm long but they can measure up to 10–25 mm when they reach the last stage of the larva period. The larva stage can last from less than 2 weeks up to 7 weeks depending on temperature.

Each larva moults four times before it reaches the pupal stage. This stage of the chironomid forms a large part of a rainbowfishes natural diet as they leave the larval tube and actively swim to the surface of the water. Those that reach the surface emerge into flying adults after a few hours and immediately fly off to mate, living only a few hours or days. The adults do not feed during their adult existence and mating normally occurs during the night. The entire life cycle can be completed in 2 weeks, although it is common for the life cycle to take longer to complete.



Midge larvae can be found in most waters with muddy bottoms. They occur in great numbers in ponds, swamps, and streams, usually 1–4 metres deep. Natural breeding sites for chironomid midges are diminishing due to urbanisation, land clearing and other changes to much of the natural environment. However, they are abundant in waste water channels, sewage treatment and settlement ponds, and other man-made water systems. It is these breeding areas that cause a variety of nuisance problems and public health agencies regularly spray these areas with insecticides to control their population and distribution.

If you wish to collect bloodworms the best time to catch them in large numbers is during the night when the larvae leave their self-made tubes and when the dissolved oxygen at the bottom of water is low. They can be caught easily using small mesh netting. Bloodworms can also be obtained by sieving the mud on the spot. The larvae and the coarse particles of detritus will remain in the sieve and then shaken into a bucket filled with water. After a while the larvae will swim to the surface where they can be fished out with a net.

#### Remarks

Attempts to propagate bloodworms have been carried out in many countries without much success. The major problem is the inability to induce swarming and mating of the chironomid midges in captivity. However, there is now some successful cultivation of bloodworms in Southeast Asia.

There have been some reports of fish with swollen abdomens and obstructions from being fed on bloodworms. The concern seems to be related to the structure of the bloodworms "undigestible" exoskeleton and its potential for creating stomach blockages.

"A note of caution here on feeding the larger imported frozen bloodworm. These worms have a chitinous exoskeleton and numerous bristles that are indigestible for fishes with small intestinal apertures. The meaty portion of the worm is readily processed, but the hard bits remain and clog up the stomach in an immovable mass. Be careful not to feed your discus, rainbows and some tetras on the larger bloodworms."

 $\sim$  Dr Jim Greenwood BVSc. Canterbury Veterinary Clinic, Victoria.

I have fed frozen bloodworms to rainbowfishes for many years and have never experienced this problem. However, there have been a number of reports on larval mortality of freshwater fish species when being fed live bloodworms.

This is because the pupae contain a chitinous body covering which is not digestible in the early larval stages of some fish species due to a lack of a chitinase enzyme in the stomach. The chitinase enzyme develops at a later stage of fry development, which helps them to digest the chitin. Usually early post-larvae cannot produce this enzyme.



Mortalities of post-larvae can occur due to not digesting the chitin as well as some toxic effects of chemicals of chitin (Steffens 1989). The chitinous body covering remain very soft up to the 3rd instar stage which is easily digestible, but becomes harder in the 4th instar and very hard in the pupal stage. Mortalities have been reported when chironomid larvae of 4th instar and pupae were fed to 4–5 days old *Clarias batrachus* post-larvae (Habib *et al.* 1993). They found intact body coverings of chironomid larvae and pupae in the intestine of *C. batrachus* post-larvae which was not digested due to absence of the chitinase enzyme.

Chitin is the principal structural component of the exoskeletons of invertebrates such as Crustacea. It is also present in the cell walls of most fungi and many algae. Endogenous chitinase, the enzyme that degrades chitin, has been described in the digestive tracts of a number of finfish (Lindsay and Gooday, 1985) and so it can be considered that chitin is available to these animals. However, the efficiency with which it is digested is unknown.

It is also claimed that some bacteria in the digestive tract of fishes participate in chitin digestion. However, enzyme activities measured in the stomach, intestine, blood, liver and lymphomyeloid tissues have all been determined to reflect endogenous enzyme capabilities of fishes, and teleost chitinase genes have been found leaving little doubt that the majority of chitinolytic activity in their digestive tracts is produced by stomach and intestinal mucosa.

There have also been some reports of respiratory and/or skin allergy to Chironomids. Cases of aquarium hobbyists developing conjunctivitis, rhinitis with or without bronchial asthma or urticaria after handling fish foods have been reported. The aetiological agents were chironomid larvae, and in the some cases, shellfish/crustaceans present in some fish foods. However, studies suggest that Chironomid allergies are rare and are mainly seen in those who handle bloodworms used for fish foods.



# Drosophila

Drosophila are small flies about 3-mm long, of the kind that accumulates around spoiled fruit. They are cultured in public Aquariums and Zoos around the world and are one of the most popular feeder insects for a variety of captive amphibians, reptiles, and fishes. The adult flies and their larvae can provide rainbowfishes with an excellent variation to their diet and supply natural vitamins and fatty acids they otherwise may not get in large enough quantities.

Drosophila melanogaster is one of the better-known species and is cultured in laboratories around the world. It is used extensively in biological research, particularly genetics and developmental biology. Intensive biological research into Drosophila melanogaster isolated a genetic mutation that only had rudimentary wings and could not fly; the mutation bred true and was further selectively bred so that a totally wingless variant was established.

The reproduction time for *Drosophila melanogaster* is rather quick and small cultures can yield large amounts of flies. Another species (*Drosophila hydeii*) is also cultured. *Drosophila hydeii* is 1.5–2 times larger than *Drosophila melanogaster* with red eyes and full wings, but their wing muscles are genetically impaired. Both species are prolific breeders, producing hundreds of offspring during their life period. Neither species can fly but they hop, climb, and attempt to fly.

*Drosophila melanogaster* has a shorter life cycle (14–15 days) than *Drosophila hydeii* (approx. 25 days). Female Drosophila lay an egg about half a millimetre long. It takes about one day after fertilisation for the embryo to develop and hatch into a worm-like larva which can be seen in the medium or on the inside of the culture jar.

The larva eats and grows continuously, moulting on day one, day two, and day four after hatching (first, second and third instars). After two days as a third instar larva, it moults one more time to form an immobile pupa. Over the next four days, the body is completely transformed to give the adult winged form, which then hatches from the pupal case and is fertile after another day. From laying the eggs, hatching, and the larva emerging and pupating can take six to ten days dependent on temperature. The pupae require another six days before the adult fly emerges and the females lay eggs and the cycle begins again. The flies can live from 20 to 30 days and the female will continue to lay eggs until she dies.

Culturing Drosophila is remarkably simple, but make sure that you obtain a culture of the wingless variety. Fly generations will usually breed true as long as no winged (wild) fruit flies infiltrate your cultures.

The normal method of raising the flies is to place a layer of medium on the bottom of a wide necked jar. Add 10–20 flies (females have a black band on the abdomen, which is slightly pointed), sit back and await the results.

Suitable containers for culturing include commercial fruit fly vials or flasks, open-mouth glass jars, and other small bottles (glass is better than plastic). The jars must be very clean, preferably sterile, or you could end up culturing a host of moulds, instead of fruit flies. The jars can be washed and then sterilised in a microwave oven. Cover the tops with a piece of paper towelling or fine mesh netting held in place by a rubber band. Small glass jars with plastic lids are very suitable. A small opening can be cut out of the plastic lid and covered by a piece of netting or fine mesh either glued or held in place with tape. This provides ventilation and prevents regular fruit flies access to the culture as they will breed with the wingless variety and ruin the culture.

Maintain the cultures at between 20 to 30°C. The optimum temperature for good reproduction without accelerating mould growth is 24–25°C, below 20°C; development takes twice as long. Many culturists suggest placing some sort of stiff plastic screen in the jar to give the flies a dry area to sit on and as a surface for the larvae to pupate on. The additional surface area not only provides somewhere for the flies to roost but also increases the quantity of flies considerably. However, supplying a roost is not entirely necessary as the adults and larvae will just use the sides of the jar.

As soon as the larvae are seen in the medium or climbing the sides you can feed the adult flies to your fishes. When adult flies hatch from the pupae, allow a day or two for them to lay eggs and use the first hatch to start new cultures. Once you have your new cultures set up the flies can be fed to your fish. Each jar will provide flies for two or three weeks before requiring cleaning and setting up again. Feed the flies to the fish daily from then on, as production will suffer if too many adult flies remain in the culture.

During the warmer months of the year you may get an explosion of larvae, and these can be harvested and fed directly to the fish which relish them. At anytime that you find the Drosophila are breeding at a faster rate than you can use them, it is a good idea to place them in a small plastic bag and freeze them for future use when supplies are less plentiful.

Culture jars are always under attack from unwanted predators. If your container is left open and receives a flying fruit fly that contaminates your existing culture, it becomes the dominant fly and that culture must be destroyed. If your cultures start to contain mites, just isolate those cultures and use the adult fruit flies for food only. Adult fruit flies often carry microscopic mites on their wings, which usually cause them no harm. Mites can, however, attack the eggs and larvae. They can be transmitted from one culture to another if not caught early on.

There is quite a lot of debate among culturists regarding which media is best, and like everything else, what works for some, may not work for others. There are a variety of homemade and commercial media available to use. Special culture mediums are available from biological suppliers or live food dealers. Following are a couple of simple recipes that can get you started. Other recipes are often available from Reptile and Herpetology magazines or books.



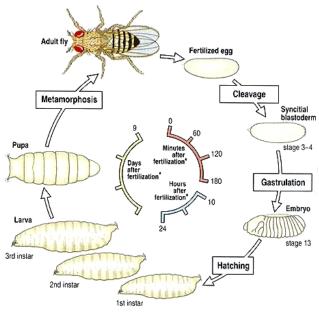
#### Recipe #1:

Equal parts banana and apple sauce - mixed to the consistency of baby food, oatmeal is then added until the mixture is stiff enough to stay in the bottom of a culture vessel. Inoculate with a sprinkle of yeast and add flies. It is reported to yield huge numbers of flies, but can have an odour that some culturists find unpleasant.

#### Recipe #2:

Mix one tablespoon of sugar with one cup of dried instant mashed potatoes, available from any supermarket. Add one inch of this blend to a wide mouth jar. Pour in water to the same level as the potato-sugar. Sprinkle a pinch of dried yeast on the surface of the mash.

There are several methods of harvesting the flies to feed to your fishes. Flies can be tapped directly onto the water surface from an established culture where the culture medium is still stiff. Where there is a risk of shaking out culture medium into the tank then simply place a jar of the same size on top of the culture and shut off the light to the culture jar. Drosophila are phototropic - that is they are attracted by light. To get the flies to leave the culture jar you should remove the jar's cover, place another jar mouth to mouth, above the jar containing the flies, then cover the lower jar completely with a dark cloth or paper to exclude as much light as possible. The flies migrate towards the light and when sufficient flies are in the top jar simply upturn and recap the culture. The flies in the top jar may now be shaken onto the water surface or as some aquarists prefer mixed with a small quantity of aquarium water and poured into the tank.



"At 25°C incubation

The body of the fly is soft and floats on the surface of the water, where it can live for several hours until eaten. Most fishes soon become accustomed to eating the flies and will feed voraciously on them floating on the water surface. In fact some species will literally jump out of the water to get at them, particularly Archerfishes. Both the fly larvae which are very good for smaller fishes and the flies can be fed.





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# Norbert Grunwald

# Daphnia

Daphnia (pronounced daff-knee-uh) are members of the order Cladocera (cladocerans), and are one of the several small aquatic crustaceans commonly called water fleas due to their small size and jerky swimming motion. They live in various freshwater environments ranging from swamps, lakes, ponds, streams and rivers. They are usually translucent or amber in colour and are an important component of the food chain in freshwater environments. Most species occur worldwide and are truly cosmopolitan, but on the other hand there are several that are endemic in distribution.

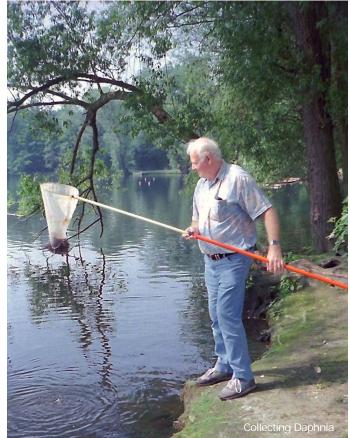
The body of daphnia is shaped rather like a clam with a slit along the belly side. A single eye capable of distinguishing light from darkness is present in the head. If a shadow is cast across a daphnia pond, the animals quickly swim out of the shadow into the light. Two large appendages behind the head (actually modified antennae) are used like oars to propel the animal in a jerky fashion through the water. Other appendages are inside the shell, and they are used to filter suspended particles on which the daphnia feed.

Most daphnia are cyclical parthenogens, so are well adapted to short seasons of reproduction such as in seasonal environments. Natural populations are composed of females who clonally reproduce until conditions within their environment deteriorate. One clutch of eggs is normally released into the brood pouch during each adult instar (an instar is the period between moults). Since these animals are crustaceans, the only way they can grow in size is to moult their exoskeleton. As long as environmental conditions remain favourable females will continue to reproduce in this manner, producing only female offspring capable of asexual reproduction.

When unfavourable conditions occur, e.g. severe temperature changes, drying, or crowding which leads to competition for food or a decrease in the quality or size of food, then the production of parthenogenetic eggs declines. Some eggs develop into males and females capable of sexual reproduction. These females have modified carapaces, which are thicker and darker dorsally than a regular carapace and produce haploid eggs that must be fertilised by the males. The fertilised egg goes through several cell divisions, the zygote enters a resting stage and cell division stops. Males are slightly smaller and different in form compared to females.

The mechanisms underlying the production of males and haploid eggs are not clear. Male production seems to be correlated with crowding and a rapid reduction in food supply (a constant low food supply simply inhibits reproduction). Short-day photoperiod seems to increase the production of ephippia in *Daphnia pulex* in contrast to the longer light periods of midsummer, i.e. as autumn approaches and days get shorter, and the number of ephippia produced seems to increase.

After these haploid eggs are fertilised by the males, the wall of the brood pouch thickens and encloses the eggs in a semi-elliptical saddle-shaped ephippium. These ephippia contain embryos in a state of arrested development. When the female moults the



ephippium is cast off. It does not disintegrate, but remains intact protecting the eggs, which will not complete their development and hatch until favourable conditions return.

Daphnia can suspend their growth and development for years or even centuries during periods of unfavourable conditions. In habitats, which dry completely, annual recruitment is entirely from the ephippia. Ephippia sink or float and are able to withstand extremes of temperature and moisture. When environmental conditions are again favourable the embryos develop into parthenogenetic females and break free of the ephippium. The ephippia are also used as a means of dispersal for many species, being carried by the wind or in the fur, feathers or digestive tracts of animals to new habitats.

The life span of daphnia, from the release of the egg into the brood chamber until the death of the adult, is highly variable depending on the species and environmental conditions. Generally the life span increases as temperature decreases, due to lowered metabolic activity. The average life span of *Daphnia magna* is about 40 days at 25°C, and about 56 days at 20°C. The average life span of *Daphnia pulex* at 20°C is approximately 50 days. The time required to reach maturity (produce their first offspring) in *Daphnia pulex* varies from six to 10 days (mean = 7.78 days) and appears to be dependent on body size.

The growth rate of the organism is greatest during its juvenile stages (early instars), and the body size may double during each of these stages. *Daphnia pulex* has three to four juvenile instars, whereas *Daphnia magna* has three to five instars. Each instar stage is terminated by a moult. Growth occurs immediately after each moult while the new carapace is still elastic.



Feeding live daphnia to rainbowfishes results in a remarkable improvement in their colouration, and health. Rainbowfishes fed an exclusive diet of daphnia grow rapidly and breed readily. Daphnia are a high bulk food; that is, a high proportion of the animal's body is covered with a chitinous exoskeleton - a sort of skeleton on the outside. This chitinous material provides food high in fibre but low in nutrients; its bulk stimulates peristalsis.

There are many species of daphnia but the most common ones cultured in the aquarium hobby are *Daphnia magna* and *Daphnia pulex* although numerous other species have been cultured successfully. The taxonomy of daphnia species is very confused and it can be very difficult to distinguish one species from another. The characteristics used to separate these species are extremely variable and many intermediate forms occur.

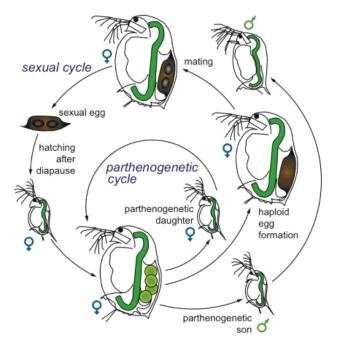
Daphnia can be easily cultured if suitable water conditions and food is provided. The best source of water is filtered stream or pond water or rainwater collected for low air polluted areas. Never use distilled or deionised water, as it does not have the minerals needed for growth. Daphnia are very sensitive to dissolved contaminants like chlorine, chloramine, and copper. *Daphnia magna* are quite resistant to phosphorus and can withstand concentrations as high as 5–7 ppm and are usually not affected by the addition of nitrogenous fertilisers for the promotion of algae growth in outside ponds.

Daphnia have a wide tolerance to temperature but grow best at a temperature range of  $18-22^{\circ}$  Celsius. Moina, on the other hand, withstand extreme temperatures better, resisting daily variations of  $5-31^{\circ}$  Celsius. Their optimum temperature range is  $24-31^{\circ}$  Celsius. This higher temperature tolerance makes this species a better choice for culturing in area where temperatures may rise above  $22^{\circ}$  Celsius. A *p*H between 6.5 and 9.5 is acceptable.

Starter cultures of daphnia can be collected from the wild but beware of predators like hydra and various carnivorous insect larvae. Obtaining a clean culture from a fellow hobbyist or biological supplier is the preferred source for a starter culture. Immerse the starter culture into your culture water and gently empty the contents (don't pour the starter culture through the air).

Daphnia magna can survive when the dissolved oxygen concentration is as low as 3 mg/L but Daphnia pulex does best when the dissolved oxygen concentration is above 5 mg/L. Therefore it is recommended that the dissolved oxygen concentration in the culture be maintained at 5 mg/L or above. Unless the cultures are too crowded or overfed, aeration is usually not necessary. Daphnia magna is principally a lake dweller, but can also be found in shallow ponds with muddy bottoms rich in organic matter. Daphnia pulex is principally a pond dweller where the oxygen content is higher, but is also found in lakes. It is generally considered a clean water species being dominant in nature during periods of low turbidity.

Daphnia can be successfully raised indoors maintained in any suitable container, although better results will be obtained when they are cultured outdoors. Do not expose indoor cultures to direct sunlight or strong artificial light.



Life cycle of a cyclic parthenogenetic Daphnia. This diagram depicts the sexual and the asexual (parthenogenetic) life cycle of a Daphnia. During the parthenogenetic cycle, females produce diploid eggs that develop directly into daughters. The same female may produce diploid asexual eggs that develop into sons. Male production is under environmental control. Furthermore, the same female may produce haploid eggs that require fertilization by males. These eggs are then enclosed in a protective shell (ephippia) and need to undergo a diapause before female offspring will hatch from them.

The variations in ambient light intensities and prevailing day/night cycles in most fishroom situations do not seem to affect daphnia growth and reproduction significantly. However, a minimum of 16 hours of illumination should be provided each day.

Start your culture with approximately 25 daphnia / litre. Although a culture can theoretically be started with a single female, always use an adequate number to develop a harvestable population quickly. If fewer are used, the population in the culture will increase more slowly; therefore, the initial quantity of fertiliser or food should be reduced to prevent overfeeding. A greater number used for inoculation reduces the time to harvesting and lessens the chance of contamination by competitors.

Cultures are usually inoculated 24 hours or more after fertilisation. However, when yeast is used, daphnia can be added to the culture after a few hours of aeration, assuming good water quality, and proper temperature. This is because the yeast cells are immediately available as food. The small amount of phytoplankton present in the water and digestive tract of the daphnia used to inoculate the culture is usually sufficient to initiate a phytoplankton bloom. Sometimes the mortality of the initial inoculation is high and an additional inoculation is required.

The health of the culture is determined by stirring the culture, removing 15 ml of the culture, and examining the sample with a 10X-hand lens or dissecting scope. Green or brown-red daphnia with full intestinal tracts and active movement indicates a healthy culture.





Pale daphnia with empty digestive tracts or females producing resting eggs are indications of sub-optimum environmental conditions or insufficient food.

The food concentration in the culture water, when examined in a clear glass, should appear slightly cloudy and tea coloured or green. Clear culture water is an indication of insufficient food. Feeding the proper amount of the right food is extremely important in daphnia culturing. The key is to provide sufficient nutrition to support normal reproduction without adding excess food, which may clog the animal's filtering apparatus, or greatly decrease the dissolved oxygen concentration and increase mortality.

Daphnia are herbivores or detritivores, feeding on phytoplankton, bacteria, or decaying organic material. They are well adapted to live in algal blooms, which are high in proteins and carbohydrates. Small particles in the water are filtered out by fine setae on the thoracic legs and moved along a groove at the base of the legs to the mouth. Although there is some evidence that certain types of food, such as particular types of algae, protozoa, or bacteria may be selected by some species, it is generally believed that all organic particles of suitable size are ingested without any selective mechanism.

You can feed your indoor culture sparingly with brewer's yeast, powdered milk or egg yolk, astaxanthin, powdered chlorella algae or hard-boiled egg yolk squeezed through a piece of cloth. Another satisfactory food is spinach juice. It has a minimum of indigestible fibrous material and can be easily made from frozen spinach in a food blender. Keep the spinach juice refrigerated and feed once or twice a day to an active culture. Powdered spirulina is another suitable food for feeding daphnia: Pre mix a suspension of 1 level tablespoon spirulina powder per litre of distilled water and fed at a rate of about 10-50 ml per 20 litres - but only if the water has cleared from any previous feeding.

Care must be taken not to overfeed with these foods, as overfeeding can quickly cause problems with water quality. Regardless of the type of media used, start with small amounts of feed or fertiliser added at frequent intervals; slowly increasing the amount used as you gain experience.

Well-established cultures reproduce very quickly and can quickly become crowded in culture tanks. High population densities of daphnia can result in a dramatic decrease in reproduction, but this is not apparently the case with moina. The maximum sustained density reported in cultures of daphnia is 500 individuals per litre. Moina cultures, however, routinely reach densities of 5000 individuals per litre and are, therefore, better adapted for intensive culture.

The batch culture method of producing daphnia uses a continuous series of cultures. Briefly, a new culture is started daily in a separate container using the procedures outlined below. When all the yeast, bacterial, or algal cells are consumed, usually about 5 to 10 days after inoculation, the daphnia are completely harvested, and the culture is restarted. This method is particularly applicable when a specific quantity of daphnia is needed each day, because daily production is much more controlled. Batch culture is also useful for maintaining pure cultures because there is less chance of the

cultures becoming contaminated with competitors (e.g., protozoans, rotifers, copepods) or predators of fish larvae or fry (e.g., hydra, dragonfly larvae).

Semi-continuous cultures can be maintained for two months or more by daily partial harvests, water changes, and regular feeding, keeping the population in a state of rapid growth. Eventually, the culture will fail to respond to additional fertilisation. When it is evident that they are not reproducing well, the daphnia should be completely harvested and a new culture started.

Daphnia can be produced either in combination with their food or as separate cultures. Combined culture is the simplest, but production from separate cultures has been reported to be approximately 1/3 higher. Production from separate cultures has the disadvantage of requiring additional containers for the cultivation of phytoplankton. Regardless of the culture method, always maintain several cultures to ensure a supply in case of a die off.

It is important to do this because once the population of daphnia becomes too crowded or the water too old or not acceptable to the daphnia for whatever reason, the females start producing male offspring and only ephippial eggs are produced. These eggs will not hatch until more favourably conditions return so your culture may just fade away.

Cultures can be maintained in 50-litre aquaria. However, this volume is usually too small to yield enough daphnia to satisfy demand. Tanks or vats (concrete, plastic, or fibreglass), and earthen ponds can be used. Wading pools, old bathtubs, discarded refrigerator liners, and cattle watering troughs also work well. Water depth should be no greater than 45 cm. The shallow water depth allows good light penetration for photosynthesis by phytoplankton. Diffuse light or shade over 1/3 of the water surface of the daphnia culture container is recommended. A greenhouse covered with shade cloth (50-80% light reduction) is ideal.

Outdoor cultures should be protected from heavy rain and screened to prevent entry of predacious aquatic insects. Filamentous algae and predators of fish fry can be especially troublesome in daphnia cultures. Gentle aeration can be used to oxygenate the water, keep food particles in suspension, and increase phytoplankton growth, but fine bubbles should be avoided.

A partial harvest every day is required to keep the culture healthy and productivity high. They can be harvested by simply dipping out the required number with a brine shrimp net as they concentrate in "clouds" at the surface. Cultures may also be harvested by draining or siphoning the culture water into a plankton collector equipped with 50 to 150  $\mu$ m mesh netting net suspended in a container of water. Turn off the aeration and allow the food particles to settle before harvesting. For semi-continuous culture, do not harvest more than 20 to 25% of the population each day, unless you are restarting the culture. Harvesting by draining the culture tank allows for a partial water exchange, improving water quality.

Daphnia maintained in outdoor ponds will feed on phytoplankton present in water, which is very rich in organic or nutrient matter. A container or pond outside that gets plenty of sun will virtually guarantee alga soup and a successful daphnia culture. The pond is filled with water, fertilised with organic matter, and allowed to stand until it turns green before a starter culture is introduced. In about three weeks, the pond should reach maximum population and the daphnia can start to be harvested.

Organic fertilisers are usually preferable to mineral fertilisers. Mineral fertilisers may be used and usually work better in earthen ponds than in tanks or vats, but can be toxic to some species of daphnia. Organic matter (manure) provides bacterial and fungal cells and detritus as well as phytoplankton as food for the daphnia. This variety of food items more completely meets their nutritional needs, resulting in maximum production. Although not necessary, the manure is frequently dried before use. Although manure is widely used to culture daphnia, micronised rice bran, and agro-industrial wastes, eg wheat bran, soybean meal, and dried blood are less objectionable to use and work well.

The fertiliser can be added to your culture in several ways. One is to soak the dry material in water until it breaks down into a thin mixture, then pour the resulting concoction into the pond, allowing it to slowly deteriorate. Another is to place the dry material in a mesh bag and suspend the bag inside the pond. Change the bag every five days. Cheese cloth, hessian, muslin, nylon, or other relatively loose weave fabrics may be used.

Nylon and other synthetic fabrics, however, do not deteriorate in water as do cotton or hessian. For smaller culture containers, nylon stockings work well for this purpose.

The use of a bag prevents large particles from being a problem when the daphnia are harvested and allows greater control of fertilisation. If fungus occurs in the culture container due to over-fertilisation, the bag containing the organic material should be removed from the culture. If fungus persists in large quantities, the culture should be discarded and restarted.

The nutritional content of daphnia varies with age, and what it has been eating. The protein content is usually around 50% of dry weight. Quite the opposite from brineshrimp, adults normally have a higher fat content than juveniles do about 20–27% for adults, and 4–6% for juveniles. Some species have been reported to have protein contents exceeding 70%.

The fatty acid composition of food is important to the survival and growth of fish fry. Omega-3 highly saturated fatty acids are essential for many species of fish. Daphnia cultured on brewer's yeast (*Saccharomyces cerevisiae*) are high in monoenoic fatty acids. By using what is called w-yeast (yeast enriched with cuttlefish oil), daphnia will contain very high levels of omega-3 fatty acids.

Commercial formulas can also be obtained from aquacultural suppliers for the enrichment of brineshrimp, rotifer, and daphnia cultures.



Differences in size, brood production, and optimum environmental conditions exist between different species and varieties of daphnia. Adjustments will need to be made in the culture technique depending on the particular species or variety you wish to produce. It may not always be possible to match production to the food demand of the fish fry. Harvested daphnia can be kept alive for several days in clean water in a refrigerator. However, the nutritional quality of the stored daphnia probably will not be optimal because of the period of starvation, so they should be enriched with algae and yeast before feeding them to the fish.

Daphnia can be stored for long periods by freezing them in flatpacks. Try to make as thin a layer as possible in order to achieve quick freezing. Otherwise it will take too long for the animals to get frozen resulting in the formation of ice crystals, which then puncture the cell walls lowering the nutritional value of the daphnia dramatically. Use plastic bags in order to prevent freezer burn and try to lower the temperature of the freezer compartment. With some practice you should end up with correctly frozen daphnia retaining their nutrients.

Although freezing does not significantly alter the nutritional content, nutrients do leach out rapidly into the water. Nearly all of the enzyme activity is lost within ten minutes after introduction in fresh water. After one hour, all of the free amino acids and many of the bound amino acids are lost. Adequate circulation is required to keep them in suspension after thawing so they will be available to the fish. But of course, live daphnia are better as their movement arouses the hunting behaviour of many fish.

Some "daphnia" cultures are not daphnia at all, and upon further investigation other cladoceran genera such as *Ceriodaphnia*, *Daphniopsis*, *Bosmina*, *Bosminopsis*, *Moina* and *Moinodaphnia* can be recognised. However, as they all are the same to culture and eagerly accepted by all rainbowfishes, it will not matter which species you have; although, there is considerable size variation between the different genera.

Moina, for example, are approximately half the maximum length of daphnia. Adult moina (700 to 1000  $\mu$ m) are larger than newly hatched brine shrimp and approximately two to three times the size of adult rotifers. Young moina (less than 400  $\mu$ m), however, are smaller than newly hatched brine shrimp and approximately the same size or slightly larger than adult rotifers. Therefore, moina are ideally suited for feeding rainbowfish fry, and many species can ingest newly hatched moina as their initial food.

Cladocerans occur throughout the world, even in Antarctica. Most are freshwater animals, but some species are estuarine and marine. Eight families (Daphniidae, Moinidae, Macrothricidae, Chydoridae, Sididae, Bosminidae, Sayciidae and Ilyocryptidae) and over 170 species of Cladocera are known from Australian inland waters. Daphnia is the most commonly known genus in this group.

# Summary

The development of commercially manufactured fish food could be considered as one of the contributing factors to the tremendous growth of the aquarium hobby over the past 50 years. However, most information on the nutrient requirements of aquarium fish is derived principally from research carried out by the aquaculture industry. These results do however, have limitations in their applicability for feeding rainbowfishes because it is based on intensive cultured conditions aimed at maximum growth in a short time period. This might be of value for the commercial farming of aquarium species, but would be unsuitable for rainbowfishes kept in home aquaria.

Not only do rainbowfishes have different nutrient requirements, but also the digestibility of various components of the diet will differ depending on their natural diet. In nature, rainbowfishes feed mainly on aquatic insect larvae, microcrustaceans, food from terrestrial origin (mainly terrestrial insects), and many other items that they encounter in their environment. When confined to an aquarium these supplemental food items are no longer available.

In captivity their survival dramatically depends on exogenous food. Hence, complete and balanced nutrition is critical for their wellbeing. However, despite the wide range of food items consumed in their natural diet, under aquarium conditions they are forced to feed on a very limited number of food items (two or three) which frequently are not part of their natural food and hence their nutritional composition is not always the most suitable for maximum growth, development and survival.

The nutritional requirements of most rainbowfish species is poorly understood. Rainbowfishes are essentially omnivorous and for this reason, it is a good management practice to feed a variety of foods to captive fish. Omnivores, by definition, eat a wide variety of food items including plant and animal material, and some detritus as well. Physical characteristics of the diet also play an important role. Food particles need to be small enough for the smaller species to ingest, but large enough to be identified and eaten by the larger species. The availability of a suitable diet is critical for successful transition of larvae to juvenile and juvenile to adult stages.

Moreover, the development and survival of rainbowfishes in the wild undergo several morphological and physiological changes which in nature are simultaneous with changes in behaviour and even habitat and type of food consumed. All these changes will affect to nutrient availability and feed utilisation by the fish in order to match their nutritional requirements. In practice, most of these problems could be simplified by the proper development of inert diets which are able to cover nutritional requirements at different stages of development. In order to achieve those diets we need, among many other important things, is to have a complete knowledge of the nutrient requirements for the different rainbowfish species.

Unfortunately, most hobbyists do not research the fish they keep, and consequently, do not provide an appropriate diet for the species they are trying to maintain.



# Rainbowfishes Species Section



Photo: Hristo Hristov

# **Rainbowfish Family**

#### Melanotaeniidae

- Cairnsichthys
  - rhombosomoides (Nichols & Raven, 1928)
- 0 Chilatherina
  - alleni Price, 1997
  - axelrodi Allen, 1980
  - bleheri Allen, 1985
  - bulolo (Whitley, 1938)
  - campsi (Whitley, 1956)
  - crassispinosa (Weber, 1913)
  - fasciata (Weber, 1913)
  - *lorentzi* (Weber, 1908)
  - pricei Allen & Renyaan, 1996
  - sentaniensis (Weber, 1908)
- 0 Glossolepis
  - dorityi Allen, 2001
  - *incisus* Weber, 1908
  - kabia (Herre, 1935)
  - *leggetti* Allen & Renyaan, 1998
  - maculosus Allen, 1981
  - multisquamata (Weber & de Beaufort, 1922)
  - pseudoincisus Allen & Cross, 1980
  - ramuensis Allen, 1985
  - wanamensis Allen & Kailola, 1979
- 0 Iriatherina
  - werneri Meinken, 1974
- 0 Melanotaenia
  - affinis (Weber, 1908)
  - *ajamaruensis* Allen & Cross, 1980
  - ammeri Allen, Unmack & Hadiaty, 2008
  - angfa Allen, 1990
  - arfakensis Allen, 1990
  - batanta Allen & Renyaan, 1998
  - boesemani Allen & Cross, 1980
  - *catherinae* (de Beaufort, 1910)
  - caerulea Allen, 1996
  - corona Allen, 1982
  - duboulayi (Castelnau, 1878)
  - eachamensis Allen & Cross, 1982
  - *exquisita* Allen, 1978
  - *fluviatilis* (Castelnau, 1878)
  - fredericki (Fowler, 1939)
  - *goldiei* (Macleay, 1883)
  - gracilis Allen, 1978
  - herbertaxelrodi Allen, 1980
  - irianjaya Allen, 1985
  - iris Allen, 1987
  - japenensis Allen & Cross, 1980
  - kamaka Allen & Renyaan, 1996
  - kokasensis Allen, Unmack & Hadiaty, 2008
  - lacustris Munro, 1964
  - lakamora Allen & Renyaan, 1996
  - maccullochi Ogilby, 1915
  - maylandi Allen, 1982
  - misoolensis Allen, 1982

- monticola Allen, 1980
- mubiensis Allen, 1996
- *nigrans* (Richardson, 1843)
- ogilbyi Weber, 1910
- oktediensis Allen & Cross, 1980
- papuae Allen, 1981
- parkinsoni Allen, 1980
- parva Allen, 1990
- pierucciae Allen & Renyaan, 1996
- pimaensis Allen, 1980
- praecox (Weber & de Beaufort, 1922)
- pygmaea Allen, 1978
- rubripinnis Allen & Renyaan, 1998
- sexlineata (Munro, 1964)
- splendida australis (Castelnau, 1875)
- splendida inornata (Castelnau, 1875)
- splendida rubrostriata (Ramsay & Ogilby, 1886)
- *splendida splendida* (Peters, 1866)
- splendida tatei (Zietz, 1896)
- sylvatica Allen, 1997
- synergos Allen & Unmack, 2008
- trifasciata (Rendahl, 1922)
- utcheensis McGuigan, 2001
- vanheurni (Weber & de Beaufort, 1922)
- 0 Pelangia
  - mbutaensis Allen, 1998
- Rhadinocentrus
  - ornatus Regan, 1914

#### Pseudomugilidae

0 Kiunga

.

• Scaturiginichthys

- ballochi Allen, 1983
- bleheri Allen, 2004
- 0 Pseudomugil
  - *connieae* (Allen, 1981)
  - cyanodorsalis Allen & Sarti, 1983
  - furcatus Nichols, 1955
  - gertrudae Weber, 1911
  - inconspicuus Roberts, 1978
  - *ivantsoffi* Allen & Renyaan, 1999
  - majusculus Ivantsoff & Allen, 1984
  - mellis Allen & Ivantsoff, 1982
  - novaeguineae Weber, 1908

signifer Kner, 1866

tenellus Taylor, 1964

- paludicola Allen & Moore, 1981
- paskai Allen & Ivantsoff, 1986

reticulatus Allen & Ivantsoff, 1986

pellucidus Allen, Ivantsoff & Renyaan, 1998

vermeilipinnis Ivantsoff, Unmack, Saeed &

Crowley, 1991

"The beginning of wisdom, as the Chinese say, is calling things by their right names."

When a fishkeeper sees a new fish, the first question most frequently asked is "what is the name of that fish" It is the nature of most aquarists to want to name every fish that he/ she maintains. In addition, biologists need to know the correct name of a fish when consulting work already carried out by others. Common names are not reliable; they are inconsistent and can be very confusing when loosely applied. Scientific names, on the other hand, enables people of all nationalities to communicate about plants and animals without confusion and allows universal recognition. To this end a complicated scientific identification system was developed to try to match the extraordinary complexity of nature. By using the scientific name of a fish you can be sure that everyone is talking about the same species. This is why name changing by taxonomists often irritates aquarists.

The terms, taxonomy and nomenclature are often confused, but have quite distinct meanings. Taxonomy is the science of classifying, describing and characterising different groups (taxa) of living organisms. Nomenclature, on the other hand, is about giving names to those different entities or groups. The system of classifying species may seem very scientific. But in fact, the system involves much human judgment. It is a system in constant flux as new knowledge is discovered. Perhaps the most modern and significant means of differentiating species is through the increase in our knowledge of genetics. It is now possible to sequence and compare the genes in different species.

#### Scientific Names

Scientific names of plants, animals, etc., follow internationally agreed rules, which are published as their respective "Codes of Nomenclature". These rules are largely the same for the different groups of organisms, but there are some differences. A scientific name is not accepted until it appears in print with a full description of the species.

Scientific names are essentially 'binomials' consisting of the name of a genus followed by the name of the species (which for plants is called the specific 'epithet'). The complete species name is comprised of the generic (genus) name and the specific (species) name. The genus is named first and is capitalised followed by the species, which is not. Both the genus and species are often italicised. It is customary to add the name of the author of the species and the date of publication, for example, *Melanotaenia angfa* Allen, 1990. Brackets around the author's name, for example, *Melanotaenia nigrans* (Richardson, 1843) indicates that, although the original description is accepted, the generic name is no longer valid and has been changed since the species was first described. The current system owes its origin to the father of modern classification or taxonomy; a Swede named Carl von Linné (1707-1778). He is better known by the Latinised name that he adopted, Carolus Linnaeus. Linnaeus devised a naming system for species that is still in use today. Linnaeus introduced the hierarchy from broadest to most specific which today with some amendment consists of a kingdom, phylum or division, class, order, family, genus, and species. The only taxonomic unit taxon (plural taxa) that actually exists in nature is the species. The species is the lowest taxonomic category and, indeed, is the only 'true' biological one; that is to say, the species is recognised as the living thing upon which natural selection operates. Species that are very similar are placed in the same genus. Similar genera are then placed in the same family and families are grouped into orders and classes. Above this the classification is subphylum and phylum (plural phyla) for animals, and division for plants. Species is either singular or plural; specific is the adjectival form. Genus is singular, genera is plural, and generic the adjectival form.

Rainbowfish taxonomy example: Phylum - Chordata Class - Osteichthyes Order - Atheriniformes Family - Melanotaeniidae Genus - Melanotaenia Species - trifasciata

The convention is that scientific names are written in italics with an initial upper-case letter for the genus and all lower case letters for the species name. The rank is not italicised. Species names are essentially adjectival in nature and thus must agree with the gender of the generic name to which they are attached. This is reflected in the endings of the names. When a species is transferred from one genus to another, the ending of the species name may also have to be altered to agree with the new genus name. For example, *Melanotaenia sexlineata* were initially described as *Nematocentris sexlineatus*. In a later review of the rainbowfish group (Allen, 1980) the name was changed to *Melanotaenia sexlineata*.

A genus name may be used on its own. Species names, however, cannot, and must always follow a genus name or its initial. A genus name should be spelt out in full the first time it is used and then may be abbreviated to an initial letter and full stop when it is unambiguous to do so e.g., *Melanotaenia trifasciata* (may be abbreviated to *M. trifasciata*).

A third level or rank can be applied to further delineate taxa into subspecies, varieties, etc. In animals only one level or rank is formally recognised – that of subspecies e.g., *Melanotaenia splendida* subsp. *splendida* and is often written without indication of rank as a "trinomial" e.g., *Melanotaenia splendida splendida*.



In plants, there are several levels below species that may be used. These infraspecific ranks are *subspecies*, *variety*, *subvariety*, *forma* and *subforma*. The last three are seldom used. In spite of there being a hierarchy, any taxon can be characterised by just using the trinomial (genus, species and infraspecies) with indication of the rank. Names must be unique within a species (that is, one cannot have a subspecies and variety in the same species with the same name but with different circumscriptions). With plants the rank must always be cited – usually as an abbreviation - and is not italicised.

#### Eucalyptus globulus subsp. bicostata Eucalyptus globulus var. compacta

Occasionally the hierarchy is included, but this is unnecessary to unambiguously define the taxon.

Leucochrysum albicans subsp. albicans var. tricolor (= Leucochrysum albicans var. tricolor).

The authors of a species name may be included, but more often than not, their inclusion can lead to error as they are seldom thoroughly checked before inclusion. They are only really necessary where the same name may have inadvertently been given to two different taxa (homonyms) within the same genus. The inclusion of the author's name following the species (or infraspecies) name can then distinguish the two names. With animal names the author name is always followed by a year; with plants, the author name or abbreviation is given alone.

Animals (Zoology):

Emydura signata Ahl, 1932

*Emydura australis* (Gray, 1841) - (the bracket indicates that Gray ascribed the species to a different genus)

#### *Emydura* (Bonaparte, 1836)

The generic name *Emydura* was derived from the Greek emys (freshwater turtle) and the Greek oura (tail), Latinised to ura. Its grammatical gender is feminine. The type species was *Emys* macquaria (Cuvier, 1829) by monotypy. The genus name *Emydura* was erected by Charles Lucien Jules Laurent Bonaparte (1803-1857) in 1836.

Plants (Botany):

Melaleuca nervosa (Lindley) Cheel

Synonym: *Callistemon nervosus* Lindley - (Lindley originally described it as a *Callistemon*; Cheel later transferred it to the genus *Melaleuca*).

#### Authorship in Scientific Names

Sometimes you will see a name or abbreviation of a name after a scientific name and even a year as well. A complete reference to a species includes not only the binomial name, but also the author(s) that described the species and gave it a name. While the scientific name is italicised, the author citation is not. This addition of authorship is usually only done once in a particular article or citation. Conventions in author citation differ somewhat between botany and zoology, and are governed by the International Code of Botanical Nomenclature and International Code of Zoological Nomenclature respectively. Botanical author citation

The name or names of plant authors are abbreviated to a standardised index of author names published by the Royal Botanic Gardens, Kew; the date of publication is not cited in brief citations.

Example: *Aponogeton elongatus* F.Muell. ex Benth. subsp. *Elongatus* - The abbreviation "F. Muell." refers to Ferdinand Jacob Heinrich von Mueller (1825-1896) and "Benth." refers to George Bentham (1800-1884).

Note: If the author of a validly published taxon ascribes it to another person, the author citation will include the ascribed author followed by the term "ex" and then the publishing author.

#### Zoological author citation

The name or names of animal authors have their surname given in full, not abbreviated, while first names are not included, or if two authors share the same surname, are given as initials. The date of first publication is also cited, with a comma between the author and date.

#### **Common Names**

Often what are called 'common' names are in reality colloquial names and may have just been coined from a translation from the scientific name. Most common names are not governed by rules. For some groups, such as fishes, guidelines and recommended common names are available at: http://www.marine.csiro.au/caab/namelist.htm Standard Names of Australian Fishes.

The Australian Environment Department, guidelines for use of common names have been developed to support consistency. These include beginning each word in the name with an initial capital i.e., Sunset Frog. With generalised or grouped names a hyphen is recommended. The word following the hyphen is generally not capitalised, except for birds where the word following the hyphen is capitalised if it is a member of a larger group.

#### Fishes – an exception to the rule:

Specific names are in lower case barramundi, golden snapper, as are groups of fish, jewfish and salmon, except at the beginning of a sentence. Australian bass is the exception, capital A is used as it relates to a country. Common names that consist of a place name, or a person's name are capitalised. However, generally it is common practice that most common names be capitalised!

The correct biological term when referring to more than a few individuals of one fish species or for two or more species, is fishes. However in common use fish and fishes are often used interchangeably.



#### **Unpublished Names**

Unpublished names can take many forms. In the interests of conservation management, threatened species often have to be listed long before they have a formal name. Sometimes, these are listed as manuscript names (e.g. *Genoplesium vernalis* D.L. Jones ms.) if they are about to be published. However, in some cases these manuscripts names remain unpublished for years or even decades.

In the 1980s in Australia, botanists agreed on a formula for use with unpublished names to avoid the confusion that was arising through the use of such things as *"Verticordia* sp.1", *"Verticordia* sp.2" etc. There was no guarantee that what was called "sp.1" in one institution was identical to "sp.1" in a second.

The agreed formula is in the form of: "Genus sp. <colloquial name or description> (<Voucher>): *Prostanthera* sp. Somersbey (B.J.Conn 4024)

*Elseya* sp. nov. (AMS – R140984)

Some zoologists use a similar convention, but it is not done so universally.

Where animal populations need to be identified, they are often done by inclusion of a form or population identifier in brackets following the species name e.g., *Melanotaenia duboulayi* (Mary River form) or *Melanotaenia duboulayi* (Mary River).

#### Synonyms

Synonyms are names that have previously been applied to a taxon, but are now generally superseded. They may be names originally ascribed to a different genus and have the same specific epithet or name - these are based on the same voucher or type specimen and are known as *nomenclatural synonyms*. *Melanotaenia duboulayi* synonym: *Atherinichthys duboulayi* 

Alternatively, they may have once been described as a separate taxon, but later studies have determined them to be the same taxon - these generally have different type specimens and are known as *taxonomic synonyms*. *Craterocephalus capreoli* synonym: *Craterocephalus anticanus* 

#### **Pronunciation of Species Name**

In general, scientific names are derived from Latin or Greek (both defunct languages and therefore not undergoing continual change), and their pronunciation should follow strict Latin or Greek pronunciation rules. General usage has, however, often anglicised or corrupted true grammatical pronunciation and more and more, names are being derived from languages other than Greek or Latin. Scientific names are often based on the appearance of the species or include the name of people or places associated with the discovery. How they are pronounced really doesn't matter providing they are understood by all concerned.

#### Abbreviations and Contractions

Sometimes you will see the generic name of an organism with sp. or spp. after it, for example:

*Melanotaenia* sp. means a species of the genus *Melanotaenia*; *Melanotaenia* spp. means species of the genus *Melanotaenia* (i. e., more than one).

There are a number of important abbreviations and contractions used in nomenclature:

cf. - confer (compare with ...) cv. - cultivar f. - form/ forma fam. - family gen. nov. (genus novus) - a newly described genus ined. - ineditus (unpublished) ms. - manuscript (unpublished manuscript name - generally follows an author name) p. p. - pro parte (in part) sect. - section/sectio s. lat. - sensu lato (in the broad sense) s. str. - sensu stricto (in the narrow or strict sense) sp. - species sp. aff. - species with affinity to ..., or close to ... (NB. 'aff. sp.' should not be used) sp. nov. (species novus) - a newly described species (NB. 'nov. sp.' should not be used) spp. - species (plural) ssp. - (not preferred - see subsp.) subg. - subgenus subsp. - subspecies subspp. - subspecies (plural) syn. - synonym var. - variety

Abbreviations of italicised words may be italicised, however they are often better not italicised in order to provide a contrast with the (italicised) genus and species names.

Examples: Eucalyptus smithii s. lat. Eleocharis sp. aff. acuta Zenarchopterus cf. buffonis





#### **Cairnsichthys rhombosomoides**

(Nichols & Raven, 1928) Cairns Rainbowfish

#### **Species Summary**

*Cairnsichthys rhombosomoides* were originally collected by Henry Raven in October, 1921 from Babinda Creek, a tributary of the Russell River in north Queensland. They were scientifically described in the American Museum Novitates Nr. 296, in 1928 by John T. Nichols and Henry C. Raven and placed in the genus *Rhadinocentrus*. Gerald Allen's revision of the family Melanotaeniidae in 1980 placed them in the monotypic genus *Cairnsichthys* in recognition of its distinctiveness.

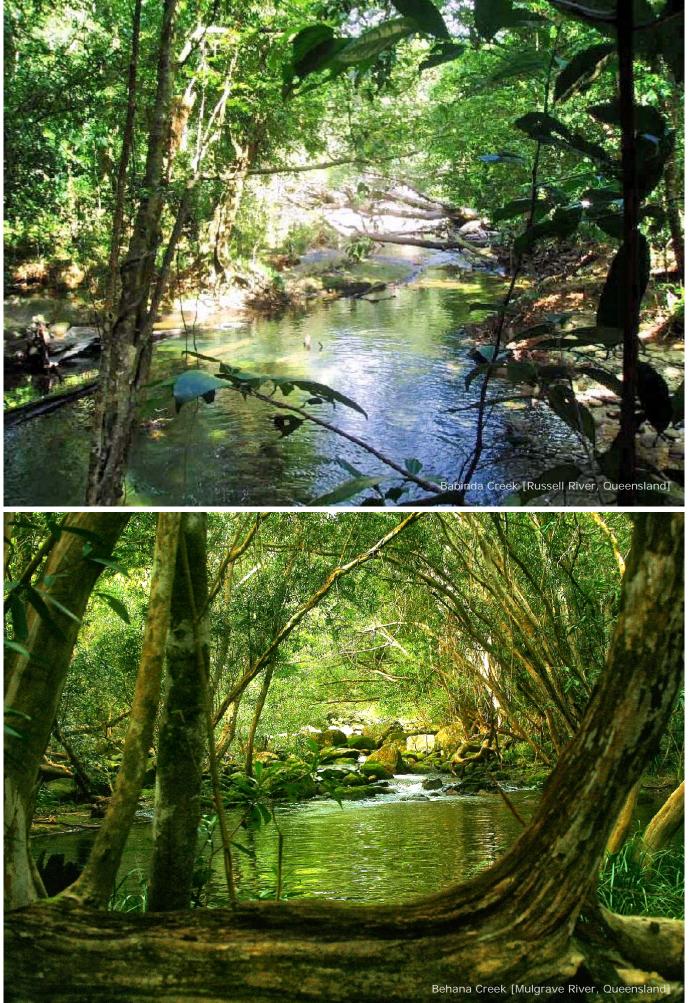
Body colouration is yellow-brown on the dorsal half of body and silvery-white below with a continuous thin dark mid-lateral band. A large silvery or yellowish spot can be seen on the operculum. Males have a yellow-orange coloured outer margin on the dorsal and anal fins. The caudal fin has a similarly coloured fan. They may reach a maximum body size of 10 cm, but are usually around 7 cm. Males can be distinguished from females by their elongated dorsal and anal fins and brighter colours - females have shorter and more rounded fins. Males are also larger and deeper bodied than females. *Cairnsichthys rhombosomoides* is believed to be a very old isolated species, whose current distribution is the remnant of a previously larger distribution range that has been reduced due to environmental changes. It is possible that they were the original rainbowfishes in rivers of northern Queensland.

#### **Distribution & Habitat**

*Cairnsichthys rhombosomoides* is a tropical species inhabiting mainly the river systems that arise in the Francis and Bellenden Ker ranges about 45 km south of Cairns in northern Queensland. They have been collected from a number of river systems draining eastward, such as the Hull, Johnstone, Moresby, Mulgrave, Russell, and Tully Rivers; Liverpool and Maria Creeks. They also occur in some small isolated coastal streams around the Innisfail region. They have a very limited distribution range and as such, have a "restricted" conservation status listing which means "a species which is not presently in danger but which occurs in restricted areas and/or are uncommon".

*Cairnsichthys rhombosomoides* can be found in both lowland and headwater tributary streams in water depth ranging from a few centimetres to about three metres. They are however, more commonly found in water between 30 and 50 cm deep.







Upland streams generally have higher water flow over a substrate composed mainly of large rocks and bedrock with sand and fine gravel with good riparian cover (remnant rainforest) and minimal aquatic plants. Small lowland streams usually have mud, sand and fine gravel substrates with abundant leaf litter. The temperature range recorded in their natural habitat is 15–29° Celsius. They are often found in company with *Melanotaenia splendida*, *M. maccullochi* and *M. utcheensis*. The *p*H in their natural habitat has been reported from 4.5–8.5 (lowland 4.5–6.8, upland 7.5–8.5). Conductivity from almost zero to 91 mS. They are generally found in small schools swimming above or among aquatic plants, woody debris and leaf litter.

#### Remarks

Although an attractive species if kept under suitable conditions, they are rarely seen in the general aquarium hobby and are mainly kept by a few aquarists who are principally interested in Australian native fishes.

#### Breeding

Very little is known about the natural life history and ecology of *Cairnsichthys rhombosomoides* in their natural environment. In their natural environment spawning fish have been observed from April through to December with a peak in August to October. Like all members of the rainbowfish family, *Cairnsichthys rhombosomoides* are egg-scatterers and generally spawn amongst aquatic plants and leaf litter, with a small number of eggs being deposited at a time. In captivity, I have on a regular basis observed this species spawning in the gravel substrate.

Fish commence spawning in their first year and mature females produce between 40 and 200 eggs. The number of eggs shed by a single female is directly related to the size of the female. Eggs adhere to water plants and hatching occurs after 5–9 days depending on temperature. Egg size is around  $1.139 \pm 0.021$  mm with larvae hatching at about 3.46-5.46 mm. Larval development is complete at around 14-15 mm body length. I found this species seems to ignore free-swimming larvae in their aquarium. Although the larvae always stay close to some form of cover and generally avoid open areas of the aquarium.







### Chilatherina alleni

Price, 1997 Allen's Rainbowfish

#### **Species Summary**

*Chilatherina alleni* is most similar to *Chilatherina fasciata* of the northern New Guinea mainland and *Chilatherina pricei* of Yapen Island in Geelvink Bay. It differs greatly from these in colouration. *C. alleni* having a dark mid-lateral stripe and reddish fins in males. It further differs from *C. fasciata* in having a shorter blunter snout with the maxillary reaching to about the level of the anterior edge of the eye (falling well short in *C. fasciata*). It further differs from *C. pricei* in the modal counts of soft anal rays (usually 19–24 in *C. alleni* vs. 24–27 in *C. pricei*), and in the number of cheek scales (16-18 in *C. alleni* vs. 19-23 *C. pricei*). *C. alleni* may reach a maximum size of 10 cm, but are usually less than 7 cm. Males are usually much larger and deeper bodied than females.

*Chilatherina alleni* have the upper half of the body brownish anteriorly becoming turquoise posteriorly, each horizontal scale row is separated by narrow brown or dull orange stripe. Midlateral band dark blue, occupying about two horizontal scales rows, from upper rear corner of eye to base of caudal fin, interrupted on its lower edge by about five, white indentations in area just behind pectoral fin. Lower half of body mainly dark silver, except for intense white stripe with lower margin of dull orange occupying scale row immediately below mid-lateral band. 4–5 diffused charcoal coloured bars on lower side between level of pelvic fin base and fin origin.

The first dorsal fin is light blue; second dorsal and anal fins translucent bluish becoming reddish on margins. Caudal fin translucent reddish. Pectoral and pelvic fins mainly translucent. Females slightly less intense with median fins mainly translucent to bluish without any red. Besides the colour differences noted above, males are typically deeper bodied than females. In addition, the second dorsal and anal fins short and blunt in females and somewhat elongated and pointed in males.

There is a wide variation in colouration of this species depending on habitat. One colour variation has a reddish back, red fins and bright yellow stripe in the middle of the body.

#### **Distribution & Habitat**

*Chilatherina alleni* were originally collected in 1994 from the Aiborei (Aboge) River, situated in the Derewo River Basin, southeast of Cenderawasih Bay in northern West Papua. They were again collected in 1998 during a Conservation International survey from tributaries of the Wapoga River. These specimens have a slightly different colour pattern.

#### Remarks

Named "*alleni*" in honour of Gerald R. Allen, in recognition of his outstanding contribution to ichthyology and his deep commitment to the study and preservation of the aquatic fauna of New Guinea. Specimens originating from the type location near Siriwo Village were introduced into the European hobby in 2009.











#### Chilatherina axelrodi Allen, 1980

Axelrod's Rainbowfish

#### Species Summary

Chilatherina axelrodi males have a body colour of bluishgrey above a blackish midlateral line and silver-grey below. The midlateral band is broken into a series of large blotches and there are several dark vertical bars on the lower side of the body. The dorsal, anal and pelvis fins are yellowish, other fins translucent. Females are an overall silvery colour and rather plain compared to the males. Males are more brightly coloured, larger, and much deeper bodied than females. Males may reach a maximum size of 10 cm, with females usually less than 8 cm SL. Spawning usually occurs from October to January, with females producing between 50 and 150 eggs, spawning over a period of several days. Eggs adhere to fine-leaved plants or among the roots of floating vegetation which hatch around 7-10 days.

#### **Distribution & Habitat**

Chilatherina axelrodi were first collected in 1979 from Yungkiri creek, a tributary of the Pual River (formerly Nemayer or Neumayer River), in the Bewani Mountains of Papua New Guinea. This location is about 40 kilometres inland from the north coast town of Vanimo, which is close to the West Papuan border. Vanimo is a relatively recent township; it was established as a patrol post and then abandoned and reoccupied several times during the Australian administration. Only after the Sepik district was divided into



east and west in 1967, Vanimo became the capital of what was then the West Sepik District and today is the Sandaun Province. Sandaun Province is the north-westernmost province of Papua New Guinea. It covers an area of 36,300 km<sup>2</sup>.

C. axelrodi were found around sub-surface vegetation, submerged logs and branches in a small, narrow slow flowing rainforest stream. The water at the collection site was slightly turbid and a temperature of 26°C and pH 7.8 were recorded. Other rainbowfishes found in the stream included Chilatherina crassispinosa and Melanotaenia affinis. Chilatherina fasciata and Chilatherina lorentzi have also been collected from the Paul River.

#### Remarks

A number of live specimens were collected by Gerald Allen, Brian Parkinson and Peter Neusinger in September 1979, but unfortunately they all died soon after arriving in Australia. However, Gerald Allen returned in 1982 and together with Heiko Bleher they collected more live specimens, which were later bred and distributed in the hobby. In 1983 further live specimens were collected by Barry Crockford and returned to Australia. Although a rather attractive species, it has never achieved much popularity in the hobby, and could be considered rare. The species was named in honour of Herbert R. Axelrod who provided funding for the first collecting expedition.





#### **Chilatherina bleheri** Allen, 1985 Bleher's Rainbowfish

#### **Species Summary**

Chilatherina bleheri is a very attractive rainbowfish. They generally have a silvery or greenish body colour on the upper back fading posteriorly to pastel shades of yellow to red. The scales on the front half of body, particularly on the dorsal region, have broad yellow-green margins; the first dorsal fin charcoal grey; second dorsal fin grey suffused with red; caudal and anal fins red; pelvic fins reddish anteriorly with remainder white or translucent; pectoral fins translucent. The lower side of the body is white with a series of faint vertical dark markings. Females lack the vivid red hues and are mainly silvery or bluish grading to grev or greenish-brown. Males display a brilliant yellow-orange stripe on the middle of the forehead during spawning. Males are larger than females with older males developing a very deep body. Males may reach a maximum size of 12 cm, but females are usually less than 10 cm. C. bleheri are essentially a carnivore, feeding on a variety of terrestrial and aquatic insects, insect larvae, and small aquatic crustaceans. Aquatic algae and fallen plant pollens are also ingested.

#### **Distribution & Habitat**

*Chilatherina bleheri* have been collected from the vegetated shoreline and feeder streams of Lake Holmes (Danau Bira) situated in the Mamberamo region of West Papua. Lake Holmes is a complex of three interconnected lakes lying at an altitude of about 430 metres above sea level and set in the foothills of the van Wees Mountains, approximately 290 kilometres west of Jayapura, the capital city of West Papua. The lakes lie within a radius of 6-7 kilometres with the main lake having a length of approximately 4.5 kilometres and maximum width of about 2 kilometres. The lakes are drained by a small stream, which flows into the Mamberamo River at a point approximately 15 kilometres directly to the north. The lake and surrounding creeks are inhabited by 11 fish species, including one other rainbowfish, *Melanotaenia maylandi*.

#### Remarks

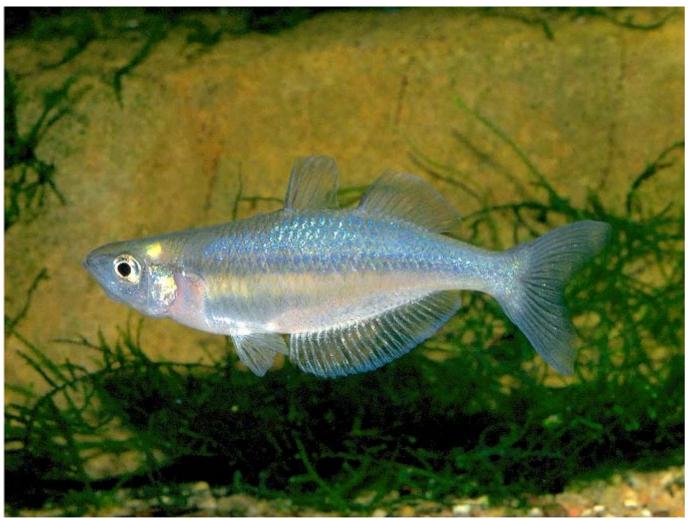
Live specimens of this species were initially collected by Gerald Allen and Heiko Bleher in 1982. When first discovered it was thought that they were just another colour variety of *Chilatherina fasciata*. However, in 1985 they were described by Gerald R. Allen as a new species and named in honour of Heiko Bleher, a well-known fish collector. The two species differ primarily with respect to size of scales and male colouration. *Chilatherina bleheri* have smaller scales.











**Chilatherina bulolo** (Whitley, 1938) Bulolo Rainbowfish

*Centratherina bulolo* Whitley, 1938 *Chilatherina bulolo* Allen, 1983

#### Species Summary

*Chilatherina bulolo* have an overall silvery body colour with a black streak on the upper and lower edges of the caudal fin. It is very similar to *Chilatherina crassispinosa*, but has a wider head, blunter head, larger eye, and shorter first dorsal fin. It also tends to be more slender, but depth increases with age. Males are usually much larger and deeper bodied than females. Males may reach a maximum size of 10 cm, but females are usually less than 8 cm. They are essentially a carnivore, feeding on a variety of terrestrial and aquatic insects, insect larvae, and small aquatic crustaceans. Algae, plant pollens, and seeds are also ingested. Stomach contents of wild caught specimens indicated a diet of small insects, particularly ants, and algae that is grazed from rocks.

#### **Distribution & Habitat**

Chilatherina bulolo is so far only known from scattered localities in the Markham (Erap Snake and Bulolo Rivers),

Ramu (Whege River) and Sepik River systems of north-eastern Papua New Guinea. They have been collected from the Erap, Snake, Bulolo, and Whege rivers. They are found mainly in mountain streams with rapid flow and coarse gravel bottoms, in water temperatures between 24-28° Celsius and pH 7.5-8.5.

#### Remarks

Live specimens were collected in 1978 and transported to Australia, but failed to become established in the hobby.







#### **Chilatherina campsi** (Whitley, 1956) Highland Rainbowfish

#### Anisocentrus campsi Whitley, 1956 Centratherina tenuis Nichols, 1956

#### **Species Summary**

*Chilatherina campsi* is a small stream dwelling species and was first collected in 1954. Gilbert P. Whitley, an ichthyologist at the Australian Museum, described them as *Anisocentrus campsi* in 1956. They were subsequently reassigned to the *Chilatherina* genus where they remain today.

*Chilatherina campsi* have a body colour of pale bluish-white with a silvery sheen; pale yellow to whitish longitudinal stripes frequently bordering scale rows and a broad blue mid-lateral stripe (most prominent on posterior part of body); fins white, sometimes with yellow suffusion; maximum size around 8–9 cm SL. Males are deeper bodied than females and their overall pattern is more intense, particularly the mid-lateral stripe. In addition the vertical fins of females are mainly translucent in contrast to the whitish fins of males. There were two different colour varieties available in the aquarium hobby, one with red fins, and one with blue fins. However, the origin of these different varieties is not known to me.

#### **Distribution & Habitat**

*Chilatherina campsi* occurs in the central highlands of Papua New Guinea, hence its common name 'Highland Rainbowfish'. The genus *Chilatherina* is usually only found in the northern areas of New Guinea but *C. campsi* have been found in southern stream habitats. It is the only member of the genus thus far found on both sides of the

Central Dividing Range, which has acted as a most effective geographical barrier to the spread of most rainbowfish species northerly or southerly.

This species was first collected in 1954 from a tributary of the Jimmi River, situated approximately 420 km from the mouth of the Sepik River via the Yuat River. They have been collected from dispersed localities in the Markham (Oomsis Creek, Wampit River), Ramu, Sepik, and Purari Rivers (Lima River; Pima River; Wahgi River). Most collection sites have been in the northern drainage division, but they have also been collected from highland tributaries of the Purari River that flows southwards into the Gulf of Papua, and from the Oomsis River near Lae.

Within their distribution range *C. campsi* are generally found in mountainous or foothill streams. They are most abundant in the smaller flowing tributaries, shallow bodies of water shaded by rainforest trees where they find shelter among aquatic plants, roots, and fallen branches. Although situated in rainforest habitat the streams are relatively open and exposed to sunlight, which is typical of the type of habitat where *Chilatherina* normally occurs. Depending on the precise location, the water is generally soft, slightly turbid with a temperature range of 21-26° Celsius and *pH* 7.6-7.8. Other rainbowfishes found co-habiting with *Chilatherina campsi* include *Melanotaenia affinis*, *Melanotaenia pimaensis*, and *Glossolepis maculosus*.

#### Remarks

The first live specimens to enter the aquarium hobby were collected by Gerald Allen and Brian Parkinson from the Wahgi River (Purari River) near Mt Hagan in 1979. Then in the early 1980's, additional specimens was collected by Barry Crockford from the Oomsis River near Lae and a small stream on the Highland Highway 105 km northwest of





Lae, both foothill tributaries of the Markham River. Heiko Bleher also collected live specimens from a tributary of the middle Ramu River in 1988. These collections formed the founding stock of current aquarium populations. *C. campsi* have never been widely available and are seldom seen in the hobby these days. Only a handful of enthusiasts are still maintaining them.







**Chilatherina crassispinosa** (Weber, 1913) Silver Rainbowfish

*Rhombatractus crassispinosus* Weber, 1913 *Centratherina crassispinosa* Regan, 1914 *Chilatherina crassispinosa* Regan, 1914

#### **Species Summary**

*Chilatherina crassispinosa* has an overall silvery body, shading to bluish dorsally and white ventrally. Fins translucent, except anal and pelvis fins and base of second dorsal fin often yellowish in males; dorsal and ventral edges of caudal fin with narrow black margin. Males have narrow orange stripes on the sides; one between each scale row and are generally deeper bodied than females. Males have more intense colouration, particularly with regards to the orange stripes and the yellow colour of the vertical fins. *C. crassispinosa* are similar to *Chilatherina bulolo*, but have a more pointed head, taller first dorsal fin, and a narrower space on top of the head between the eyes. Maximum size of males to about 10 cm SL, females to about 8 cm.

#### **Distribution & Habitat**

Known from foothill tributaries of the Torricelli Range on the northern side of the Sepik River Basin. They also occur in the upper Ramu system and streams in the Bewani Mountains flowing into the Neumayer River, both in Papua New Guinea, north of the Sepik. The range extends into northern West Papua where it is known from several coastal streams just to the west of Jayapura and from a few scattered locations in the Mamberamo system. This species was initially collected from the Tawarin River, West Papua during 1903. It is widely distributed in northern New Guinea. It is found in the following rivers system: Begowre, Buarin, Gogol, Mamberamo, Markham, Pual, Ramu, Sepik and Sermowai. It also occurs in a number of smaller independent drainages along the northern coast.

*Chilatherina crassispinosa* are found in slow flowing, clear water, streams and quiet pools, in water temperatures between  $24-28^{\circ}$  Celsius and *p*H 7.5-8.5. These streams are usually situated in hilly (rainforest) terrain. The fish congregate in exposed sections, which receive full sunlit for most of the day. Other rainbowfishes sometimes found together with this species include *Chilatherina lorentzi*, *Chilatherina fasciata*, and *Melanotaenia affinis*.

#### Remarks

This species was previously recorded from the Markham system of Papua New Guinea by Allen and Cross (1982), but in a subsequent paper by Allen (1983) the Markham population was shown to be a distinct species, *Chilatherina bulolo*. Live specimens were collected in 1980 and 1983, and transported to Australia. However, they were never popular or widely available and their status in the hobby today is uncertain.





## Chilatherina fasciata

(Weber, 1913) Barred Rainbowfish

*Rhombatractus fasciatus* Weber, 1913 *Chilatherina fasciata* Regan, 1914

#### **Species Summary**

As with many rainbowfishes, *Chilatherina fasciata* often display a great variation in body colour and markings depending on location. Throughout New Guinea there are effective natural barriers that isolated various populations, thus contributing to the number of different colour variations.

Generally, they have a body colour of brown to bluish-green on the upper half, white to yellowish on the lower half with a diffuse dark mid-lateral stripe. Scales of this region often bordered with pale yellow. Males usually have several diffuse blackish bars on the lower sides, above the front half of the anal-fin base; fins dusky grey to yellowish.

Males are generally deeper bodied than females, this feature becoming more obvious with increased growth. In addition, the posterior profile of the dorsal and anal fins is more pointed and elongated in males. In contrast to males, which have longer posterior dorsal rays, females have the longest rays at the anterior part of the fin. Finally, mature males are more colourful than females often exhibiting reddish or yellowish dorsal and anal fins. While spawning the males colour becomes very intense and the top of the head radiates a brilliant bronze to vermilion hue. Females are basically silver to olive overall with clear fins.

Perhaps the most distinguishing feature of *Chilatherina fasciata* is their deep, laterally compressed body that increases with age, particularly in males. Males may reach a maximum size of 12 cm, but females are usually less than 10 cm.

#### **Distribution & Habitat**

Type-locality: Mouth of the Sermowai River, New Guinea. *Chilatherina fasciata* is probably one of the most abundant and widely distributed rainbowfishes in northern New Guinea. They have been found in tributaries of the Markham, Ramu, Sepik, Neumeyer, Grimé and Mamberamo River systems of northern New Guinea. They have also been collected from the following lakes: Sentani, Wanam and Nenggwambu (Kali Biru).

*Chilatherina fasciata* have been collected mainly in clear, slow-flowing rainforest streams, generally inhabiting deeper pools that are exposed to sunlight for most of the day. These streams usually have a substrate consisting mainly of gravel or sand and littered with leaves and other debris. The natural *p*H and temperature ranges have been reported as 6.2–8.1 and 27–32° Celsius.

Diet includes filamentous algae, small crustaceans, terrestrial insects (particularly ants and tiny beetles), and aquatic insect larvae.











#### Remarks

Pieter Nicolaas van Kampen first collected *Chilatherina fasciata* in June 1910 in a stream near Njao, West Papua. Live specimens were collected for the aquarium hobby by Barry Crockford and Gerald Allen during several trips to Papua New Guinea in the late 1970's and early 1980's. Around 1983, live specimens were also collected from the Jafuri River, an outlet of Lake Sentani which flows into the Pacific Ocean near the Papua New Guinea border. In 1991, another live collection was made by Gilbert Maebe from the Jafuri River and taken back to Europe.

In 1981, Barry Crockford and Neil Travis collected specimens (*Clearwater Creek*) from a tributary of the Markham River, approximately 60 km east of Lae, PNG. The creek had a water depth of about 50 cm. The water conditions reported were pH 7.5, hardness 90 ppm and temperature 24°C. The creek was slow-flowing with a very muddy substrate and vegetation lining the shore.

Males of this variety have an upper body colour of brilliant bronze while the lower half shows a silvery-white colouration with a diffuse dark mid-lateral stripe. The lower half of the body has several diffused brownish-black vertical bars. These sometimes change to the same colour as the mid-lateral stripe. During spawning, the lower half of the males' body deepens to a golden-yellow colour (as shown in the accompanying photo) and the dorsal region deepens to a burnished copper colour, while the darker body markings intensify to near black. The first two rays of the first dorsal and first ray of the second dorsal change from their normal orange colour to a brilliant vermilion. The nape region from the dorsal fin to the tip of the nose becomes a flashing reddish-bronze colour. The colour in the lower part of the iris develops a beautiful blue-violet opalescence ringed by orange-gold. This rich gold is also brilliant on the central area of the gill plate. During spawning the central section of the caudal fin is much lighter than the blue of the outer lobes. The scales on the sides of the fish above the lateral line show some subtle violet hues in the mid-scale region.

The female is much more slender than the male. It is silvery in colour, with shorter and more rounded finnage. The female has a violet-blue reflective colour in the mid-lateral region. The base colour above the lateral line is a dull olive-green with dark scale outlines. A dark continuous stripe runs back from the eye into the peduncle at the lateral line. The peduncle is very slim and shows only a narrow edge of olive above and below the dark line. The lower part of the chest is a dull yellow with a strong sprinkling of black speckles from the lower jaw back to the ventrals. The fins are all a dull olive-yellow which fade to white toward the edges. The gill cover has a strong orange-copper toning rather deeper in hue than the colour of the iris. Several rather indistinct vertical bars occur on the lower sides of the female.







Heiko Bleher has collected live specimens for the aquarium trade from various locations in New Guinea. The latest colour variant was collect in 1999 from Lake Nenggwambu (Kali Biru) in West Papua. He collected only males and had to return a second time in order to collect females. This variety has a broad orange stripe from the tip of its snout to the first dorsal fin. There is a vigorously flowing outlet stream, but no apparent inlet, indicative of a subterranean connection with neighbouring lakes via the limestone substratum. Water was relatively clear and maximum depth was estimated to be at least 10–15 metres, *p*H 7.8, and conductivity 60  $\mu$ S/cm. The lake is surrounded by secondary forest and aquatic plants were abundant, but relatively few species were evident. Fishes were most strongly congregated around the outlet, where vegetation was very dense. *Glossolepis dorityi* was also collected from this location. There have also been a number of collections by individual aquarists from other various locations and these are frequently available in the hobby.







# **Chilatherina lorentzi**

(Weber, 1908) Lorentz's Rainbowfish

*Rhombatractus lorentzi* Weber, 1908 *Rhombosoma lorentzi* Regan, 1914 *Chilatherina lorentzi* Regan, 1914

# **Species Summary**

*Chilatherina lorentzi* have a basic bluish body colour grading to silvery-blue ventrally and a broad darker blue midlateral band, with clear to yellowish fins. They may attain a length of about 12 cm with a body depth of around 3–4 cm.

# **Distribution & Habitat**

Type-locality: Tawarin River, northern New Guinea. Known only from the Sermowai, Tawarin and Mamberamo Rivers on the north coast of West Papua, and from Puive Creek, a tributary of the Pual River, near Vanimo, Papua New Guinea in 1979. They were collected from slow-flowing stream that was enclosed by dense rainforest. The bottom was covered entirely with leaf litter. The fishes were confined in the deeper sections of the stream in the vicinity of waterplants and debris. Temperature 28° Celsius and *p*H 7.8.

### Remarks

This species is poorly known and live specimens so far have not been collected for the aquarium hobby.

In 2008, a number of surveys were conducted by a team from the Manokwari University in cooperation with Conservation International around the Haya Village region (02°48.951'S and 138°05.903'E) in the Mamberamo. Three species of rainbowfishes were collected during these surveys which included *Chilatherina fasciata*, *C. lorentzi* and *Glossolepis multisquamata*. *G. multisquamata* were found in the lakemarsh habitats, *C. fasciata* were found in creeks near the village, and *C. lorentzi* was found in clear rocky streams about 6 km from the village.





# **Chilatherina pricei**

Allen and Renyaan, 1996 Price's Rainbowfish

# **Species Summary**

*Chilatherina pricei* is a small stream dwelling species, growing to a length of about 7 to 10 cm. They were described by Gerald R. Allen and Samuel J. Renyaan in 1996 on the basis of 23 specimens collected in 1991 and 1995 from the Reifafeif River near Warironi Village on Yapen Island, off the central north coast of West Papua. They are similar to *C. fasciata* from the northern New Guinea mainland, but differ in colouration and have a shorter, blunter snout with the maxillary reaching the level of the eye (falling well short of eye in *C. fasciata*). It is also similar to *C. alleni* from the Derewo and Wapoga river systems of West Papua, but lacks a prominent midlateral stripe and reddish fins (males), and differs in modal counts of soft anal rays (usually 24–27 in *C. pricei* versus 19–24 in *C. alleni*), and number of cheek scales (usually 19–23 in *C. pricei* versus 16–18 in *C. alleni*).

The body colour is overall silvery-grey to bluish; scales on back sometimes with golden or yellow margins, wider on upper and lower edge of scale and forming a longitudinal yellow stripe between each scale row. Scales on caudal peduncle sometimes entirely yellow. About 5–10 dull grey to dark blue bars on lower half of side between level of pelvic fin base and middle of anal fin; some individuals have 2–6 small dark blue spots on mid-lateral scale row or on scale row immediately above, on rear half of body. Fins mainly translucent bluish, except dorsal and anal fins of mature males often golden yellow.

### **Distribution & Habitat**

The Reifafeif River habitat is typical of numerous streams on Yapen Island, which no doubt are populated by this species. Yapen Island is situated in Cenderawasih Bay (formerly Geelvink Bay) off the northern coast of West Papua. It is a narrow, elongate (approximately 170 km long and up to 25 km wide) island with a central spine of mountains which rise to a maximum elevation of 1500 metres, and was once part of the New Guinea mainland. It has an area of approximately 2424 km<sup>2</sup>. The mountain range is dissected by numerous independent drainage systems, such as the Reifafeif River. The streams rise in the steep mountains and plunge to a relatively narrow (3-5 km wide) coastal plain, which is the home of C. pricei and most other freshwater fishes on Yapen. The main channel of the Reifafeif River on the coastal plain is approximately 15-30 m wide and in non-flood periods averages about 1-2 m in depth. Water temperatures and pH in July 1995 ranged between 24.2°-26.4°C and 7.5-8.1. Progressing inland, the river quickly narrows as it approaches the mountains and is characterised by scenic rapids, waterfalls, and deep pools. C. pricei is abundant in the main channel, usually over rock or boulder bottoms. It forms midwater aggregations that feed on algae and small invertebrates, particularly insects. Although the river flows through rainforest and gardens, it is generally open to sunlight over the coastal plain. Melanotaenia japenensis is sometimes found with C. pricei, but seems to be more common in smaller, shaded tributaries, rather than the main channel.

#### Remarks

This species was named after David Price, in recognition of his keen interest in the natural history of New Guinea. He collected the type specimens of *Chilatherina pricei*.





### **Chilatherina sentaniensis** (Weber, 1908) Sentani Rainbowfish

*Rhombatractus sentaniensis* Weber, 1908 *Chilatherina sentaniensis* Regan, 1914

#### Species Summary

Perhaps the most distinguishing feature of the *Chilatherina* genus is their deep, laterally compressed body that increases with age, particularly in males, and *Chilatherina sentaniensis* is no different. Their overall body colouration is silvery-blue or greenish on the upper back fading laterally to silverly orange. They have a diffuse blue or green mid-lateral band and narrow silver or light blue stripes between each horizontal scale rows. However, colour can be variable depending on captive conditions. Males are more brightly coloured, larger, and deeper bodied than females. Males may reach a maximum size of 12 cm, but females are usually less than 10 cm.

#### **Distribution & Habitat**

*Chilatherina sentaniensis* are endemic to Lake Sentani and its tributaries streams. Lake Sentani is located some 10 kilometres west of Jayapura at the NE extremity of West Papua. It is an irregularly shaped lake with approximate dimensions of 28 km (E-W) by 19 km (N-S) and a surface area of 104 km<sup>2</sup>. Its bluegreen waters are dotted with at least 16 small islands, and it is surrounded by hillsides in the south and the Cyclops Mountains in the north, which separate the lake from the Pacific Ocean. Lake Sentani is by far the largest of the West Papuan lakes and has a catchment area of about 600 km<sup>2</sup>. About 35 small rivers flow into the lake, and there is one natural outlet in the southeastern tip, via the Jafuri and Tami rivers to the Pacific Ocean near the Papua New Guinea border. The lake is divided into three main sections with recorded depths of 7 to 52 metres. According to surveys in 1970-71, 1984 and 1987 the lake is thermally unstratified, with surface temperatures of 29–32° and pH 6.2–6.8. Rainbowfishes are generally found around the margins of the lake. Large numbers are found congregating around submerged aquatic vegetation, fallen tree branches etc.

Because of its proximity to the provincial capital Jayapura and the large population around it, Sentani is no longer the pristine lake it once was. A survey by Samuel J. Renyaan in 1993 recorded 33 species of fish, of which 13 were introduced. Surveys have shown an increase in introduced species but the impact on the total fish population has not been documented. Fish are extensively raised in ponds and cages around the perimeter of the lake and the introduction of species (particularly carp and tilapia) has been both accidental and intentional.

#### Remarks

*Chilatherina sentaniensis* was originally collected from Lake Sentani and the Sekanto River during the 1899–1900 Siboga Expedition to the Dutch East Indies (West Papua). A large collection of specimens were also obtained from the lake by Marinus Boeseman during a collecting expedition for the Leiden Museum in 1954–1955.

The lectotype of *C. sentaniensis*, preserved in the Amsterdam museum, shows a very long snout, which according to Gerald Allen, is an important distinguishing mark. In his 1982 book, "Rainbowfishes of Australia and New Guinea" Allen showed *C. sentaniensis* among the colour sketches, because he didn't have any photographs of live specimens.





In 1982, live specimens of *Chilatherina fasciata* were collected from the Jafuri River and during the 1980s and early 1990s they were being distributed in the hobby as *Chilatherina sentaniensis*. In his *Field Guide to the Freshwater Fishes of New Guinea* of 1991 Gerald Allen noted that probably all records of *C. sentaniensis* in the aquarium literature related to *C. fasciata* varieties and that the most recent collection of the true *C. sentaniensis* was made in 1954. In yet another book, *Rainbowfishes in Nature and in the Aquarium*, published in 1995, Gerald Allen showed a picture of a live specimen, with the remarkably long snout, that was collected in a small tributary stream flowing into the north-eastern end of Lake Sentani in 1991. Several specimens were netted along with *Chilatherina fasciata* and *Glossolepis incisus*. None of these specimens however, were collected live for the aquarium hobby.

The "genuine" *Chilatherina sentaniensis* does however, exist in the hobby and they have a much longer, more pointed head and usually 9–12 soft dorsal rays compared to 11–16 rays in *Chilatherina fasciata*. Live specimens were collected from the lake in 1991 by Charles Nishihira and distributed in the aquarium hobby. Specimens were also collected from a small tributary stream in 2004 and 2005. However, they have not been widely available and only a handful of enthusiasts are maintaining them in captivity.

There is some concern that *C. sentaniensis* may not occur in Lake Sentani anymore, only in some of the feeder streams. The only currently known location is "Carwash Creek" which is crossed by the road from Sentani to Jayapura.

This creek is nowadays heavily polluted by upstream mining activities. In the last years, attempts to re-collect *C. sentaniensis* from this creek have been unsuccessful. It hasn't been found at any other location so far (J. Graf 2009, *pers. comm.*).



*Chilatherina sentaniensis*: Dorsal spines: 5–7; Dorsal soft rays: 9–12; Anal spines: 1; Anal soft rays: 21–26. 22-27 scales in front of dorsal. 10–12 soft dorsal rays (Weber, 1922).

*Chilatherina fasciata*: Dorsal spines: 5–8; Dorsal soft rays: 11–16; Anal spines: 1; Anal soft rays: 21–28. 19–21 scales in front of dorsal. 13–16 soft dorsal rays (Weber, 1922)







# **Glossolepis dorityi** Allen, 2001

# **Species Summary**

*Glossolepis dorityi* males have a body colour that is generally greenish with silvery reflections on the back, nape, and side of head. They have a dull orange or bronze stripe between each scale row of upper half of body; red-orange stripe between each scale row of lower half of body, especially prominent during courtship activities. Fins greenish to translucent, but with pinkish or red-orange hue on ventrals and basal half of anal and second dorsal. Female generally greenish with silvery reflections and lacking orange or red-orange stripes between scale rows. Males generally possess a deeper body and have elongated posterior rays on the dorsal and anal fins. Males may reach a maximum size of 10 cm, but females are usually less than 8 cm SL. This species is named "*dorityi*" in honour of Dan Dority for his efforts in collecting the type specimens.

# **Distribution & Habitat**

*Glossolepis dorityi* is currently known only from the Grimé River region of northern West Papua. The area was formerly known by the Dutch administrators as the Nimboran (Grimé) Plain. It is located roughly 50 kilometres west of Lake Sentani.

The type locality consists of a small round lake (Lake Nenggwambu or Lake Kali Biru). There is a vigorously flowing outlet stream, but no apparent inlet, indicative of a subterranean connection with neighbouring lakes via the limestone substratum. Water was relatively clear and maximum depth was estimated to be at least 10–15 m. The lake is surrounded by secondary forest and aquatic plants were abundant, but relatively few species

were evident. Fishes were most strongly congregated around the outlet, where vegetation was very dense. *G. dorityi* was the most abundant fish species and a second rainbowfish, *C. fasciata* was also common. The body colouration of this *C. fasciata* variety is mostly an orange to mauve and shows a golden luminescent nuptial stripe on their forehead which is switched on and off during spawning.

# Remarks

Dan Dority and David Price collected *Glossolepis dorityi* in April 2000. Heiko Bleher found this species in May 1999 in Lake Nenggwambu, but was unable to collect any specimens. However, in a nearby lake (Lake Jaigum) he collected specimens of the same species. Unfortunately only two males survived the journey back to Europe. He returned in November of the same year - but once again was unsuccessful in collecting specimens. However, during his third journey, at the end of 2000, he succeeded in catching six adult specimens. He also collected more specimens from Lake Jaigum. He eventually returned to Europe with four males and one female. Further live specimens were collected in 2008 by Dan Dority, Gary Lange and Johannes Graf. Specimens from both lakes are currently available in the hobby.

The rainbowfishes in Lake Nenggwambu are in serious danger of extinction due to the introduction of predatory fishes such as *Channa striata* and *Cyprinus carpio*. The formerly crystal-clear water is now muddy and dark brown. Water plants (*Ottelia sp.*) are completely gone. *G. dorityi* and *C. fasciata* are also heavily infested with *Lennaea sp.* (anchor worms) due to the introduction of the exotic fish species. The population of *G. dorityi* in Lake Jaigum is now thought to no longer exist in the lake (J. Graf 2009, *pers. comm.*).





▼ Lake Nenggwambu





# **Glossolepis incisus** Weber, 1907

Salmon-Red Rainbowfish

# **Species Summary**

Female *Glossolepis incisus* have a yellowish olive body colour with a golden iridescence to the scales, and clear fins. The males on the other hand are brilliant, the entire body and fins are a bright salmon-red colour. Some of the scales have a silvery sheen, which creates a most unusual effect over the red background colour. Young fish are all rather dull in colour, being an overall olive greenish colour with a trace of silvery sheen. However, once the fish reach a length of 4 to 5 cm the males begin to colour up. Once the colour change begins to occur it progresses quite rapidly. By the time the fish are 7 to 8 cm long the males should have their full intense red colouration. Males are typically deeper bodied than females and have a high rounded back which gives them the appearance of having a relatively small head and disproportionately large eyes. Males may reach a maximum size of 15 cm, but females are usually less than 12 cm.

*Glossolepis* differ from other melanotaeniids by a combination of characters which includes distinctly crenulate scale margins, a high gill raker count, spine at the beginning of the second dorsal fin taller than first spine of first dorsal fin, relatively elongate pectoral fins, a unique premaxillary dentition and characteristic profile of the head, nape, and dorsal and anal fins.

# **Distribution & Habitat**

Glossolepis incisus are found only in Lake Sentani. Lake Sentani is located some 10 kilometres west of Jayapura at the NE extremity of West Papua. It is an irregularly shaped lake with approximate dimensions of 28 km (E-W) by 19 km (N-S) and a surface area of 104 km<sup>2</sup>. Its blue-green waters are dotted with at least 16 small islands, and it is surrounded by hillsides in the south and the Cyclops Mountains in the north, which separate the lake from the Pacific Ocean. Lake Sentani is by far the largest of the West Papuan lakes and has a catchment area of about 600 km<sup>2</sup>. About 35 small rivers flow into the lake, and there is one natural outlet in the south-eastern tip, via the Jafuri and Tami rivers to the Pacific Ocean near the Papua New Guinea border. The lake is divided into three main sections with recorded depths of 7 to 52 metres. According to surveys in 1970-71, 1984 and 1987 the lake is thermally unstratified, with surface temperatures of 29–32° and pH6.2-6.8. Rainbowfishes are generally found around the margins of the lake. Large numbers are found congregating around submerged aquatic vegetation, fallen tree branches etc.





# Remarks

*Glossolepis incisus* was originally collected from Lake Sentani during the 1899~1900 Siboga Expedition to the Dutch East Indies (West Papua). The discovery and collection of rainbowfishes in remote places in New Guinea has always been very difficult and it wasn't until 1973 before live specimens of this species were collected. In 1973, A. Werner, Jr. of Munich, and E. Frech of Memmingen, Germany collected live specimens during a collecting trip to Java, Celebes, and West Papua. They took a number of colourful fishes back to Europe, including *Glossolepis incisus*. Werner and Frech are also credited with the introduction of another beautiful and appealing rainbowfish to the aquarium hobby - *Iriatherina werneri*. *Glossolepis incisus* came into the Australian hobby in 1977.

Comparison of certain characters for Glossolepis pseudoincisus and G. incisus		
Character	G. pseudoincisus	G. incisus
Horizontal scale rows	12-16	16-20
Vertical scale rows	38-43	50-60
Predorsal scales	27-34 (x = 31, N = 39)	30-36 (x = 36, N = 13)
Preopercle scales	21-29 (x = 25, N = 39)	26-38 (x = 31, N = 13)
Predorsal-Preanal distance	Predorsal > Preanal	Predorsal < Preanal
Pectoral fin colour	uniformly pale	outer portion dusky brown
Pelvic fin colour	mainly pale	mainly dusky brown





# **Glossolepis kabia**

(Herre, 1935) Sepik Rainbowfish

*Melanotaenia kabia* Herre, 1935 *Melanotaenia rosacea* Herre, 1935 *Lomanetia multisquamata* Whitley, 1936 *Glossolepis multisquamatus* Allen, 1980

# **Species Summary**

*Glossolepis kabia* have an overall body colouration of greenish or olive to silvery with a rosy glow across the sides. There is a series of narrow orange stripes between each scale row and the fins are usually clear or greenish but sometimes nearly black. They may reach a maximum size of 12 cm, but are usually less than 10 cm. Most fish collected have been within the range 6-10 cm. Adults become very deep bodied especially the males (6-8 cm), although this difference is not obvious in young adults less than about 6 cm SL. The orange stripes on the sides are brighter in mature males and their dorsal and anal fin membranes often have a silvery to yellow sheen. Named *kabia*, from *kabi*, the native name at Koragu.

The original description described their living colours as dusky silvery above, the lower half with alternate vertical stripes of golden orange or deep orange and silver or steely blue, with an orange band at the caudal base. The top of the snout and interorbital are black, the opercles and preopercles silvery with yellow or golden. The fins are more or less dusky in some, in others the membranes of the dorsal and anal are orange, wholly or only basally, the rays and outer part blackish.

### **Distribution & Habitat**

*Glossolepis kabia* is found in the Sepik and Ramu river systems in New Guinea. They are found in floodplains and swampy lagoons, lakes, and small tributary streams. It is the only rainbowfish that is abundant in the extensive floodplain regions of the Sepik River. They are usually found where there is an abundance of aquatic vegetation in moderate turbid to clear, still to slow-flowing water. Young fish form aggregations around submerged logs and branches or among reeds and other shoreline vegetation.

The Sepik River (formerly known as the Kaiserin Augusta River) is the second largest river in Papua New Guinea (PNG). The river is about 1100 kilometres long with a catchment area of approximately 80,000 km<sup>2</sup>. It is the largest river system in PNG in terms of catchment area, but has a lower discharge than the Fly River. Biologically, the region holds some of the most diverse and least described ecosystems on Earth, and is probably the largest uncontaminated freshwater wetland system in the Asia-Pacific region. There are no large mining projects, no industrial plants and no large timber extraction projects operating within the region and, compared to other areas of New Guinea, much of the area has a low rate of population growth.

The Sepik begins in the Victor Emanuel Range in the central highlands. It leaves the mountains abruptly near Yapsei on the border with West Papua where it becomes a strongly braided channel flowing in a north-westerly, then northerly direction. Turning east it follows the great Central Depression, receiving numerous tributaries draining from the Bewani and Torricelli





Mountains in the north and the Central Range in the south before entering the Bismarck Sea. For most of its lower course the river meanders through a wilderness of swamps and lagoons with large floating islands of vegetation.

The water temperature at lower elevations on the Sepik River usually remains within the range 27-29°C. It is thought that average temperatures in the Sepik tributaries stay relatively low at lower altitudes than in most equatorial regions. This is partly due to the unusual topography of the basin in which lowland swamps are surrounded on most sides by relatively steep mountains resulting in cooler water being transported rapidly into the lowlands. Secondly, the degree of riparian shade covering feeder streams and rivers is pronounced in the Sepik in comparison with many other regions which have lost much of their vegetative cover through human activity. Surface water temperature commonly varies between 28°C and 35°C, and the bottom waters between 24.8°C and 30°C. Conductivity levels in the Sepik are high (110–250  $\mu$ S/cm) although this is most likely related to hardness and dissolved organic matter, rather than to nutrients. The Sepik water chemistry is dominated by calcium and bicarbonate ions. In the Middle Sepik water conditions were reported as conductivity 103-125 µS/cm and total hardness 45–56 mg CaCO<sub>3</sub>; slightly acidic pH and low alkalinity. Seasonal of the lakes have different water chemistry.

Formerly called the Ottilien River, the Ramu is one of the longest rivers in Papua New Guinea, rising in the southeast on the Kratke Range and flowing northwest through the great central depression, where it receives numerous streams draining the Bismarck (south) and Finisterre and Adelbert (north) ranges. For the last 100 km of its approximately 720-kilometre long course, it flows directly north. This swampy portion receives the river's principal tributary, the Sogeram River. The Ramu enters the Bismarck Sea just 32 km southeast of the mouth of the Sepik. The lower reaches of the Sepik and Ramu rivers are now inter-connected by numerous channels that pass through an area of low-lying alluvium. The freshwater ichthyofaunas of the Sepik and Ramu basins are very similar, although there is a modest degree of species endemicity in both regions.

#### Biology

*Glossolepis kabia* generally become sexually mature at about 6 cm (females) and 7 cm (males). Individuals with either ripe or spent gonads are rarely found below this size, which is relatively large compared with most rainbowfishes. Males and females in spawning condition have been found throughout the wet and dry seasons indicating that *Glossolepis kabia* breed all year round. However, fish in spawning condition was significantly lower in the dry season in both sexes which suggests a reduced reproductive rate at that time. The dry season is defined as May to October and the wet season as November to April.

The reproductive capacity of *Glossolepis kabia* is considered to be high compared with most other Sepik River fish species spawning in freshwater. Fecundity in *Glossolepis kabia* is



perhaps twice that of equivalent sized, stream dwelling, *Melanotaenia fluviatilis*. One factor increasing fecundity in *Glossolepis kabia* is egg size which is much lower than reported for other rainbowfish species. Most rainbowfishes have egg diameters of greater than 1.0 mm and up to 2.0 mm. The eggs of *Glossolepis kabia* ( $0.69 \pm 0.052$  mm) are, therefore, between about 4% and 35% of the size of eggs from other species on a volume basis. Reduced egg size results in a much higher fecundity than occurs in stream dwelling rainbowfish species.

The diet of *Glossolepis kabia* appears similar to observations on other species of rainbowfish and the species exhibits little feeding specialisation within this group. A similar diet has been recorded for four other species of rainbowfishes which inhabit the Sepik River streams. This generalised feeding habit is also confirmed for several Australian rainbowfishes and one species from the Fly River in southern Papua New Guinea. They are carnivorous, feeding predominantly on a wide variety of small invertebrates taken mainly from mid-water or from the water surface. The percentage of insect larvae and insects from terrestrial sources increase slightly in the wet season, with reductions in the percentage of these and slight increases in the percentage of crustaceans, detritus and 'other material' in the dry season. Food intake increases during the wet season and decreases during the dry season.

#### Remarks

Glossolepis kabia was originally collected during the Crane Pacific Expedition in May, 1929. The Crane Pacific Expedition was sponsored by Cornelius Crane for the Field Museum in Chicago, USA. The expedition left Boston on November 16, 1928, in the yacht Illyria and spent January to May 1929 visiting a variety of countries and island groups in the Pacific region, including the Marquesas, Tahiti, Fiji, Vanuatu (then the New Hebrides), New Britain and New Guinea. Their longest stay was in northern Papua New Guinea, where they sailed up the Sepik River and stopped at several villages and mission stations, then continued on up the May River, visiting two or three more villages. On their return, they look a short detour to the Keram River then revisited a few settlements on the Sepik. Very few fish were obtained but many of those collected were new. Among the fish collected were some rainbowfishes, later described by Albert W. Herre as Melanotaenia kabia, M. rosacea, and Rhombosoma sepikensis. The type specimens of Glossolepis kabia were collected from the Sepik River at Nyaurangai, about 300 kilometres from the sea. Additional specimens were also collected at Koragu and the Kerame River.

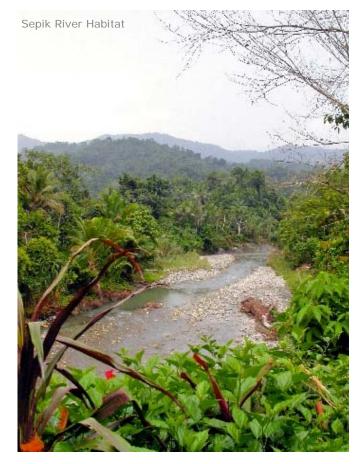
In 1979, Gerald Allen collected a number of live rainbowfishes while collecting in the Sepik-Ramu River region, and brought them back to Australia where they were subsequently bred and distributed in the hobby. He later identified them as *Glossolepis multisquamatus*. Heiko Bleher also collected live specimens from the same area and another collection near the Ramu River and took them back to Europe. However, it is thought that none of these fish still remain in captivity.

In 1992, Heiko Bleher collected a number of *Glossolepis* specimens from the Mamberamo River area in West Papua. It was unclear whether these specimens represented an undescribed species or perhaps just a colour variation of *G. multisquamata*.

Heiko Bleher had collected this form a couple of times and reported that in his opinion they are quite different from the Sepik/Ramu form. However, the specimens collected from the Mamberamo River are the true *G. multisquamata*.

The Mamberamo River specimens are quite different, but the most obvious difference is they have quite large anal fins, which are reminiscent of *G. wanamensis*. They also have a really bright red eye too which seems somewhat unique in rainbows (P. J. Unmack 2009, *pers. comm.*). These species were bred and distributed in Australia under the common name of "Red-eyed Tiger Rainbowfish". In Europe, they were generally known as *Glossolepis* sp. (Mamberamo). The fish from the Mamberamo River system are *G. multisquamata* while those from the Sepik-Ramu River system are considered as *G. kabia*.

Additional live specimens of *G. Kabia* were collected by Kent Webster in 2002 from Paro Village in the middle Sepik River region and introduced to the aquarium hobby.







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# **Glossolepis leggetti**

Allen and Renyaan, 1998 Leggett's Rainbowfish

# **Species Summary**

*Glossolepis leggetti* was described from 79 specimens collected in 1998. The males have a body colouration that is iridescent green on the back, dull orange to whitish on the breast and lower sides. A diffused blue midlateral band, its colouration most intense posteriorly and bordered immediately below by narrower stripe of light metallic blue. Sides with scattered dark blur flecks or narrow bars. Fins generally translucent although dorsal and anal may be bluish, particularly in adult males. Females are basically the same colour, but less ornate with a narrow midlateral stripe and with the dark flecks and narrow bars on the side considerably reduced. May reach a maximum size of 10 cm, but usually less than 8 cm SL.

*Glossolepis leggetti* is most closely related to *Glossolepis kabia* from the Ramu, Sepik, and Mamberamo river systems of northern New Guinea. The two species are similar in general appearance and coloration, although *Glossolepis kabia* usually lacks scattered dark markings on the side of the body, which are typical of *G. leggetti*. Moreover, *G. leggetti* usually has a higher number of soft dorsal rays and fewer predorsal scales (18–23 versus 24–31). In addition, males (in excess of 50 mm SL) of *G. leggetti* are generally more slender than those of *G. kabia*.



# **Distribution & Habitat**

*Glossolepis leggetti* is currently known only from the Wapoga River system of northern New Guinea. They were found in relatively clear, quiet pools of the Tiawiwa River, a major tributary of the Wapoga, in the vicinity of Siewa airstrip. They were found together in the Tiawiwa River with *Chilatherina alleni* and *Melanotaenia rubripinnis*, but unlike these species which are most abundant in shallow relatively rapid sections, it favours deeper pools, with minimal flow.

# Remarks

*Glossolepis leggetti* was named 'leggetti' in honour of Ray Leggett of Brisbane, Queensland in recognition of his contributions to the knowledge of freshwater fishes of the Australian-New Guinea region. This species is not currently available in the aquarium hobby.





# **Glossolepis maculosus**

Allen, 1981 Spotted Rainbowfish

#### **Species Summary**

Glossolepis maculosus males have an overall body colour of greenish-bronze with faint pale-blue reflective scales and a series of dark spots or blotches along the lateral line. These may number from 4 to 10 but can vary considerably. Each individual has its own pattern of distinctive spots along the sides. Even the left and right sides of each individual can be differently marked. Below the lateral line the body colour is whitish anteriorly and yellowish posteriorly. Fins are blueygreen with a slight hint of orange and black margins. The colouration of the female is drab compared to the male, but they still show distinct spots, but otherwise are pale olive with a silvery belly. Mature, older females often show colouration similar to subordinate males, but are usually easily identified by a shallower body/chest depth and smaller, more rounded fin edges. Mature males are usually much larger and deeper bodied than females and have a higher first dorsal fin, which overlaps the origin of the second dorsal fin when depressed. Glossolepis maculosus may reach a maximum size of 8 cm, but usually less than 6 cm.

# **Distribution & Habitat**

*Glossolepis maculosus* are so far only known from a few localities in the Markham, Ramu and Sepik river systems of northern Papua New Guinea. Initially discovered and collected by Barry Crockford during 1979, in a small tributary of the

Oomsis River about 22 km west of Lae, Papua New Guinea. He collected a small number of live specimens and returned to Australia. He later sent a coloured drawing of the fish to Gerry Allen at the Western Australian Museum in Perth who confirmed that he had collected a new species of rainbowfish. A year later Gerry Allen returned to this location with Barry and collected several more live fish, which were later bred and distributed in the Australian hobby. Some years later Heiko Bleher collected a small number of live specimens from the Ramu River valley and introduced them to the European hobby. They have been found cohabiting with *Glossolepis kabia*, *Melanotaenia affinis*, and *Chilatherina campsi*.

*Glossolepis maculosus* are a stream dwelling species and inhabit slow-flowing streams, swamps, and quiet backwaters. Occurs most frequently in relatively still, clear water, in water temperatures between  $18-28^{\circ}$  Celsius. Water conditions recorded at one collecting site were temperature  $25^{\circ}$  Celsius, *p*H 7.8 and hardness 80 ppm. They are usually found along grassy banks, or around sub-surface vegetation, submerged logs, and branches.

# Remarks

*Glossolepis maculosus* have never been widely available and even today could still be considered uncommon. Only a handful of enthusiasts are maintaining them in captivity. Due to the small number of founding stock collected, the colouration of specimens in captivity has changed over the years. Nevertheless, they are still a very attractive species and if someone is willing to spend some time with them, they could end up with a beautiful aquarium fish.





# **Glossolepis multisquamata**

(Weber and de Beaufort, 1922) Mamberamo Rainbowfish

*Melanotaenia multisquamata* Weber & de Beaufort, 1922 *Glossolepis multisquamatus* Allen, 1980 *Glossolepis multisquamata* Eschmeyer, 1998

# **Species Summary**

*Glossolepis multisquamata* have an overall body colouration of olive-green to silvery with a rosy glow across the sides. There is a series of narrow orange-red lines between each scale row and the fins are usually clear, greenish-yellow with a hint of red, or sometimes nearly black. The orange-red lines on the sides are much brighter in mature males giving the body an overall reddish colour. Males also have a bright red eye. They may reach a maximum size of 14 cm, but are usually around 10–12 cm. Most fish collected have been within the range 6–10 cm. Adults become very deep bodied especially the males (6–8 cm), although this difference is not obvious in young adults less than about 6 cm SL.

# **Distribution & Habitat**

Glossolepis multisquamata is currently found in lakes, slow-flowing streams and backwaters of the Mamberamo River

system in West Papua. They were originally collected from the Idenburg River (= Taritatu River) in the Mamberamo region by the Dutch zoologist and explorer W. C. van Heurn in 1920. They were also collected from the Doorman River, a major tributary of the Taritatu.

The Mamberamo region is not well-studied but possesses a wealth of biodiversity. The first scientific fish collections from the Mamberamo were made by van Heurn in 1920-21. These collections were mainly described by Weber and de Beaufort (1911–1962). There have been some other expeditions; 1938–1939 (Archbold expedition) and Gerald Allen did some surveys in this area. Several locations within the Mamberamo catchment, including Danau Biru, Obogwi, Faui, Kordesi, Dabra, Nevere, and Senggi were surveyed by Gerald Allen between 1982 and 1991.

The Mamberamo River (also called Tarikaikea) is the largest in northern West Papua, draining a catchment that encompasses all northward flowing streams descending from the New Guinea central mountains between the Papua New Guinea border and approximately 137° west longitude. The source of the river is formed from the confluences of its upper tributaries, the Tariku, Van Daalen and Taritatu Rivers. The Tariku River (previously known as the Rouffaer River) in the west flows eastward and the Taritatu River (previously known as the Idenburg River) in the east flows roughly westward. They meet





in the Meervlakte Basin to form the main Mamberamo River. Extensive inland swamps surround the Taritatu and Tariku rivers in the central depression of the Lakes-Plains province. Beyond the confluence of Tariku and Taritatu, the Mamberamo flows abruptly northwards 175 km through the Van Rees Range to reach the lowland marshes of its broad river delta on the coast at Cape D'Urville on the northeast margin of Cenderawasih Bay. The Mamberamo River has a total length of about 1000 km; the Taritatu is about 467 km long; the Tariku 327 km; and the Mamberamo itself is about 283 km long.

#### Remarks

Gerald Allen collected a number of live rainbowfishes in 1979, while collecting in the Sepik-Ramu River region, and brought them back to Australia where they were subsequently bred and distributed in the hobby. He later identified them as *Glossolepis multisquamatus*. Heiko Bleher also collected live specimens from the same area and another collection near the Ramu River and took them back to Europe. However, it is thought that none of these fish still remain in captivity.

Heiko Bleher also collected a number of *Glossolepis* specimens from the Mamberamo region in 1992. It was unclear whether these specimens represented an undescribed species or perhaps just a colour variation of *G. multisquamata*.

Heiko Bleher had collected this form a couple of times and reported that in his opinion they were quite different from the Sepik/Ramu form (H. Bleher *pers. comm.*). These species were bred and distributed in Australia under the common name of "Red-eyed Tiger Rainbowfish". In Europe, they were generally known as *Glossolepis* sp. (Mamberamo).

Around 1993, Heiko Bleher collected another rainbowfish from Lake Kli in the Mamberamo valley. It is generally known in the hobby as *Glossolepis* sp. (Lake Kli) or as the Fringefin Rainbowfish. Since then there has been a number of collections by individual aquarists and some of these forms are currently available in the hobby.

As it turns out, the specimens from the Mamberamo region are the true *Glossolepis multisquamata* while those from the Sepik-Ramu River system are considered to be *Glossolepis kabia*. The Mamberamo River specimens are quite different, but the most obvious difference is they have quite large anal fins, which are reminiscent of *Glossolepis wanamensis*. They also have a really bright red eye which seems somewhat unique in rainbows (P. J. Unmack 2009, *pers. comm.*).

Additional live specimens were collected and introduced into the aquarium hobby in 2009.











# **Glossolepis pseudoincisus**

Allen and Cross, 1980 Tami River Rainbowfish

# **Species Summary**

From October 1954 through to May 1955 Marinus Boeseman took part in a collecting expedition for the Rijksmuseum van Natuurlijke Historie to Netherlands New Guinea (West Papua). His task was to provide a thorough knowledge of the fish fauna by intensively surveying as many rivers and lakes as was possible. This task was taken to heart and in a relatively short period many localities were visited, resulting in a rich collection for the museum in Leiden. Among the places he visited was Lake Sentani, Tami River, Biak Island, Lake Jamoer (Yamur), Wissel Lakes, Ajamaroe (Ayamaru) Lakes, Lake Aytinjo, Merauke and the Digul River. This collection included many rainbowfishes, but a thorough study of this material or descriptions of any new species was never made by Boeseman.

As part of his preparation for the revision of the rainbowfish family, Gerald Allen studied the Boeseman collection of 1954–55 during 1975 and 1977. He discovered no less than four new rainbowfish species, which he described in 1980 together with Norbert Cross. These species were *Melanotaenia boesemani*, *M. ajamaruensis*, *M. japenensis* and *Glossolepis pseudoincisus*.

*G. pseudoincisus* is most closely related to *G. incisus*. They differ from the other members of the genus by possessing more gill rakers on the first arch (26–32 *vs.* 19–23) and by having more pronounced crenulations on the scale margins. These species differ from one another on the basis of the characters presented in Fig. 1.

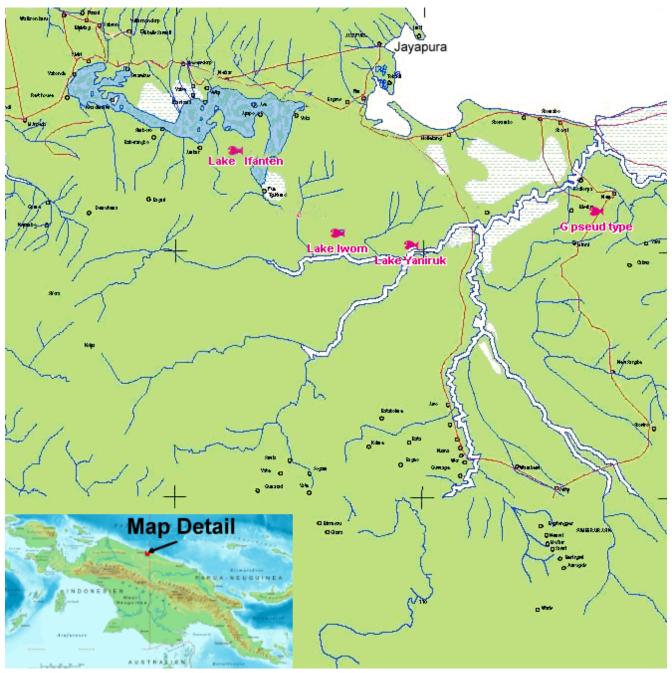
# **Distribution & Habitat**

Specimens of *G. pseudoincisus* were collected by Boeseman in November, 1954 in an ox-bow lake of the Tami River, about 30 kilometres to the east of Lake Sentani. They are also known from Lake Ifaten, Lake Iwom and Lake Yaniruk. The species was named 'pseudoincisus' with references to its similar appearance and geographic proximity to *G. incisus*.

#### Remarks

Gerald Allen and Heiko Bleher went looking for this species in 1982, but were not able to find it. However, in January 2001, Heiko collected some *Glossolepis* species from Lake Ifaten, an isolated crater lake situated in the mountains near Lake Sentani in West Papua. Lake Ifaten is about 300 metres above sea level. The lake has a diameter of around 250 metres. Water temperature  $28^{\circ}$ C, *p*H 9.0.





Distribution map of known locations of *Glossolepis pseudoincisus* [Johannes Graf]

# Fig. 1

Comparison of certain characters for Glossolepis pseudoincisus and G. incisus		
Character	G. pseudoincisus	G. incisus
Horizontal scale rows	12-16	16-20
Vertical scale rows	38-43	50-60
Predorsal scales	27-34 (x = 31, N = 39)	30-36 (x = 36, N = 13)
Preopercle scales	21-29 (x = 25, N = 39)	26-38 (x = 31, N = 13)
Predorsal-Preanal distance	Predorsal > Preanal	Predorsal < Preanal
Pectoral fin colour	uniformly pale	outer portion dusky brown
Pelvic fin colour	mainly pale	mainly dusky brown





Only five specimens (4 males/1 female) survived the long journey back to Europe. The fish look similar to *G. incisus*, but the scales are different (they're smaller and differently aligned).

The Lake Ifaten rainbowfish is not as big as *G. incisus*; the body shape is more compact; the red coloration is more intense and the fin marking and shape is more pronounced. Also the Lake Ifaten rainbowfish shows colour after three month of age (*G. incisus* needs nearly a year before they show proper colours). At the age of four months and with a body size of hardly more than 4 cm, they showed the full colour (H. Bleher, *pers. comm.*).

The Lake Ifaten Rainbowfish has a very typical *Glossolepis* head. Very prominent at an age of one year, are the marks on the gill plate. A unique pattern of red lines that - so it appears - are drifting criss-cross. The females of the Lake Ifaten Rainbowfish are differently coloured. They have strong horizontal zigzagging yellow stripe colouration across the whole body. They also remain small - up to 6 cm total length.







▲ ▼ Glossolepis pseudoincisus (Lake Yaniruk)



Christopher Mailliet



# **Glossolepis ramuensis** Allen, 1985

Ramu Rainbowfish

# **Species Summary**

Glossolepis ramuensis was described on the basis of a single male specimen, found in October 1983 in a tributary of the Ramu River, about three kilometres south of Walium Village in northern Papua New Guinea. Further specimens were collected in 1987. Males are greenish-brown to purplish on the back and white or mauve on the lower half. There are several narrow orange horizontal lines on the sides; those on the middle above and below the midlateral band are the most vivid. Mature males are usually much larger and deeper bodied than females and have a higher first dorsal fin, which overlaps the origin of the second dorsal fin when depressed. Similar to G. maculosus, young G. ramuensis also show a pattern of small spots on the sides, which disappears with ageing. G. ramuensis may reach a maximum size of 10 cm, but are usually less than 8 cm. Spawning occurs from October to December, with females producing between 50-100 eggs.

# **Distribution & Habitat**

Known only from the Ramu Valley and tributaries of the Gogol River near Madang, Papua New Guinea.



*G. ramuensis* inhabit small freshwater streams flowing through rainforest. The streams generally have clear water, a gravel bottom, and very few aquatic plants. Temperature and pH ranges from 26–29° Celsius and pH 7.4–7.9. It is sometimes found together with *Chilatherina campsi* and *Melanotaenia affinis*.

#### Remarks

Live specimens were collected for the aquarium hobby from the Gogol River in 1988 by Heiko Bleher. *Glossolepis ramuensis* have never been widely available and are still considered as uncommon. Only a handful of enthusiasts are maintaining them in captivity.









### **Glossolepis wanamensis** Allen and Kailola, 1979 Lake Wanam Rainbowfish

# **Species Summary**

Generally, the body colour of adult male *Glossolepis wanamensis* is an overall greenish colour with a rosy flush on the breast. The anterior scale rows below the lateral line are marked with narrow orange lines becoming green or bluish towards the tail. The upper half of the body often shows a metallic green colouration. The anal fin of the male is very large and elongated reaching a depth of nearly 3 cm. Females are generally a dull greenish colour, have shorter fins and rather drab compared to the males. Males are easily distinguished from females by their brighter colours and the extremely large anal fin. Males may reach a maximum size of 10 cm, but females are usually less than 8 cm. With a body depth of 4–5 cm, adults become very deep bodied especially the males.

#### **Distribution & Habitat**

*Glossolepis wanamensis* has only been found in Lake Wanam. They were collected in shallow, clear, sunlit water around sub-surface vegetation, submerged logs, and branches, or among reeds and other shoreline vegetation.

The temperature and pH recorded at the lake was 28° Celsius and pH 7.0–7.8. Lake Wanum is a freshwater lake centred on 6° 38'S and 146° 47'E, and located in the vicinity of Mount Ngaroneno, at the southern margin of the lower Markham Valley, near Oomsis, Morobe Province about 25 km inland from the Huon Gulf. It has an irregular outline and, with a maximum width of about 3 km, is the largest of a number of lakes and swamps in the vicinity.

As there is no permanent stream inflow into the lake much of its water is derived by precipitation directly onto the lake surface. The lake has a maximum depth of around 19 metres, and is subject to seasonal fluctuations in level. The only outflow of the lake is a small channel in the extreme south-west corner of the basin that runs into Oomsis Creek. This flows only intermittently and often dries out completely in the dry season when the level of the lake becomes lower. This channel also serves as an inflow when the creek is in flood. Oomsis Creek is the only permanent watercourse in this locality but it too may cease flowing under seasonal conditions of sustained dry weather.

#### Remarks

Lake Wanum is home to two rainbowfishes, *Glossolepis* wanamensis and *Chilatherina fasciata*. *G. wanamensis* was first collected by C. Ellway in 1975 but it wasn't until 1979 that they were scientifically described. This followed their







collection by Gerald Allen and Brian Parkinson in October 1978. Brian Parkinson had previously collected specimens there and sent them to Patricia Kailola, then working for the Fisheries Department at Port Moresby. Fifty-five specimens of *Glossolepis wanamensis* and four specimens of *Chilatherina fasciata* were collected. After just two days there were only five survivors and of these only two made it back to Australia. Barry Crockford brought more live specimens to Australia in 1980. Five survived, which included 2 females. A year later further live specimens were collected. The fish collected on these two trips formed the breeding stock of all *Glossolepis wanamensis* in Australia to the present day. Water conditions at the time were reported as *p*H 7.6, temperature 28°C and hardness 80 ppm.

In 1992, Heiko Bleher collected live specimens from the lake and introduced them to the European hobby. He again collected in 1994 and reported that "An exhaustive search produced just one small group of rainbowfishes, adults 2-3 years old, seven males and a single elderly female. No juveniles, no eggs. The water was murky from surface to substrate, with tilapias as far as the eye could see." He returned in 1995 and noted that Tilapia infestation of the Lake had increased even further and only two very old male specimens were collected. He reported in Aqua Geographia (1998), that Glossolepis wanamensis was becoming extinct in the Lake. It was thought that the introduction of carp, tilapia, and gambusia may have caused this drastic and worrying reduction in numbers. Oreochromis mossambica were introduced into the lake by the Department of Agriculture, Stock and Fisheries reportedly during 1966, and their increasing population may have made some impact on the lake's ecosystem.

During the 1998 ANGFA Convention in Brisbane, discussions were held with Heiko Bleher about the situation at Lake Wanum and it was decided that further survey work would need to be undertaken. It was felt that the endemic Lake Wanum rainbowfish was in such low numbers as to cause concern for its future in the wild. Matt Vincent and Gary Slater from the Melbourne Zoo travelled to PNG to discuss the problem with Peter Clarke, Director of the Rainforest Habitat in Lae. These discussions led to the setting up of a tripartite agreement between ANGFA, Melbourne Zoo and the Rainforest Habitat. These three bodies formed the Lake Wanum Management Project on 21 December 1998.

An extensive survey of Lake Wanum was undertaken in June of 1999. The results of this survey can be found in Fishes of Sahul 13(3): 621-629. This survey found that *Glossolepis wanamensis* were in reasonably large numbers in the lake, but that the *Chilatherina fasciata* found in the lake on previous collecting trips had all but disappeared. Despite extensive sampling of areas known to contain hundreds of specimens in the past, not a single fish was collected. However, among all the *G. wanamensis* collected only two juvenile specimens were noted. The remainder of the *G. wanamensis* were approximately two years of age. The survey team noted small groups of fry but was concerned by the low numbers within these groups (approx 20 specimens).



It was thought that during a severe El Niño drought in 1997, when water levels fell by 11.2 metres, it caused a massive fish kill in the lake. Apparently dead fish were floating everywhere and thick on the shoreline. *G. wanamensis* are known to withstand high temperatures and perhaps this event allowed the population to recover temporarily. However this fluctuation in population numbers did highlight the vulnerability of this species and it was decided to establish a separate breeding population at the Rainforest Habitat in Lae.

About eighty *G. wanamensis* were collected from Lake Wanum. Oomsis Creek was also surveyed and about twenty *Chilatherina campsi* were collected. Both collections were taken back to the Rainforest Habitat. *G. wanamensis* did not travel well and some losses occurred. The fish were placed into a tank and treated with medication. *C. campsi* travelled better and most of these were released directly into the ponds at the Rainforest Habitat. The following day the fish seemed to have improved, but more losses occurred (mostly females). About ten *G. wanamensis* were released into a pond in the Butterfly House and these improved dramatically so it was decided to select the best and fittest females from the tank and place these in that pond. It appeared at the time that the pond fish were surviving and that a breeding colony would be achieved.

The Rainforest Habitat began operations in 1994 on a tenhectare section of the University of Technology campus in Lae, Papua New Guinea. The Rainforest Habitat and its sister company the Insect Farming and Trading Agency are both operated by the University of Technology as self-funding community development initiatives. Today the Rainforest Habitat is maintaining populations of three rainbowfish species: *Glossolepis wanamensis, Melanotaenia affinis* and *Chilatherina campsi.* 

The current situation in Lake Wanum and its rainbowfish inhabitants is unknown. Very little information on the lake and the health of its ecosystem is available. However, small captive populations of *G. wanamensis* and *C. fasciata* from Lake Wanum still exist in Australia and internationally.





# Iriatherina werneri Meinken, 1974

Threadfin Rainbowfish

# **Species Summary**

In 1973, two visiting German aquarists collected some small freshwater fishes in a rice paddy field on the outskirts of the town of Merauke in New Guinea. They were transported back to Europe and a number of them were given to Herman Meinken, a well known aquarist and ichthyologist, who realised that they were an undescribed species. In 1974, Meinken published the scientific description of the fish in the German aquarium magazine Das Aquarium (Aqua Terra) and they were named *Iriatherina werneri* after one of the collectors, Arthur Werner. They are commonly known as the 'Threadfin' or 'Featherfin' Rainbowfish. In Australia the 'Standard Names of Australian Fishes' published by the CSIRO lists them as 'Threadfin Rainbowfish'.

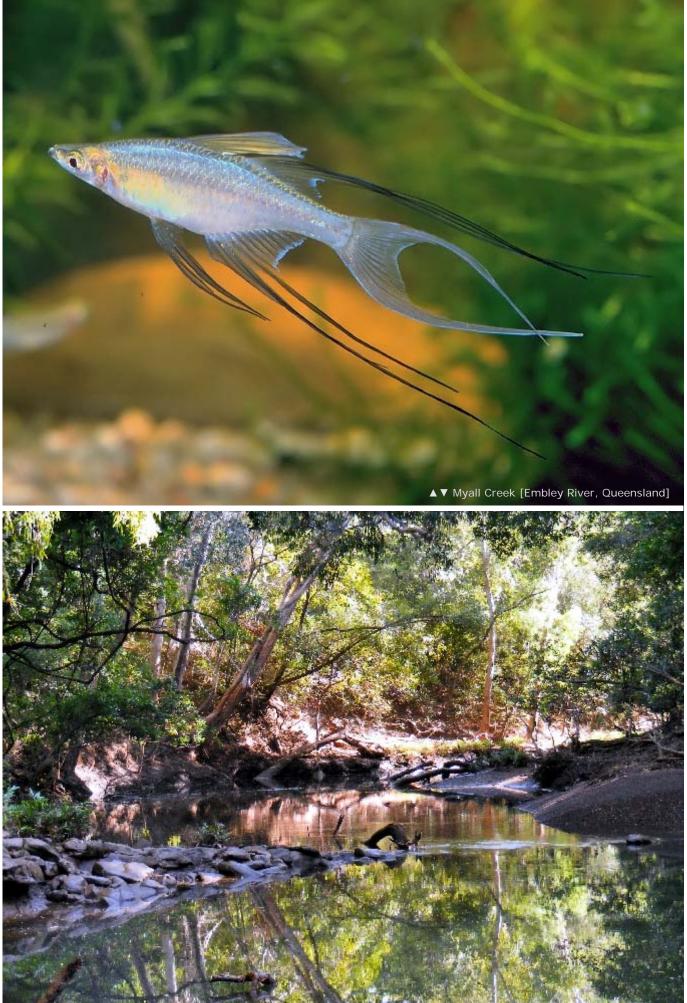
When *Iriatherina werneri* were first released to the aquarium trade in Europe only males were sold to the unsuspecting aquarium public. This of course meant that all attempts to breed this new species failed. Then in 1978, more of these delightful little jewels were found in swamplands of the Jardine River in Cape York Peninsula. Their discovery in Australia opened up the trade in the species with both males and females being freely available, much to the delight of aquarium hobbyists. *Iriatherina werneri* may grow to a maximum size of 5 cm, but are more commonly seen at around 3–4 cm. Mature males have a first dorsal fin that is fan shaped, while the second dorsal fin has exceptionally long filaments. The anal fin is similarly extended. This elegant finnage is used in a remarkably vivacious display for females and rival males. The body is slender, laterally compressed and general metallic silver with slightly visible dark vertical bars. The colours of the dorsal, anal and pelvic fins are black with a reddish-tan wash. The tail fin is deeply forked, transparent, and edged in a rustic red colouration.

There are also small differences in fin shape and colouration of male specimens from different locations. Some males have a narrow and high sail-like first dorsal fin, while this fin is lower and broader in other males. Specimens from the Cadell River in the Northern Territory often have a more lyre-tail shaped tail fin, and the fins may be darker and longer. Females however, pale in comparison to the males, although their tail fins are adorned with a pinkish margin and the edges of the pelvic and anal fins are edged with black.

The colouration of the fish in their natural habitat is usually much more intense than specimens maintained in the confines of an aquarium. Specimens found in New Guinea are usually darker than the Australian variety. A yellow finned variety has been collected from a tributary of the Embley River in northern Queensland.







Preliminary genetic studies (P.J. Unmack 2009, *pers. comm.*) have revealed significant genetic variation between the geographically distinct populations of *Iriatherina werneri* that occur in north Queensland and the Northern Territory that warrant taxonomical separation at the species level. Ongoing studies looking at the New Guinea populations may also indicate a third species.

# **Distribution & Habitat**

In New Guinea, *Iriatherina werneri* have been collected from several coastal rivers between the Merauke and the Fly Rivers. In the Fly River they have been collected in swampy lagoons along the mainstream of the river more than 500 km upstream from its mouth. In 1975 they were collected from Lake Bosset. Then in 2005, they were collected in 2 sites, Lake Bosset and Lake Kala. In Australia, the Jardine River swamps were believed to be the only place where they could be found. However, in 1985 (Hansen, 1987) they were found in the Edward River catchment on the western side of Cape York Peninsula. More recent surveys have found that they have a much wider distribution in a number of rivers both on the western and eastern sides of Cape York Peninsula. They have also been collected from the Arafura Swamps and a number of streams in the Northern Territory.

*Iriatherina werneri* are generally found in clear, slow flowing streams, grassy wetland swamps and lagoons that have abundant vegetation. They are most commonly found along the heavily vegetated margins of lagoons and small streams at depths of 0.5 to 1.25 metres, and usually in open water not far from clumps of vegetation. Small schools of females and juveniles can be seen moving slowly through the water while the more mature males display around them. A temperature range of 22° to 30°C and pH values of 5.2–7.5 have been recorded in their natural habitats.

# Keeping & Caring

Very little is known about the natural life history and ecology of the Iriatherina werneri in the wild. Most information is mainly based on aquarium observations. They have been a popular aquarium fish with Australian native fish enthusiasts for many years and are easy to maintain under standard aquarium conditions. Although they are a fish that requires a little more attention than most other rainbowfishes. They should not be kept in aquaria less than 60 cm (55 litres) and are best kept in groups of 5 or more individuals. They can be maintained and bred in water conditions that are suitable for most aquarium species. Best results will be achieved if maintained at a temperature range of 23–28°C; pH 6.0–7.0; and water hardness of 10-70 ppm (mg/L). I did however, maintain this species for 16 years and successfully spawned and raised numerous generations under the following water conditions: Temperature 24-31°C, pH 5.9-8.2, Hardness 100-220 ppm, Alkalinity 25-100 ppm and conductivity 390-820 µS/cm.

*Iriatherina werneri* is one of the most peaceful of all aquarium fishes, almost to the point of exclusion as a potential "community" tank fish. Their timidity, gentle manner, small

size and dainty feeding habits put them at a disadvantage in an aquarium with most other types of tropical fishes. This is probably why they are not readily available in aquarium stores and are mostly maintained by rainbowfish enthusiasts. Nevertheless, they can be kept in a mixed community aquarium containing other small native species such as *Pseudomugil gertrudae*. A breeding tank can be easily set up and the fish induced to spawn throughout the year.

In their natural environment they feed mainly on large quantities of unicellular, planktonic algae and diatoms. However, in captivity they seem to thrive on live brine shrimp nauplii, daphnia, copepods, mosquito larvae, and worms such as grindal and microworm. The challenge of attacking larger live foods is usually declined. Floating dry foods, such as small bite-sized pellets or flake foods are acceptable as their dorsally projected jaws are designed for surface feeding.

I first obtain this species in 1982. They were part of a collection of fishes from the Jardine River. No information was given regarding the water conditions of the collecting site. I set them up in a 55 litre breeding aquarium with subgravel filtration containing aged tap water with a pH 7.6 and a hardness of 140 ppm. Temperature varied between 24–27°C, a woollen spawning mop was added to complete the set-up. The spawning mop was checked daily and when eggs were found the mop was removed to a hatching tray and replaced with another. Getting them to spawn and hatch was the easy part, raising the fry was another story. The fry were fed infusoria and commercial fine powdered food. Mortalities were high; in fact it took almost two months before I was successful in raising any offspring.

High mortality rates can often occur, especially during the early feeding stages. The main reason for this is that the developing larvae are very small, extremely fragile, and generally not physiologically fully developed. For example, their small mouth size is a limiting factor in proper feed selection and use during the early first-feeding period. However, mortality can be the result of several factors including inferior water conditions and improper hatching conditions. I found old acidic water (water that is less frequently changed) to be detrimental to their long-term health. Good aquarium conditions with regular partial water changes are the most important requirement for successful maintenance and breeding.

In their natural environment spawning usually occurs during the warmer period of the year (Spring-Summer) when water temperatures are around 24-32°C. Spawning in captivity can be attempted in a number of ways. They can be placed in a specially set up breeding aquarium with a one male; two females combination. A bunch of aquatic moss or spawning mop is placed in the tank, on which the fish will readily spawn. The spawning medium, with attached eggs, can then be removed each day, and place in a special hatching container. Alternatively, in a permanent aquarium environment purposely set up for breeding *I. werneri*, a self-sustaining population can be maintained. In fact, this is probably the most reliable method of breeding for the general hobbyist. Such an aquarium need not be larger than 50 litres, although a 90x45x45 cm aquarium tastefully decorated with river stones,





driftwood overgrown with aquatic mosses and some floating duckweed or water sprite can be very attractive. An aquarium designed accordingly and containing a school of displaying *Iriatherina werneri*, is a sight not easily forgotten.

Iriatherina werneri will normally eat their own larvae (fry) if they are given the opportunity, but what generally will happen is that in a well-planted aquarium some of the fry will survive because the adults cannot get to them. Once they get to a certain size you will see them coming out of the aquatic moss and start swimming with the adults. They (fry) know when they can safely come out. What happens is you end up with a lot of fry at different growth stages. This is also good because as the fry grow they need larger food and seeing that you are feeding the adults you don't need to know when you need to feed them larger food as the food is already there. In a permanently established aquarium some of the fry will find sufficient edible particles or infusorians to survive. You will often see newly hatched larvae clinging to and obviously feeding from algae and other material growing on the sides of the aquarium.

I found that any of the aquatic mosses or similar plants provides a very fine cover and plenty of it. Normal aquatic plants do not always provide the hiding places. You need the "fern or moss" type plants. Aquatic mosses are also good where you don't have overhead lights as they seem to grow without bright lighting. After a while the moss usually takes over the whole tank and you have to start removing some of it. A floating type plant that has a lot of fine hairy roots would probable also be suitable. The fry are very small and remain at the water surface so the aquatic mosses need to be close to the surface of the water. In a permanently established aquarium some of the fry will find sufficient edible particles or infusorians to survive. You will often see the newly hatched larvae clinging to and obviously feeding from algae and other material on the sides of the aquarium.

In an aquarium when they are maintained in a small group the males will regularly display to each other and to females. The male appears to prefer a site under a floating plant where he extends his first dorsal fin like a sail, the second dorsal and anal fins are then "flicked" in rapid movement up and down, enticing the female to his side. Incidentally, Pseudomugil gertrudae from the Jardine River also exhibits this flicking motion of the fins, whereas the east coast populations displays in similar fashion as rainbowfishes (fins held erect). The Jardine species of *Pseudomugil gertrudae* also have larger finnage than most of the east coast populations and exhibit different colours. During this procedure the colour of the body and fins intensifies. If the female is responsive, she will look for a suitable site for egg laying. Once the female remains in the same spot, the male will follow and together they will swim among the spawning medium and leave behind a small cluster of eggs. Spawning will often continue throughout the day. Females produce a small number of eggs each day for several days. Eggs adhere to fine-leaved plants or among the roots of floating vegetation. Any of the aquatic mosses are a suitable spawning medium for egg laying. Depending on water temperature, eggs will hatch in 7 to 10 days.

The requirement for successfully raising Iriatherina werneri is the implementation of a suitable feeding regimen for the larvae. The major difficulty for the aquarist is providing organisms appropriate to the size of the larvae at the first feeding stage and then supplying the large numbers of feed organisms necessary to maintain them. The preferred size of prey for larval fish increases as mouth size and feeding competency increase and different types of live foods have to be cultured for the different stages in the larval development. For example, different species of microalgae (phytoplankton) range from 2 to 20 µm; rotifers from 50 to 200 µm, and brine shrimp nauplii 400 to 800 µm. Apart from these main groups, a few other live feeds are used on a more limited scale including microworm (Panagrellus redivivus), vinegar eels (Turbatrix aceti), Moina and Daphnia spp. Brine shrimp nauplii are used primarily because brineshrimp eggs are easy to acquire and hatch.

Apart from live foods you can also successfully raise the larvae using one or more of the commercially available fry foods. I have found OSI 'Micro-Food' an excellent first food for *Iriatherina werneri* larvae. Just use it dry and sprinkle it over the surface of the water. Other products such as 'Sera Micron' are also suitable as a first food. As the larvae grow it is important to increase the size of the food until they are large enough to take brine shrimp nauplii and microworm. When weaning fish to a new food, introduce 10% of the new food daily while reducing the same percentage of the initial food until 100% of the new food is accepted. Commence feeding adult foods as soon as the juveniles are big enough to eat it and feed them often (at least twice daily). However, care should be taken not to overfeed.

The growth rate of the Iriatherina werneri is generally slow, with little variation until around 14 days. After that time growth rates increased. Feeding live foods generally results in higher growth rates. At 14-21 days post hatching the fry should be around 14-15 mm in size. It should be noted that the hatching of eggs might vary, resulting in the presence of larvae at different stages of development. As the larvae increased in age, the variation in length between individuals also increased. If you wish to raise an entire spawning, you may have to sort the growing fish by size, as the larger ones will eat their smaller siblings or repress their growth rate. If you have a batch that differs greatly in size, you will often find that the smaller ones are females. Size grading separates the faster and slower growing fish. When these smaller fish are transferred to another tank, their growth rate is no longer negatively impacted by the faster growing individuals. They should increase their growth rates to compensate for the initial retarded growth rates that developed during the nursery phase.

The continued growth and development of the fish will vary from one hobbyist to another and is largely conditional upon aquarium conditions such as temperature, water quality, and feeding regime.





# Kiunga ballochi Allen, 1983

Kiunga Blue Eye

# Species Summary

*Kiunga ballochi* have a mainly transparent body with a silvery coloured stomach and opercula. The scales are thinly outlined with pepper-like melanophores. The midlateral line, ventral edge of caudal peduncle, and bases of dorsal, caudal, and anal fins have dense concentrations of melanophores. The dorsal fin spines are translucent yellow; the second dorsal, caudal, and anal fins have bold black borders and yellow submarginal bands. The remaining portions of these fins are transparent except the dorsal and anal fins that have narrow strips of yellow basally and the anterior half of the anal fin has mainly yellow membranes. The pectoral fins are transparent. All fins with soft rays are faintly outlined with black. They have a moderately deep body for a blue-eye. Maximum size to about 3 cm SL.

# **Distribution & Habitat**

*Kiunga ballochi* were initially collected by Gerald Allen and John Paska in 1982 from small tributaries of the Ok Smak River, about 40 kilometres north of Kiunga on Tabubil Road, Papua New Guinea. They were found in several small shallow tributary streams flowing through dense rainforest with occasional sunlit clearings. A temperature of  $24-25^{\circ}$  Celsius and a *p*H of 7.8 were recorded at the collecting site. However, the full extent of their distribution range is unknown.

# Remarks

*Kiunga ballochi* was named in honour of Dr. David Balloch, a biologist with the Ok Tedi Mining Company. A few live specimens were brought back to Australia by Barry Crockford, but they were all destroyed in a tragic house fire in February 1983. Heiko Bleher visited this area in 1993 and despite sampling 37 streams along the entire stretch of road; he was unable to find any specimens. In 2007 Mark Allen and Philip Atio collected this species in streams along the Tabubil-Kiunga Road. However, they are not currently available in the hobby.

# Kiunga bleheri

Allen, 2004 Bleher's Blue Eye

# **Species Summary**

Gerald Allen described this species in 2004. On first glance their general appearance looks similar to *Kiunga ballochi*. However, the morphological differences, nevertheless, are also obvious. It differs from *Kiunga ballochi*, also from the Kiunga area, on the basis of its much shorter second dorsal and anal fins, significant modal difference in the number of second dorsal fin rays, 6 versus 5 transverse scale rows on the body, and in usually having most of the second dorsal and anal fin rays unbranched. Body colour is general semitransparent with the second dorsal fin faintly yellowish on outer portion with irregular black margin. Maximum size about 2.8 cm.

# **Distribution & Habitat**

Specimens of *Kiunga bleheri* have been collected in clear, shallow rainforest streams along the Kiunga-Konkonda Road, approximately 12 km west of Kiunga, Papua New Guinea. However, the full extent of their distribution range is unknown.

*Kiunga bleheri* were reportedly first collected by Heiko Bleher in 1991 from Tare Creek. Unfortunately on that occasion the live specimens were lost by the airline. On a second attempt in 1993 he caught just three individuals and they didn't breed. Then in 2003 he managed to catch a larger number in the same stream and these were successfully transported to Europe and eventually released into the aquarium hobby. Additional specimens were collected in 2003 by Heiko Bleher. Water parameters recorded in 2003 were: *p*H 5.9; conductivity 29  $\mu$ S/cm; temperature 27°C.

Live specimens of this species were collected by Charles Nishihira in 1994 and released into the aquarium hobby. In 2007 Mark Allen and Philip Atio also collected this species. However, today there are no live specimens in captivity.





Melanotaenia affinis (Weber, 1908)

New Guinea Rainbowfish

*Rhombatractus affinis* Weber, 1908 *Rhombosoma sepikensis* Herre, 1935 *Rhombosoma affinis* Whitley, 1938 *Melanotaenia affinis* Allen, 1980

# **Species Summary**

Melanotaenia affinis are an attractive species that are found only in the northern regions of New Guinea. As with many rainbowfishes, their colouration is variable depending on location and water conditions. They generally have a body colouration of olive to bronzy-yellow dorsally grading to white ventrally; a blue to blackish stripe from the snout to caudal fin base, often faint or absent in the pectoral fin region and becoming more intense and broader on the caudal peduncle where it is bordered by orange or reddish stripes (at least in males). The sides often have a series of narrow orange stripes between each scale row; fins whitish-translucent to yellowish; anal fin and frequently soft dorsal fin of mature males bright yellow-orange. Maximum size about 12-15 cm with a body depth of around 3–4 cm. Males are deeper bodied than females and have pointed posterior tips of the dorsal and anal fins. These features become more obvious with increased growth. The overall colour pattern of males is more intense, particularly the orange stripes and dark mid-lateral stripe. The vertical fins of females are either translucent or only faintly yellow compared to the bright yellow-orange of males.

This species often exhibits geographic colour pattern variation. Generally this is related to the amount or intensity of orange or reddish striping on the sides, width and intensity of the dark mid-lateral stripe and intensity of yellow or orange colouration in the vertical fins. There may also be morphological variations, for example in relation to body depth and shape of the snout. Two populations in particular are noteworthy because of these features; one from the highland tributaries in the vicinity of Baiyer River and the other from lowland tributaries near Pagwi. The Baiyer River population is characterised by a relatively slender body and tends to have a higher soft dorsal ray count than specimens from other localities (17-20 vs. 13-16). The fish from streams near Pagwi differs by having a more pointed snout and a mid-lateral stripe that is more uniform in width (i.e., not broadly expanded posteriorly) and bordered by brilliant reddish stripes.

Males and females generally mature before the end of their first year or at a standard length of 5–6 cm. *M. affinis* are carnivorous, feeding on a variety of small invertebrates taken from mid-water or from the surface. Gut content included crustaceans, insect larvae, and terrestrial insects such as ants and small beetles.





# **Distribution & Habitat**

*Melanotaenia affinis* are widely distributed in northern New Guinea (north of the central dividing range). The range extends from the Oomsis River near Lae, Papua New Guinea westward into West Papua to at least the vicinity of Nabira. It is the most common rainbowfish in tributary streams of the Markham, Ramu and Sepik Rivers of Papua New Guinea. Found in some mountainous headwater streams of the Sepik in the Western Highlands, such as in the Baiyer River, and also recorded from the Taritatu River (Mamberamo system) in West Papua.

They are found most frequently in rainforest streams, in water temperatures between  $18-28^{\circ}$  Celsius. They are mainly found around sub-surface vegetation, submerged logs, or branches in small tributary streams but can also occur in lakes, swamps, and lagoons often together with *Chilatherina* and *Glossolepis* species. Their natural environment is subjected to seasonal variations with water temperature, pH, and hardness levels varying considerably. They are usually found in clear water, but sometimes in turbid conditions.

# Remarks

*Melanotaenia affinis* were one of the earlier New Guinea rainbowfishes to be introduced to the aquarium hobby. They first appeared in the Australian hobby around 1959. Further live specimens were collected near Lae by Barry Crockford and Gerald Allen in the late 1970's and subsequently reintroduced to the hobby in Australia. Three main varieties have been maintained in the hobby. The 'standard' coloured variety is widespread in a number of locations and is indistinguishable in colour pattern. Live specimens of this variety were collected and transported back to Australia by Barry Crockford in the late 1970's.

The 'Pagwi' variety, known only from small tributaries of the Sepik River near Pagwi Village have an olive-greenish upper body colour and white below. The mid-lateral line is blue, prominent and has bright red-orange upper and lower margins on the rear half of the body. There is also a broad pale yellow anterior scale row just below the mid-lateral band. Live specimens of this variety were collected and transported back to Australia by David Coates and Gerry Allen in 1982.

Another, the 'Bluewater Creek' variety was collected from a stream near Madang, Papua New Guinea by Gerald Allen in 1978. The upper body is greenish-blue with silvery reflections and the lower body white. The mid-lateral band is blue-black having a broad white margin on the lower edge, which is separated from the lower side by a diffused bluish-black stripe. Live specimens of this variety were collected by Heiko Bleher in 1988 and taken back to Europe.







## Melanotaenia ajamaruensis

Allen & Cross, 1980 Ajamaru Rainbowfish

## **Species Summary**

From October 1954 through to May 1955 Marinus Boeseman took part in a collecting expedition for the Rijksmuseum van Natuurlijke Historie to Netherlands New Guinea (West Papua). Among the places he visited was Lake Sentani, Tami River, Biak Island, Lake Jamoer (Yamur), Wissel Lakes, Ajamaru Lakes, Lake Ajtinjo, Merauke and the Digul River. This collection included many rainbowfishes, but a thorough study of the collection or description of the fishes was never made by Boeseman.

As part of his preparation for the revision of the rainbowfish family, Gerald Allen studied the Boeseman collection of 1954–55 during 1975 and 1977. He discovered no less than four new rainbowfish species, which he described in 1980 together with Norbert Cross. These species were *Melanotaenia boesemani*, *M. ajamaruensis*, *M. japenensis* and *Glossolepis pseudoincisus*.

From field notes it was stated that *Melanotaenia ajamaruensis* have a metallic blue to yellowish or green with orange and yellow longitudinal stripes and dark scale edges. The body is

ovate and laterally compressed. Mature males have a higher first dorsal fin, which overlaps the origin of the second dorsal fin when depressed. They grow to a length of around 11 cm; males are usually deeper bodied than females. They were named '*ajamaruensis*' with reference to the Ajamaru Lakes, the type locality and only known collection site for this species at that time (see remarks).

In 1980 Allen & Cross described *M. ajamaruensis* as a species of *Melanotaenia* with the following combination of characters: dorsal rays IV to VI, 15 to 19; anal rays I, 21 to 27; pectoral rays 13 to 15; horizontal scale rows 7 or 8; vertical scale rows 34 to 37; predorsal scales 13 to 16 preopercle scales 9 to 16; colour generally reddish-brown on back and anterior half of body grading to yellow or tan posteriorly with series of red-brown horizontal stripes on side; in life ground colour metallic blue to yellowish or green with yellow longitudinal stripes.

*M. boesemani* is readily separable from *M. ajamaruensis* on the basis of soft ray counts for the second dorsal and anal fins. The former species has 10 to 14 (usually 12 or 13) dorsal rays and 17 to 23 (usually 18 to 21) anal rays compared with 15 to 19 (usually 15 to 17) and 21 to 27 (usually 22 to 24) for *M. ajamaruensis*. Although these species possess a similar colouration and general shape, the stripes on the sides tend to be more pronounced in *M*.



*ajamaruensis*, particularly the mid-lateral one and the stripe just below it. *M. ajamaruensis* further differs from *M. boesemani* by being more slender, and by having the first dorsal fin origin in front (by about one half eye diameter) of the anal fin origin compared to the approximately even position of these fins in *M. boesemani*.

### **Distribution & Habitat**

*M. ajamaruensis* is a lake and stream dwelling rainbowfish found in relatively clear alkaline water, with abundant aquatic vegetation. Museum specimens were collected in March 1955 by Marinus Boeseman and his companions in the Ajamaru Lakes, a complex of lakes on the Ajamaru River in the centre of the Vogelkop Peninsula at the western extremity of New Guinea. The Ajamaru Lakes region is located about 120 km east-southeast of Sorong, at the headwaters of the Ajamaru River in a mountainous region of the Vogelkop Peninsula, West Papua. The region contains a number of small freshwater lakes and associated marshes. The largest lake, Lake Ajamaru drains east via the other two lakes (Lake Hain and Lake Ajtinjo) into an upper tributary of the Kais River that eventually flows into the Ceram Sea to the south.

Two species of rainbowfishes have been reported from these lakes: *Melanotaenia ajamaruensis* and *Melanotaenia boesemani*. It is possible that *M. ajamaruensis* inhabit other areas on the Vogelkop Peninsula, but most of the region remains unsampled.

Lake Ajamaru has an area of approximately 22 km<sup>2</sup> and is located in a rather flat terrain, at about 250 metres altitude. The lake has variable depths with clear water and abundant vegetation. In the wetter months (April-June) the lake can rise by up to 5 metres from its dry season level; it never dries out completely, but the shoreline recedes several hundred metres. It has a muddy bottom, and the sediments of the shores are reportedly white, either sand or kaolin clay. The lakes and streams have a pH of 6.4–7.8 (de Vries, 1962) and temperate 26–27° Celsius. Heiko Bleher reported the water conditions as pH 9.0, hardness 5° dGH, and conductivity 145 mS/cm. When Marinus Boeseman collected his specimens, he reported a pH of 6.4–6.5.

## Remarks

In November 1982, Gerry Allen had the opportunity to collect live specimens during a visit to the remote Vogelkop Peninsula in West Papua. Heiko Bleher, a well-known aquarium fish collector, had accompanied Gerry Allen on the trip and was able to transport a number of live specimens captured during the expedition back to Europe, whereupon they were subsequently bred and distributed in the aquarium hobby as *M. boesemani*. At the time it was thought that females of *M. boesemani* were *M. ajamaruensis*.

The natural colours of *M. ajamaruensis* at the time remained unknown. The type specimens preserved in the Leiden museum were the only ones that had so far been collected.

In 2007, a number of surveys were conducted by the Papuan National Marine and Fisheries Research, the Academy of Fishery Sorong, and the Institut de recherche pour le développement (IRD) Jakarta in five bioregions of West Papua. During the collecting trip to the Sorong region they collected approximately 352 rainbowfishes, and among them were a number of specimens that were considered to fit the type specimens of *M. ajamaruensis* as described by Allen & Cross in 1980.

They were collected in an upstream section of a small river near Lake Ajamaru. This river is an old outlet to the western side of the lake, but does not flow into the lake. Instead it flows into a subterraneous zone. There is no connection between the lake and the river and *M. boesemani* wasn't found to occur in the river. This is currently the only known location of *M. ajamaruensis* today and it is thought that they no longer exist in Lake Ajamaru. The colouration of *M. ajamaruensis* can be much more intense than that shown in the accompanying photograph; displaying a brilliant red to red-orange colour (L. Pouyaud 2009, *pers. comm.*).

*Melanotaenia ajamaruensis* are currently not available in the aquarium hobby.

*Note*: The interesting thing about *M. ajamaruensis* is their remarkable similarity with *M. boesemani*. Since its introduction to the aquarium hobby, *M. boesemani* has steadily increased in popularity and today, it could be considered the most popular rainbowfish in the hobby. By 1989 Ajamaru villagers were catching so many live fish for the aquarium trade the species was on the brink of becoming endangered. An estimated 60,000 male rainbows were captured each month for shipment to Jakarta exporters. Eventually the Indonesian Government placed some controls on the activity (*Polhemus et al. 2004*).

Is it possible that specimens of *M. ajamaruensis* may have been mixed in with some of the early collections of *M. boesemani* from Lake Ajamaru—or perhaps *M. ajamaruensis* never occurred in Lake Ajamaru in the first place?





## Melanotaenia ammeri

Allen, Unmack and Hadiaty, 2008 Ammer's Rainbowfish

## **Species Summary**

The male colouration of Melanotaenia ammeri is generally bluish on the upper body with a series of alternating mauve to blue-grey and pale yellow stripes corresponding with each of the horizontal scale rows on the side of the body. The midlateral stripe, at the level of the upper pectoral-fin base is usually much darker than the others. The blue stripes below become progressively more inconspicuous and forming interrupted dotted lines. A broad, horizontally bluish streak immediately above the abdomen. The upper portion of the head is blue or greyish, while the lower half is silvery white. The dorsal, anal, pelvic and caudal fins are pale yellow. The pectoral fins translucent with a white base. The colour pattern of the females are similar to that of the males except the blue and yellow hues of the body stripes are generally less vivid and the median fins are mainly translucent, only slightly yellow. The distinctive pattern of alternating mauve to blue-grey and yellow stripes is unique among species of Melanotaenia.

Males are generally deeper bodied than females and have a more elongate, pointed shape posteriorly on the soft dorsal fin. The longest soft dorsal-fin rays of males are located in the posterior-most portion of the fin, in contrast to that of females, which are situated in the anterior half of fin. In addition, the depressed first dorsal fin of adult males extends to the base of the second or third soft ray of the second dorsal fin, compared with the spine of first ray in females. This species was named "*ammeri*" in honour of Max Ammer of Sorong, West Papua.

## **Distribution & Habitat**

Melanotaenia ammeri is currently known only from the type locality; a small creek flowing into the northern part of Arguni Bay near Gusimawa Village. It no doubt occurs in nearby streams, but the exact limits of distribution remain to be determined. The type locality consisted of a narrow (2–3 metre wide), relatively shallow (to about 0.5 metre) stream with gradual gradients flowing through second growth forest, about one kilometre upstream from the sea. The type specimens were collected over sand and gravel bottoms with substantial leaf litter and dead tree branches.

## Remarks

*Melanotaenia ammeri* was collected from a small creek near Gusimawa Arguni Bay, West Papua by G. R. Allen and M. Ammer in 2008. This species is not currently available in the aquarium hobby.





## Melanotaenia angfa

Allen, 1990 ANGFA Rainbowfish

#### **Species Summary**

*Melanotaenia angfa* are bright yellow (including fins) with a narrow midlateral band, which is interrupted along its length by narrow yellow streaks. They grow to a length of around 13-cm, males are usually deeper bodied than females.

#### **Distribution & Habitat**

*Melanotaenia angfa* have only been collected from two small creeks in the Yakati River system of West Papua. The Yakati River is located in the narrow isthmus connecting the Vogelkop Peninsula and the remainder of West Papua. Both streams are characterised by slow to rapid-flowing water with rock or sand bottom and minimal aquatic vegetation.

The region contains a number of river systems that empty into Bintuni Bay and include the Wasian, Muturi, Bokor, Tirasai, Sumber, Kodai, Rarjoi, Kamisayo, Tatawori, Sorobaba, Yakati, Yensei, Sobrawara and Naramasa Rivers. Most of these rivers are fast-flowing during the wet season, particularly in the upper part of the rivers, sometimes causing flooding. During the wet season the colour of the water turns brown from soil erosion. Some rivers always have flowing water, while others are dry and only flow during the wet season.

#### Remarks

This species was discovered by Gerald Allen in 1989 during a conversation survey of Bintuni Bay and named '*angfa*' in honour of the Australia New Guinea Fishes Association. Live specimens were collected by Heiko Bleher in 1999 and have been distributed in the aquarium hobby.





### Melanotaenia arfakensis Allen, 1990 Arfak Rainbowfish

#### Species Summary

Melanotaenia arfakensis have a basic body colour that is mauve with silvery reflections. There is a bluish midlateral band, about one scale wide, and a narrow yellow-orange stripe between each horizontal scale row on the sides of the body. Fins are translucent with a bluish to mauve shading. The second dorsal fin has a sub-marginal band and narrow white margins. The caudal fin has black upper and lower margins. Males may reach a maximum size of 10 cm, but females are usually less than 8 cm. Males are more brightly coloured, larger, and deeper bodied than females.

#### **Distribution & Habitat**

Melanotaenia arfakensis were originally collected by Gerald Allen in 1989 from the Prafi River, about 20-30 kilometres west of Manokwari, West Papua. This area is bordered on the western side by the Arfak Mountains. The mountains are a water catchment area for several river systems. The Arfak area is important historically and scientifically for being the site of the first extensive zoological expedition to West Papua led by d'Albertis and Beccari in 1872-73.



Melanotaenia arfakensis is a stream dwelling rainbowfish mainly found around sub-surface vegetation, submerged logs, or branches in small tributary streams, but can also occur in swamps and lagoons. Their natural environment is subjected to seasonal variations.

#### Remarks

Live specimens of Melanotaenia arfakensis were collected by Heiko Bleher in 1990 and introduced to the aquarium hobby.

In 2007 surveys were conducted by the Papuan National Marine and Fisheries Research, the Academy of Fishery Sorong, and the Institute of Research for Development of France in the Arfak Mountains (Manokwari) region in West Papua. M. arfakensis were collected from the Prafi River. Additional rainbowfishes were collected from the Ati (Ani) River that were very different from M. arfakensis. Also in the Kebar River, another different rainbowfish species than those found in the Ati River were collected. This species was also believed to be a new species and not M. arfakensis. However, none were kept alive for breeding purpose.



## Melanotaenia australis (Castelnau, 1875)

Western Rainbowfish

Neoatherina australis Castelnau, 1875 Melanotaenia solata Taylor, 1964 Nematocentris australis Allen, 1975 Melanotaenia splendida australis Allen, 1980 Melanotaenia australis Allen, Midgley & Allen, 2002

## **Species Summary**

*Melanotaenia australis* was first described by Castelnau as *Neoatherina australis* in 1875. The original type specimens were collected from Weeli Wolli Creek, Hammersley Range, and Millstream homestead in Western Australia. In earlier days they were commonly known as the 'Westralian Sunfish'. In 1964 another species of rainbowfish collected from the Northern Territory was named *Melanotaenia solata*. After Gerald Allen's review of the rainbowfish family in 1980, these two species were considered as one and he placed them in the large "*splendida*" group as a sub-species, and named them *Melanotaenia splendida australis*, but genetic studies indicate they are clearly a distinct species.

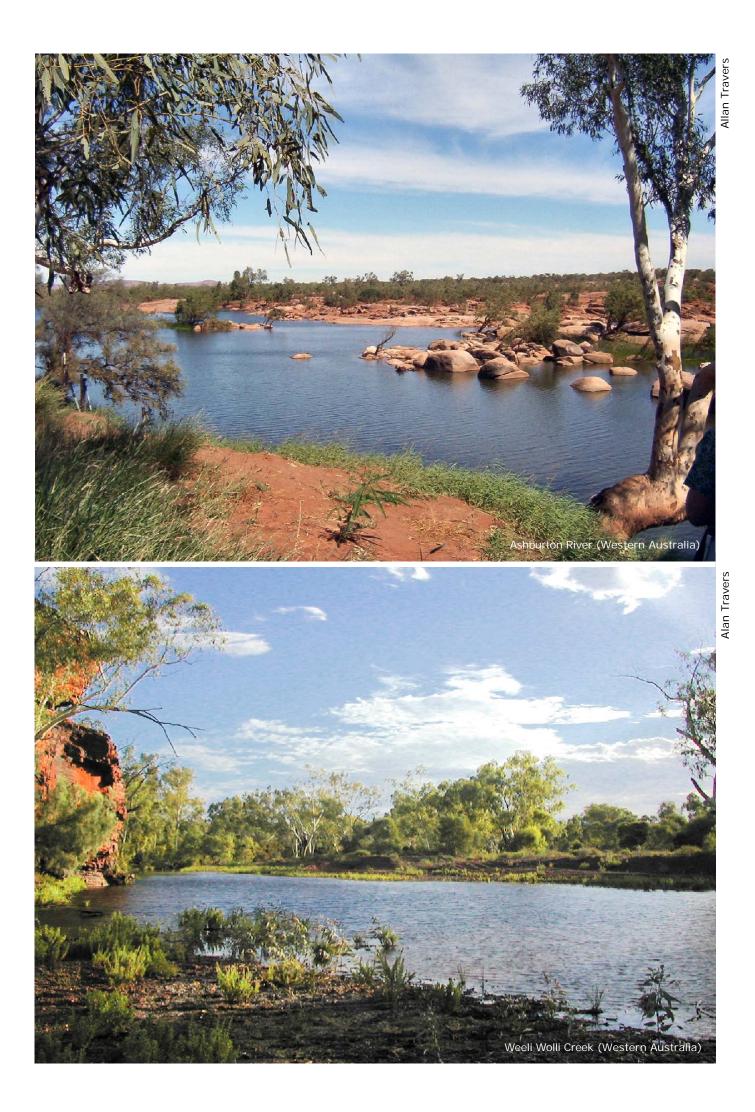
*Melanotaenia australis* can grow to a length of around 10 cm, but are more common at 8 cm or less. Males are usually much larger and deeper bodied than females. Their colouration,

particularly the fins and intensity of the mid-lateral stripe, is highly variable depending on their geographic locality. This is mainly due to a rapid speciation in the rainbowfish group and is further enhanced by the degree of random morphological variation occurring among the various populations. Colouration generally consists of 1-2 broad, dark mid-lateral stripes and a series of narrow reddish stripes corresponding with each scale row. Fins range from nearly colourless to deep red, or clear with red or green flecks. Gerry Allen notes that the real trademark of this species is the zigzagging blackish lines on the lower sides, just above the anal fins. The black mid-lateral line may be very prominent or scarcely apparent. Males are easily distinguished from females by their brighter colours and longer and more elongated dorsal fin rays. Fish from the Drysdale River of Western Australia are generally smaller in statue and possess a distinctive double mid-lateral black stripe.

## **Distribution & Habitat**

*Melanotaenia australis* have a restricted disjunct distribution in northern Western Australia and the Northern Territory. They are widespread throughout the Pilbara region of Western Australia between the Ashburton and DeGrey Rivers and in the Kimberley region in the extreme northern part of Western Australia between the Fitzroy River and the Northern Territory border. They also occur in streams of the north-western sector in the Timor Sea drainage of the Northern Territory, just east of Darwin.







*Melanotaenia australis* is a stream dwelling rainbowfish mainly found around sub-surface vegetation, submerged logs, or branches in small tributary streams, but can also occur in swamps and lagoons. They are most commonly found in backwaters or along the shoreline where there is minimal flow and grassy vegetation. Their natural environment is subjected to seasonal variations with water temperature 22–28°C, *p*H 6.5–8.0, and hardness levels varying considerably. There is often a large fluctuation in water conditions between the dry and wet seasons.

#### Remarks

*M. australis* show considerable variation in morphology across its known distribution and genetic studies suggest that there maybe more than one species in the "*australis*" complex. Genetic studies in 2000 (McGuigan, *et al.*) found that *M. australis* is represented by two distinct lineages. These lineages are geographically restricted to Western Australia and the Northern Territory. The difference between the two groups could indicate the presence of two distinct species. If this is true, then the name *Melanotaenia solata* (Taylor 1964) could be applied to the Northern Territory populations following redescription. The study suggested that irrespective of the situation in the Northern Territory, West Australian populations of *M. australis* should be accorded species status. The research did not support the inclusion of either group within *Melanotaenia splendida*.

Allen (1980) considered that *Melanotaenia solata* (Taylor 1964) fell within the range of *Melanotaenia splendida australis* with regard

to colour pattern, morphometrics and meristics. Melanotaenia solata have been reported from several localities east of Darwin, including the upper South Alligator River and Yirrkala, Groote Eylandt, and Bickerton Island. William R. Taylor described them as a species of Melanotaenia with a rather slender, compressed body; with complete dentition, with poorest developed in upper jaw; with a rather faint brown lateral body band and with numerous, characteristic, golden yellow life stripes through brown band as well as along the body. Large adults with diffuse dark band and about ten brilliant golden longitudinal stripes on each side; basal half of caudal fin bright yellow; bronze bar behind eye crossing preopercle and diffusing downward on opercle; belly and bases of second dorsal and anal fin pinkish; the inter-radial membranes paler outward; inter-radial membranes of first dorsal fin red. In specimens from Yirrkala, the dorsal and anal fins red; caudal fin yellowish orange; pelvic fins deep red; lower side bluish silvery; scale centres on side generally golden bronze; they form about five longitudinal rows, the lowermost of which is approximately on a level with the ventral surface of the caudal peduncle.

More recent studies (Phillips, 2004) have shown that there is also a very large genetic difference between the Pilbara and east Kimberley populations of *M. australis*. These studies provide support for the separation of *M. australis* into two species: a southern one from the Pilbara area and the remaining northern populations. Pilbara populations are quite distinct and Allen (1995) noted that Pilbara populations lacked the mid lateral band that is prominent in many other *M. australis* populations.









# Melanotaenia batanta

Allen and Renyaan, 1998 Batanta Rainbowfish

### **Species Summary**

Melanotaenia batanta was described from 12 specimens collected at Batanta Island in 1998. They have an overall blue body colouration with darker scale margins. Lower half of body whitish to silvery with a triangular grey area on the lower side, above the pelvic fins. Narrow orange stripes are visible between each horizontal scale row on the upper two-thirds of side. A dark blotch consisting of a concentration of melanophores immediately behind eye on uppermost part of operculum. Fins mainly translucent except dorsal, anal, and caudal, frequently with bluish tint. There is little difference between male and female, an unusual feature for rainbowfishes. Unlike most Melanotaenia there is a lack of pronounced sexual dimorphism. Males, in particular, lack the pronounced elongation of the posterior dorsal and anal fin rays, but rather the anterior or middle rays tend to be longest. Moreover, they have poorly developed palatine teeth or lack them entirely. Males may reach a maximum size of 10 cm, but females are usually less than 8 cm.

*M. batanta* is most closely related to *M. fredericki*, but there is a modal difference in number of dorsal fin rays and it possesses fewer cheek scales (11–13 *vs.* 17–20).



#### **Distribution & Habitat**

Known only from Batanta Island in the Raja Ampat Islands lying immediately west of the Vogelkop Peninsula, West Papua. Batanta Island is home to three separate species of rainbowfishes. *M. batanta* were collected from Warmon Creek, on the northern side of the island. The rainbowfish was restricted to a very small portion of this stream, essentially a 400 metre stretch bounded by brackish mangrove habitat and a 10 metre high waterfall upstream.

#### Remarks

This species was named batanta, with reference to the type locality. No live specimens have so far been collected for the aquarium hobby. This species should not be confused with another rainbowfish distributed in the hobby as the "Batanta Island Rainbowfish" (*Melanotaenia synergos*).





## Melanotaenia boesemani

Allen and Cross, 1980 Boeseman's Rainbowfish

#### **Species Summary**

The colour pattern of male *Melanotaenia boesemani* is completely different from most other rainbowfishes and show a half-and-half colouration when fully matured. The head and front portion of the body are a brilliant bluish-grey, sometimes almost blackish, with the fins and posterior half of the body largely bright orange-red. Between these two areas, or roughly just behind the pectoral fin, there are alternating light and dark vertical bars. Their wild colouration can fade somewhat in captivity, possibly due to something lacking in the diet, or from the nature of captivity itself. They may reach a maximum size of 12 cm, but are usually less than 10 cm.

Males are easily distinguished from females by their different colour and longer and more elongated dorsal fin rays, and are usually much deeper bodied than females. Females display a broad dark mid-lateral stripe accompanied by a series of narrow yellow or reddish-orange longitudinal stripes corresponding with each scale row that deepen or lighten according to mood. Mature, older females often show colouration similar to subordinate males, but are usually easily identified by a shallower body/chest depth and smaller, more rounded fin edges.

#### **Distribution & Habitat**

*Melanotaenia boesemani* have been found in Lake Ajamaru and a few surrounding tributaries. They also occurs in Lake Ajtinjo, Lake Hain and Lake Uter. The lakes are located about 120 km east-southeast of Sorong, at the headwaters of the Ajamaru River in a mountainous region of the Vogelkop Peninsula, West Papua. The largest lake, Lake Ajamaru drains east via the other lakes into an upper tributary of the Kais River that eventually flows into the Ceram Sea to the south. The lakes are positioned centrally on the Ajamaru Plateau which extends for 20–30 km to the south and south-west of the lakes before giving way to a broad zone of relict alluvial landforms dissected by wide flooded river valleys.

Lake Ajamaru has an area of approximately 22 km<sup>2</sup> and is located in a rather flat terrain, at about 250 metres altitude. The lake has variable depths with clear water and abundant vegetation. In the wetter months (April-June) the lake can rise by up to 5 metres from its dry season level; it never dries out completely, but the shoreline recedes several hundred metres. It has a muddy bottom, and the sediments of the shores are reportedly white, either sand or kaolin clay. The lakes and streams have a *p*H of 6.4–7.8 (de Vries, 1962) and temperate 26–27° Celsius. Heiko Bleher reported the water conditions as *p*H 9.0, hardness 5° dGH, and conductivity 145 mS/cm. When Marinus Boeseman collected his specimens, he reported a *p*H of 6.4–6.5.





In August 1959, G. A. Reeskamp surveyed the lakes with the objective of determining the potential fisheries of the lakes. He reported that the lakes were shallow and interconnected by channels that might perhaps be better termed as "broads". The three lakes average approximately 7 feet. (2.13m) in depth and drain in an easterly direction into the Kais River. During the rainy season the water level rises to approximately 9 feet. (2.74m) and at the dry season large areas of these broads become dry. The greatest depth was found close to the southern margin of the lakes where a basin about 60 feet. (18.28m) diameter was discovered with a depth of approximately 20 feet. (6.09m). The outstanding characteristic of the lakes was the clearness of the water. Owing to the clarity of the water there is complete light penetration to the bottom with the resultant abundant bottom flora of aquatic plants. Samples of the waterplants were stiff to the touch, indicating a high lime content. The pH of the water was recorded as 7.8. Fish in these lakes appeared to be extremely scarce in relation to the large area of available water. In the shallow creeks along the margins, however, one obtains an impression of the fairly rich fauna but in the open water few fish may be seen and in general the fish appear to remain in the shallow margins of the lakes where food such as water insects, snails, fish fry, etc., are more plentiful.

The Ajamaru lakes only support a small number of fishes and most of these are of very small size and diversity. The Dutch introduced some larger fish species, such as *Cyprinus carpio* and labyrinth fishes into the lakes in the mid-1930s to provide new sources of animal protein. As early as 1938, *Trichogaster pectoralis*, *Helostoma temminckii* and *Cyprinus carpio* were introduced into Lake Ayamaru to supply the requirements of a Dutch military post in that area. The two first-mentioned species are still found there as a result of a highly successful acclimatisation. *Cyprinus carpio* was introduced to the lake in 1938, 1951 and 1969. *Gambusia (affinis)* was introduced in 1959 for malaria control.

Reeskamp reported that "the local natives benefited by the somewhat improved stocks of fish in the lake since the native species were apparently seriously depleted many year ago. Generally speaking, the methods of fishing are very primitive and there is considerable destruction of fish by poisons, locally known as "akar kajoe" or "akar boreh", derived from the Derris (*Derris elliptica*). This system of fish poisoning seems to be increasing and must no doubt have disastrous results on the existing stocks and will inhibit any development unless it can be fully prevented. Very large numbers of firy are killed by the poison and it is certainly in the interests of the natives themselves that this practice should be prohibited." Reeskamp also recommended that plant-eating fish should be introduced into the lakes to utilise the vast quantities of submerged aquatic vegetation.

*Melanotaenia ajamaruensis, Melanotaenia boesemani*, and *Pseudomugil reticulatus* have been reported from the lakes and surrounding streams.

The waterplant *Ceratophyllum demersum* has been recorded from the lake and *Eichhornia crassipes* was introduced in 1980s, but it covered only a small part of the lake. Formerly there were two species of submerged macrophyte (species not reported), but these disappeared or became very scarce after the introduction of *Cyprinus carpio*. As a result, one small species of fish local known as 'bobok' was reported to have become extinct because of the disappearance of its habitat (the submerged macrophytes). Heiko Bleher reported that the lake is almost filled with aquatic plants. Mainly *Vallisneria*, *Ceratophyllum* and *Najas* species.

Boeseman described Lake Ajtinjo as "... a widened river, flowing southeast, with a length of 4 km and strongly varying width with a maximum of about 350 m. At the north-western, end the principal river widens to become a lake which consists of two parts separated by considerable rapids and small cataracts; at the south-eastern end the lake abruptly stops, but a subterranean connection with the Kais River is supposed to exist here. The mountains at most places closely surround the lake which has steep and rocky shores, almost perpendicular at some places but elsewhere allowing some wider marshy banks. The water is clear, pH about 6.5, flowing rather strongly only at the narrower parts of the lake, including the upper reaches. The bottom is rocky, at most places covered with sand, stones or large rocks, but muddy at some places. Both the aquatic and terrestrial vegetation are dense, at least where the stony substratum allows growth."

### Remarks

*M. boesemani* were originally collected from Ajtinjo Lake by Sten Bergman during the Swedish New Guinea Expedition 1948-1949. Specimens are maintained in the Swedish Museum of Natural History. From October 1954 through to May 1955 Marinus Boeseman took part in a collecting expedition for the 'Rijksmuseum van Natuurlijke Historie' to Netherlands New Guinea (West Papua) with L.D. Brongersma and L.B. Holthuis. His task was to provide a thorough knowledge of the fish fauna by intensively surveying as many rivers and lakes as possible in western New Guinea.

This task was taken to heart and in a relatively short period many localities were visited, resulting in a rich collection for the museum in Leiden. Among the places he visited was Lake Sentani, Tami River, Biak Island, Lake Jamoer (Yamur), Wissel Lakes, Ajamaroe (Ajamaru) Lake, Lake Ajtinjo (Aytinjo), Merauke and the Digul River. This collection included many rainbowfishes, but a thorough study of this material or description of these species was never made by Boeseman.





As part of his preparation for the revision of the rainbowfish family, Gerald Allen studied the Dutch collection of 1954–55 during 1975 and 1977. He discovered no less than four new rainbowfish species, which he described in 1980 together with Norbert Cross. These species were *Melanotaenia boesemani, M. ajamaruensis, M. japenensis* and *Glossolepis pseudoincisus. M. boesemani* and *M. ajamaruensis* were collected in March 1955 by Boeseman and his companions in the Ajamaru Lakes, a complex of lakes on the Ajamaru River in the centre of the Vogelkop Peninsula. Specimens of *M. boesemani* was also found in Lake Ajtinjo, 25 kilometres to the southeast of Ajamaru village and from 'Djitmau', about 3 km south of the Ajamaru Lakes. The specimens preserved in alcohol still showed the unusual colour pattern.



*M. boesemani* is readily separable from *M. ajamaruensis* on the basis of soft ray counts for the second dorsal and anal fins. The former species has 10 to 14 (usually 12 or 13) dorsal rays and 17 to 23 (usually 18 to 21) anal rays compared with 15 to 19 (usually 15 to 17) and 21 to 27 (usually 22 to 24) for *M. ajamaruensis*. Although these species possess a similar colouration and general shape, the stripes on the sides tend to be more pronounced in *M. ajamaruensis*, particularly the midlateral one and the stripe just below it.

In November 1982, Gerry Allen had the opportunity to collect live specimens during a visit to the remote Vogelkop Peninsula. Heiko Bleher had accompanied him on the trip and was able to transport a number of live specimens captured during the expedition back to Europe, whereupon they were subsequently bred and distributed in the aquarium hobby. In 1998, Heiko Bleher collected more live specimens of *M. boesemani* from Ajtinjo Lake and they too, have been distributed in the aquarium hobby.

Since its introduction to the aquarium hobby, *M. boesemani* has steadily increased in popularity and today, it could be considered the most popular rainbowfish in the hobby. By 1989 Ajamaru villagers were catching so many live fish for the aquarium trade the species was on the brink of becoming endangered. An estimated 60,000 male rainbows were captured each month for shipment to Jakarta exporters. Eventually the Indonesian Government placed some controls on the activity (*Polhemus et al. 2004*).

#### **Other Notes**

In 2007 surveys were conducted by the Papuan National Marine and Fisheries Research, the Academy of Fishery Sorong, and the Institute of Research for Development of France in five regions of West Papua. Fifteen species of rainbowfishes were collected during these expeditions. During the collecting trip to the Sorong region they collected 352 rainbowfishes, and among them were a number of undescribed species. *M. boesemani* were collected from Lake Ajamaru, Lake Ajtinjo and Lake Uter.

Lake Uter was inhabited by thousands of *M. boesemani*. They also sighted Tilapia and Goldfish. Based on their observations, the specimens caught in the upstream Lake Uter were attacked by bacteria and fungi and were suffering from malnutrition. According to the local villagers the rainbowfishes started showing disease after the introduction of the goldfish and Tilapia a few years before. The *M. boesemani* specimens collected from Lake Ajamaru had bright yellow posterior, whereas specimens from Lake Uter had a body colour of skyblue and orange posterior.

*M. boesemani* was named in honour of Dr. Marinus Boeseman, the collector of the type specimens. According to labels accompanying the type specimens the native name for this species is 'sekiak' and 'ikan rascado'. Marinus Boeseman was born on June 22, 1916 in Enkhuizen, a small port on the Zuiderzee in Holland. After the untimely death of his father, Marinus, aged 11, his two elder sisters and his mother moved to Oegstgeest, a neighbour town of Leiden where he continued his primary and secondary education. In 1935 he entered Leiden University to study biology. On November 1, 1947 he was appointed curator of fishes at the Rijksmuseum van Natuurlijke Historie in Leiden, and held that position until his retirement on 30 June 1981. He died on July 14, 2006 at the age of 90







## Melanotaenia caerulea

Allen, 1996 Blue Rainbowfish

## Species Summary

Melanotaenia caerulea have a body colour of bright iridescent blue on the sides and back, becoming whitish or pinkish ventrally. There is a faint dark blue midlateral band on the posterior half of body, about one scale row wide. Each horizontal scale row on blue portion of body is separated by narrow pinkish-orange stripe. There is a short brown stripe about pupil width from the rear of the eye to the area just above the pectoral fin, frequently continuing as a pair of narrow brown stripes on the upper and lower edge of the midlateral band, and linking posteriorly with the dark blue midlateral band mentioned above. Fins are bluish to translucent, anterior edge of first dorsal fin and outer portions of second dorsal and anal fins sometimes reddish or dusky blackish in males. Pelvis fins mainly grey to reddish, but sometimes slightly dusky grey to reddish. Pectoral fins translucent. Males may reach a maximum size of 8 cm, but females are usually less than 6 cm. Males are generally deeper bodied and have more elongated, somewhat pointed shape posteriorly on the soft dorsal and anal fin rays. Females have smaller rounded dorsal and anal fins.

*Melanotaenia caerulea* belongs to the "*maccullochi*" group of rainbowfishes, and appears to be most closely related to *M. ogilbyi*. It differs from other members of the group in having a largely blue colouration, and is separated from *M. ogilbyi* by significant modal differences in the number of soft dorsal, anal, and pectoral rays.

## **Distribution & Habitat**

*Melanotaenia caerulea* was collected in Papua New Guinea at several sites in the lower and middle Kikori drainage system, spanning a distance of approximately 125 km. They inhabit small tributary streams flowing through rainforest, except at one location where it was collected in a small tidal creek-fed pond in open sunlight. The Kikori River rises in the central mountains of southwestern Papua New Guinea and flows southward for nearly 250 km before forming a major delta at the head of the Gulf of Papua. The river mouth is situated about 340 km northwest of Port Moresby, and approximately 140 km northeast of the Fly River entrance.

The water quality of the mainstream rivers of the Tagari-Hegigio and Lake Kutubu-Digimu-Mubi sub-basins are typical of other mainstream rivers in Papua New Guinea that are near neutral to mildly alkaline (pH 7.4–8.2) and calcium-bicarbonate dominated. These properties are indicative of water draining a limestone catchment area. The lower calcium concentration, alkalinity and hardness of the Ai'io River, which drains to the upper Hegigio River, probably reflect the predominantly volcanic and sedimentary terrain at this location. Water hardness in all rivers except the Ai'io River (31 mg/L CaCO<sub>3</sub>) is moderate (60–119 mg/ L CaCO<sub>3</sub>) to hard (120–179 mg/L CaCO<sub>3</sub>). Conductivity values are generally similar in all streams, with median values ranging between 167 and 267  $\mu$ S/cm.

## Remarks

*Melanotaenia caerulea* was named *caerulea* (Latin: blue) with reference to the characteristic colour pattern. This species is not currently available in the aquarium hobby.





## Melanotaenia catherinae

(de Beaufort, 1910) Waigeo Rainbowfish

*Rhombatractus catherinae* de Beaufort, 1910 *Melanotaenia catherinae* Allen, 1980

### **Species Summary**

Melanotaenia catherinae are bluish to purplish brown on the back and white on the lower side with a blue midlateral band up to  $2\frac{1}{2}$  scales wide. Dorsal and anal fins bluish with a maroon-red wash on the margins. They may reach a maximum size of 10 cm, but usually less than 8 cm. They are closely related to Melanotaenia synergos, which is found on Batanta Island. The two species share similar meristic and morphological features. However, they differ in modal counts for pectoral-fin rays and lateral scales. They also exhibit slight colour pattern differences related to the width of the dark midlateral stripe, which is generally narrower in M. synergos, covering one and a half scale rows for most of its length versus 2 to 3 scale rows for M. catherinae. Moreover, the midlateral stripe of Melanotaenia synergos is nearly covered entirely by the pectoral fin, whereas it is broadly exposed (at least one scale row) above the pectoral fin of Melanotaenia catherinae. Analysis of genetic relationships indicates a close relationship between the two species.

#### **Original Description**

Dorsal profile nearly straight, sloping down from dorsal to snout, a little more convex in large specimens. Ventral profile strongly convex in large specimens. Height in smaller specimens (to 100 mm.) 2.5–3.2, 3–3.75 in length with caudal, in specimens above 100 mm. 2.2–2.5, 2.7–3 in length with caudal. Head 3.2–3.7, 4–4.5 in length with caudal. Eye 3–3.8, about 1.5 in interorbital space, which is about equal to postorbital part of head. Snout rather obtuse, 2.6–3 in head and only a little longer than eye.

Upper jaw prominent. Mouth opening reaching to vertical through front border of eye. Conical teeth in several rows in the jaws, extending to the outside of the lips, which are thickened, especially in their anterior part. A patch of teeth on the vomer and perhaps a few on the hinderpart of the palatines, none on tongue. Two rows of scales on suborbital part of cheeks. Operculum with large scales, excepting the superior ones, which are small. Dorsal separated by 16 scales from occiput. Spine of first dorsal scarcely longer than that of second dorsal, shorter than postorbital part of head and much shorter than that of anal, which is about equal to eye. Origin of anal opposite to that of first dorsal. Length of base of anal longer than distance between origin of first dorsal and end of second dorsal. Pectorals longer than head without snout. Scales nearly smooth, with indication of crenulations. Caudal peduncle longer than high in small specimens, in large specimens considerably higher than long. In life, the colour of the lateral band is dark-blue and the scales have wine-red margins, which form about 8 longitudinal stripes. Proximal part of anal and second dorsal wine-red.

## **Distribution & Habitat**

The type-locality was a brook flowing into the Rabiai River, on Waigiou (Waigeo) Island. However, they have been collected from several streams including the Rabiai River, Wai Semie and the Wai Meniel. *Melanotaenia catherinae* is so far found only on Waigeo and Batanta Islands in the Raja Ampat Group lying immediately west of the Vogelkop Peninsula, West Papua. The Raja Ampat Islands are a group of islands comprising Waigeo, Batanta, Salawati and Misool located to the west of Sorong, on the northwest tip of the mainland of New Guinea. The Raja Ampat Islands are situated immediately west of the New Guinea mainland, between 0°20' and 2°15' S latitude, and 129°35' and 131°20' E longitude. The Archipelago and surrounding seas occupy approximately 40,000 km<sup>2</sup>.







A number of rainbowfishes have been collected from several other islands off the coast of New Guinea. *Melanotaenia misoolensis* from Misool and *Melanotaenia japenensis* from Japen, which are endemic to these islands off the north coast. The Aru Islands off the south coast are inhabited by *Melanotaenia goldiei* (*trifasciata*) and *M. splendida rubrostriata*, both of which are widely distributed on the southern New Guinea mainland. All of these insular areas were formerly connected to the New Guinea land mass and are presently separated by shallow (less than 50 fathoms) seas.

## Remarks

The first aquarium specimens were imported to Germany by Heiko Bleher in 1992, where they were bred and distributed internationally.







## Melanotaenia corona

Allen, 1982 Corona Rainbowfish

## **Species Summary**

*Melanotaenia corona* was described on the basis of two specimens collected in 1911 from the Sermowai River, near Walckenaer Bay, northern New Guinea, about 2°47'S, 140° 00'E. It differs from other members of the genus in the shape of the dorsal and anal fin outline and colour pattern. The body is laterally compressed and elongated. Two dorsal fins, very close together, the first much smaller than the second. May reach a maximum size of 12 cm, but usually less than 10 cm. The second dorsal and anal fins are unusually tall compared with other members of the genus. The longest rays are situated in the middle part of these fins, a feature that is typical of some *Glossolepis*. Live colours unknown.

*Melanotaenia corona* is easily distinguished from other member's of the genus on the basis of colour pattern, particularly the combination of the four broad dark stripes on the back and the very dark coloration of the dorsal and anal fins. Moreover, it is the only member of the genus in which the longest soft rays of the dorsal and anal fin are located in the middle part of these fins, a character which is also present in the genus *Glossolepis*.

## **Distribution & Habitat**

Known only from the upper Sermowai River on the north coast of West Papua, about 75 kilometres west of Jayapura. They were collected by Knud Gjellerup in 1911. Four other species of rainbowfishes were collected by Gjellerup from the Sermowai River: *Chilatherina crassispinosa, Chilatherina fasciata, Chilatherina lorentzi* and *Melanotaenia affinis.* However, it is not known if they share the same habitat with *Melanotaenia corona.* 

#### Remarks

The only two known specimens must have escaped Weber and de Beaufort's attention. The holotype is preserved in Amsterdam, the only paratype was sent to the Western Australian Museum in Perth.

Today *Melanotaenia corona* is still awaiting rediscovery. The specific name is Latin, meaning rim, or border, alluding to the distinctive white margin on the dorsal fins. This species is not currently available in the aquarium hobby.





## Melanotaenia duboulayi

(Castelnau, 1878) Crimsonspotted Rainbowfish

Atherinichthys duboulayi Castelnau, 1878 Aristeus lineatus Macleay, 1881 Aristeus perporosus De Vis, 1884 Rhombatractus lineatus Gill, 1894 Rhombatractus perporosus Ogilby, 1896 Chirostoma duboulayi Waite, 1904 Melanotaenia nigrans Regan, 1914 Melanotaenia nigrans Jordan & Hubbs, 1919 Melanotaenia splendida fluviatilis Allen & Cross, 1982 Melanotaenia duboulayi Crowley, Ivantsoff & Allen, 1986

## **Species Summary**

*Melanotaenia duboulayi* were initially collected in the 1870's from the Richmond River in northern New South Wales by a man named Duboulay (du Boulay). They were later scientifically described as *Atherinichthys duboulayi by* Castelnau in 1878. They were also later known as *Nematocentris fluviatilis* and *Melanotaenia fluviatilis*. Following a review of the rainbowfish group by Allen in 1980, they were reclassified as *Melanotaenia splendida fluviatilis*. Their current scientific name follows from a study of its early life-history stages by Crowley, *et al.*, 1986. This study resulted in *Melanotaenia splendida fluviatilis* being separated into two species, *Melanotaenia duboulayi* from the eastern coastal

drainage systems of northern New South Wales and southern Queensland, and *Melanotaenia fluviatilis* from the inland Murray-Darling River system.

M. duboulavi can reach a maximum size of 12 cm SL, but are usually less than 10 cm. They have a slender and compressed body shape with depth increasing with age. Two dorsal fins, very close together, the first much smaller than the second. They exhibit considerable colour variations over their wide geographical range. Generally, the body is silvery-blue or green ranging through deep bluish or yellow tones. The scale rows are marked with narrow yellow lines and overlaid with orange to brilliant red. A prominent spot of crimson red is seen on the operculum, the fin colours are variable from clear, yellowish to red, with red flecks and dark margins. These dark margins become intensely black in males during spawning activities. The larger males are easily distinguished from females by their brighter colours and can usually be identified from the elongation of posterior rays in the second dorsal and anal fins. Females have rounded dorsal and anal fins, which are smaller and lack the dark edges.

*M. duboulayi* are not easily distinguished from *M. fluviatilis*. Principal variations are body depth, fin counts, and colour pattern. In addition, there are clear differences in egg characteristics and larval development. *M. fluviatilis* often have a broader head and blunter snout compared to *M. duboulayi*.











#### **Distribution & Habitat**

*Melanotaenia duboulayi* inhabits the coastal drainages east of the Great Dividing Range from the Hastings River, New South Wales, approximately 400 km north of Sydney to Baffle Creek, Queensland. They are a subtropical species found in relatively still, clear water, in water temperatures between 16–28° Celsius. Habitat includes freshwater rivers, streams, billabongs, reservoirs, swamps, and lagoons with dense aquatic vegetation. Their natural environment is subjected to seasonal variations with water temperature, *p*H (5.4–7.8), and hardness levels varying considerably.

*Melanotaenia splendida* occupies the east coast drainages of Queensland north of the Burnett River region in the south, to Scrubby Creek, just south of the Lockhart River in the north. The exact species boundaries are unknown and it may be that these two species live sympatrically in some locations.

Melanotaenia duboulayi are usually found in open water areas and around sub-surface vegetation, submerged logs, or branches in mid to lower depths. They usually spend most of their time in the open water areas where they form small groups, with one or two fish breaking away to explore occasionally. The behaviour between the sexes also appears to vary with females forming the basis of the group while the males cruise in search of spawning or feeding opportunities. In sunny conditions groups of juveniles occurred near the water surface feeding on floating material at the surface, but larger fish tended to occur at the bottom near submerged vegetation, often utilizing the aquatic plants as a refuge and food source. In the middle of the day, juveniles and small fish seemed to show behavioural thermoregulation at the surface in the warmest site. Under cloudy conditions, however, fish of all sizes preferred deeper water.

#### **Biology**

Most information on rainbowfish biology is mainly based on aquarium observations. Spawning occurs from September to December before the onset of summer rains. Spawning occurs during the early morning or evening just before dark. Each female lays several eggs a day, which are fertilised by the male. Eggs adhere to fine-leaved foliage plants or among the roots of floating vegetation by several long, thin filaments originating at one point on the egg membrane. The water hardened eggs have a diameter of 0.98-1.8 mm and hatch after 5-9 days after fertilisation at water temperatures between 24 and 29°C. At hatching, larvae 2.5 to 4.2 mm in length have a reduced but still present yolk-sac. The newly hatched larvae congregate near the water's surface within a few hours and begin feeding within 12 hours. At 32 days after hatching, the mean larval length is about 14-15 mm and at 72 days 21-25 mm. Juvenile fish grow quickly and reach maturity in the year following hatching. Sexual maturity occurs at about 4-5 cm for both sexes. Strong sexual dimorphism is present in the species with males typically being larger and brighter in colouration.

#### Remarks

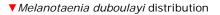
*M. duboulayi* is the original "Australian Rainbowfish" and were being maintained in the aquarium hobby around the turn of the twentieth century. *M. duboulayi* were commonly known as the 'Crimson-spotted Sunfish' and mistakenly identified as *Melanotaenia nigrans*. Amandus Rudel was a founding member of the Aquarium & Terrarium Society of Queensland, and in 1927 he introduced the Australian rainbowfish to the international aquarium hobby when he sent specimens of *M. duboulayi* by steamship to Germany. Speaking of *Melanotaenia duboulayi*, Amandus said, "*I was astonished at the beauty of this fish the first time I saw it. Like a living rainbow, there is no other fish which can compare with its beauty. Naturally it has been my favourite ever since.*"

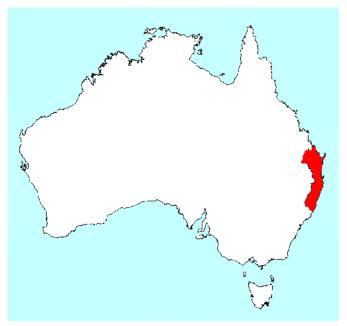
It is believed that from this initial shipment *M. duboulayi* were introduced to the organised aquarium hobby throughout Europe, and then to North America. They are probably the species upon which today's common name "Rainbowfish" is based. In 1930 a number of *M. duboulayi* were collected in the Mississippi River. This was one of the earliest accounts of an introduced ornamental fish found in the USA.

#### Rainbowfish found in US waters...

Three specimens of Melanotaenia nigrans were collected with a small seine from the edge of a sandbar of the Mississippi River, Randolph County, in July 1930. It was supposed that these fish were escapes from a tropical fish establishment in the St. Louis area, about 40 miles upstream. However, an aquarium release cannot be ruled out. The record of O'Donnell (1935) represents one of the earliest accounts of an introduced ornamental fish taken in U.S. open waters. Specimens were identified by Carl Hubbs. There are no known voucher specimens.

These fish would undoubtedly be Melanotaenia duboulayi!









### Melanotaenia eachamensis Allen and Cross, 1982

Lake Eacham Rainbowfish

## **Species Summary**

*Melanotaenia eachamensis* is a small species, slender and compressed but depth increasing with age. Two dorsal fins, very close together, the first much smaller than the second. May reach a maximum size of 8 cm SL, but usually less than 6 cm. Males can be distinguished from females on the basis of differences in colouration and shape of the dorsal, anal and ventral fins. The original males collected from Lake Eacham had an overall bronze body colouration. The first dorsal fin was jet black while the second dorsal and anal fins had a maroon red colouration. Females are rather plain compared with males and have smaller more rounded dorsal and anal fins. Gerald Allen and Norbert Cross described the new species on the basis of differences in colour and body shape from surrounding populations of *Melanotaenia splendida*.

This species was formally described in 1982. A survey of Lake Eacham in 1978 revealed the existence of a rainbowfish along with a hardyhead (*Craterocephalus stercusmuscarum*) and a gudgeon (*Mogurnda mogurnda*). This survey and subsequent collection resulted in the rainbowfish being described as a new species and was thought to be endemic to the lake.

## **Distribution & Habitat**

*Melanotaenia eachamensis* were initially found in Lake Eacham, a 43 ha crater lake located on the Atherton Tablelands about 40 km south-west of the north Queensland town of Cairns. The lake has a northeast-southwest length of around 1.5 km and 1 km wide. Water in the lake is supplied entirely from the catchment area within the crater rim. The water of the lake is neutral with a low level of dissolved salts. The lake is permanent and deep, and fluctuates seasonally with a maximum depth of 65.5 m during the wet season.

#### Remarks

*Melanotaenia eachamensis* is without doubt the most wellknown Australian rainbowfish. This is not because it is the most desirable species to keep but because it was believed to be the first Australian freshwater fish species to ever become extinct. It was one of the most widely publicised examples of the impact of translocated fishes.



# The "eachamensis" Story

*Melanotaenia eachamensis* were initially collected from Lake Eacham, a crater lake located on the Atherton Tablelands about 40 km south-west of Cairns in northern Queensland. The lake has a northeast-southwest length of around 1.5 km and 1 km wide. Water in the lake is supplied entirely from the catchment area within the crater rim. The water of the lake is neutral with a low level of dissolved salts, with water temperatures between 18–28°C. The lake is permanent and deep, and fluctuates seasonally with a maximum depth of 65.5 metres during the wet season.

There are a number of volcanic crater lakes or lake remnants located on the Atherton Tablelands including Lynch's Crater, Strenekoff's Crater, Mobo Crater, Bromfield Swamp, Lake Barrine, Lake Eacham and Lake Euramoo. Three of these volcanic lakes (Eacham, Barrine and Euramoo) occur within close proximity of each other. Lake Euramoo has a relatively small catchment area of about 4500 m<sup>2</sup> with no inflow or outflow channels. The lake has a water depth averaging around 20 metres in the northern end and 16 metres in the southern end, though there are seasonal fluctuations in water depth of between 2 and 3 metres. Lake Barrine is on average 67 metres deep. It is about 1 km in diameter, has a shoreline of almost 4.5 km and is the largest of the natural volcanic lakes in the area.

The surrounding area of both Lake Eacham and Lake Barrine contain creeks that flow into Tinaroo Dam (Barron River) but are not associated with the lakes themselves. Both of these lakes are very close to the impoundment area of Tinaroo Dam into which Wright Creek and Congoo Creek flow. Lake Barrine has flood-flow connections to Toohey Creek, an upper tributary of the Mulgrave River.

*M. eachamensis* is a small rainbowfish species. They may reach a maximum size of 8 cm SL, but are usually less than 6 cm. They have a slender and compressed body with depth increasing with age. *M. eachamensis* is not an overly attractive species but it does have its own distinctive colour and characteristics. They can be very colourful when kept in a suitable captive environment. The original males collected from Lake Eacham had an overall bronze body colouration. The first dorsal fin was jet black while the second dorsal and anal fins had a maroon red colouration. Males can be distinguished from females on the basis of differences in colouration and shape of the dorsal, anal and ventral fins. Females are rather plain compared with the males and have smaller more rounded dorsal and anal fins.

Gerald Allen collected the Lake Eacham rainbowfish in 1978 and they were described as *M. eachamensis* in 1982, although it was considered to be closely related to the widespread *Melanotaenia splendida*. Allen and Cross described the new species on the basis of differences in colour and body shape from surrounding populations of *M. splendida*. In his original description of *M. eachamensis*, Allen points out this very close relationship, and makes clear that it is defined as being separate only because it falls outside the range of parameters for *M. splendida* on several counts. Small freshwater fish species had been reported from Lake Eacham as early as 1925. The first report of rainbowfishes in Lake Eacham, however, was in 1965 by members of the Townsville Aquarium Society. They were generally considered not to be as brightly coloured as other rainbowfishes from the coastal regions. They were never collected however, (well, not officially anyway), and there is no record of them having being maintained in the hobby in those early years. Allen and Cross's description of the Lake Eacham rainbowfish helped to stimulate interest in keeping the fish and fortunately a few specimens were collected for the aquarium hobby in 1980 and 1982 before they disappeared from the lake and were still being maintain by some hobbyists, otherwise this fish would have been lost forever.

Surveys of Lake Eacham in 1973, 1974 and 1978 revealed the existence of a rainbowfish along with a hardyhead (*Craterocephalus stercusnuscarum*) and a gudgeon (*Mogurnda mogurnda*). *M. eachamensis* were abundant within the lake at the time, but during surveys in 1983, 1984 and 1985 four native fish species (*Amniataba percoides, Glossamia aprion, Nematalosa erebi* and *Toxotes chatareus*) were found in the lake. All these fish were presumed to have been translocated to the lake by person or persons unknown.

In 1987, Barlow *et al.* surveyed the lake but failed to locate any rainbowfishes, although the four introduced species were plentiful. Apart from the complete absence of *M. eachamensis*, the survey also failed to locate any gudgeons or any specimens of the native crayfish (*Cherax cairnsensis*), which had been very abundant in the lake in the early 1980's. Thus, two of the three fishes (and potentially a crayfish as well) naturally occurring in the lake disappeared during the same period that four translocated native fishes established breeding populations there. The decline of the rainbowfishes in the lake must have been very dramatic, as I visited Lake Eacham in early 1984 and large numbers of rainbowfishes were still visible, particularly around the floating pontoon that was accessible from the shore.

As a result of the 1987 survey, *M. eachamensis* was declared "extinct in the wild" (some remained in captivity) at the 1987 Australian Society for Fish Biology Conference. Thus, within a few years of its formal recognition, the Lake Eacham rainbowfish was regarded as the first freshwater fish in Australia to have become extinct since European settlement.

This extinction was attributed to a harmful interaction with the translocated species, in particular predation by the mouth almighty (*Glossamia aprion*). Although other rainbowfish species coexist with the translocated species elsewhere in northern Australia, it was assumed that being isolated from predators, *M. eachamensis* was unable to survive.

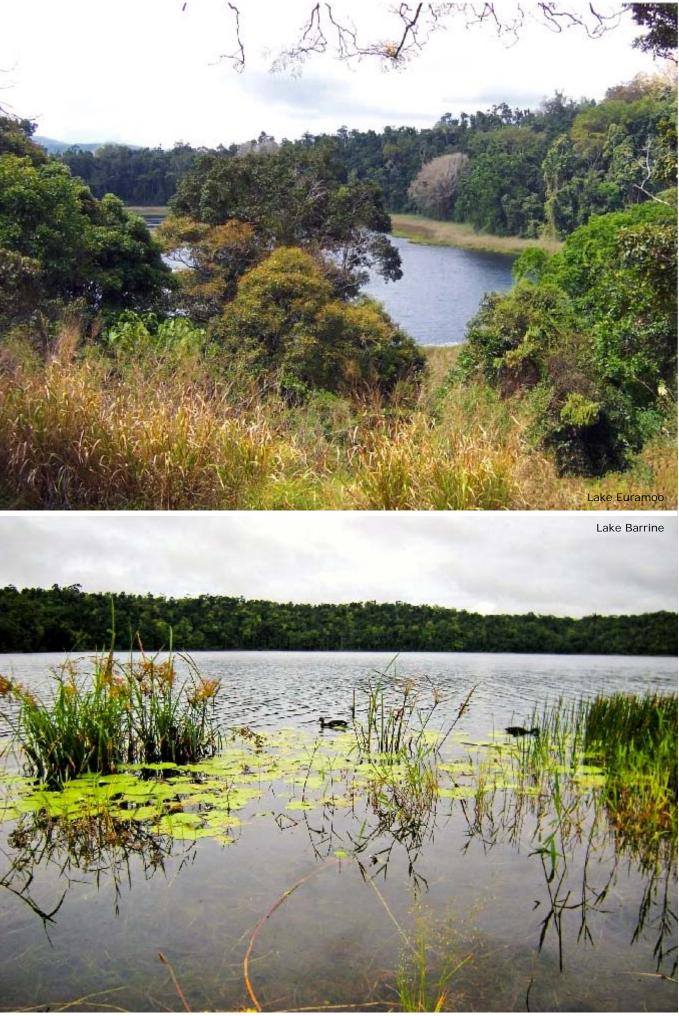
In addition to the above translocations, *Lates calcarifer* were collected there in 1990–1991. *Hephaestus fuliginosus* and *M. splendida* have also been translocated there and are still present. Ironically, *M. splendida* are now present in large numbers and another species, believed to be an exotic has also been observed. All these fish species have been stocked into the lake unofficially. The natural condition of the lake has also declined and sadly it is no longer the pristine lake it once was.











Immediately after the "extinct-in-the-wild" announcement in 1987, members of the Australia New Guinea Fishes Association (ANGFA) at their first national conference in Sydney undertook a survey of members throughout Australia, which revealed the existence of a number of small captive populations of *M. eachamensis*. These stocks were designated as the 'Bowman' and 'Tappin' populations. ANGFA instigated a captive-breeding program known as "Project Eachamensis" to stimulate and actively encouraged the establishment of new captive populations. Within two years, numerous 'Bowman' and 'Tappin' populations were established in the Australian hobby. In addition, eggs were sent to North America and Europe where populations were established, and the species was considered secure in captivity.

Much finger pointing followed as to who was responsible for the demise of the Lake Eacham rainbowfish until public embarrassment got the better of all and government funding was found to investigate the genetic distinctiveness of *M. eachamensis* and to evaluate the genetic composition of existing captive stocks with a view to re-establishing the rainbowfish back into its natural habitat. Not only was this the first reported extinction of a native fish but Lake Eacham is situated in a National Park under control of the Queensland National Parks administration and also in a World Heritage area under the control of the Wet Tropics Management Authority.

A number of young *M. eachamensis* were supplied to the Walkamin Research Station (inland from Cairns) to establish a research and breeding stock. A similar number of specimens were supplied to Sydney's Taronga Park Zoo Aquarium for display, to heighten public awareness, and for breeding stock; however, owing to a mycobacteria disease outbreak this group all died several years later.

At Walkamin Research Station a breeding pond was set aside and within 18 months several thousand rainbow fish had been produced. In November 1989, 3,000 *M. eachamensis* bred from captive stocks were released into the lake but none could be located in surveys just three months later and throughout 1990. As this reintroduction attempt was unsuccessful, and the removal of the translocated fishes from Lake Eacham were considered unlikely, there were no further attempts to restore *M. eachamensis* to the lake.

In the meantime, while this fascinating chronicle of events was evolving, ANGFA member, David Liddle was exploring a number of streams on the Atherton Tablelands looking for rainbowfishes and discovered an "eachamensis" look-alike in Dirran Creek (an upland tributary of the North Johnstone River), located about 22 km south of Lake Eacham. Another unusual rainbowfish species was also found in Lake Euramoo, a crater lake situated about 14 km north of Lake Eacham.

In 1991, Crowley and Ivantsoff conducted an electrophoretic analysis of a number of the captive-bred Lake Eacham rainbowfishes and reported that they could not distinguish electrophoretically between *M. eachamensis* and *M. splendida*. However, studies by Moritz *et al.* (1995) reported contrasting conclusions. mtDNA analyses of rainbowfish collections from catchments adjacent to Lake Eacham and captive bred specimens confirmed *M. eachamensis* as a separate species distinguishable from *M. splendida* from surrounding areas. Furthermore, it was concluded that both Lake Euramoo and Dirran Creek, and possibly Charappa Creek (an upland tributary of the South Johnstone River) exhibited a 'pure' lineage with *M. eachamensis*. Lake and stream populations were also genetically distinct and these differences were retained in captive bred populations. Following on from there, a flurry of scientific work was undertaken that resulted in a better understanding of this species complex and the distribution of the scattered populations. As well, fascinating behavioural research revealed that the long isolation of some of these populations robbed them of protective predator avoidance responses and this led to accelerated losses and their ultimate rapid extinction by translocated (but still native) predators.

Lake Barrine also contained rainbowfishes of uncertain identity, although these were listed as M. splendida at the time. However, in a brief survey of Lake Barrine in 1991 numerous Glossamia aprion were found, but no rainbowfish. As Lake Barrine has a flood-flow connection to the upper Mulgrave River via Toohey Creek, the rainbowfish in Lake Barrine may have been the common M. splendida or if the waterfalls in the upper Mulgrave River and Toohey Creek prevented colonisation by M. splendida, they may have been another form of rainbowfish. Rainbowfish from Lake Euramoo were analysed as part of the genetic studies of the Lake Eacham rainbowfish (Zhu et al. 1998, McGuigan et al. 2000). However, no specimens from Lake Barrine were included in the study as the researchers could not locate any there. A number of predatory native fishes have also been translocated into Lake Barrine. Incidentally, translocated native predators also now occur in Dirran Creek.

In 1983, I obtained wild-caught specimens of another rainbowfish of uncertain identity from Mobo Crater. Mobo Crater is located between Lake Euramoo and Lake Barrine. This fish was being distributed within the rainbowfish hobby as *M. eachamensis* (Mobo Crater) as late as 1999, but the original wild-caught fish looked nothing like the Lake Eacham rainbowfish. This form however, now seems to have disappeared from the aquarium hobby in Australia. Limited stocks are available in Europe.

As a representative of ANGFA, I attended a 'Lake Eacham Rainbowfish Workshop' in Cairns during September 1995. This workshop was attended by representatives from Walkamin Research Station, Queensland Fisheries Management Authority, University of Oueensland, Oueensland Department of Environment & Heritage, Queensland Department of Primary Industries, Wet Tropics Management Authority, James Cook University, Sydney's Taronga Zoo, and the Australia New Guinea Fishes Association. At this workshop, a number of problems had to be taken into account before developing a conservation strategy for M. eachamensis. However, while it was considered desirable that the Lake Eacham Rainbowfish be returned to the lake, the eradication of the translocated fishes from Lake Eacham was considered unlikely to be achievable, economical or sustainable using existing technology. It was also recommended that the conservation status of *M. eachamensis* be downgraded from "extinct-in-the-wild" to vulnerable under the Nature Conservation Act 1992.





Using an analysis of morphological and meristic characters, Pusey *et al.* (1997) believed *M. eachamensis* to be even more widespread, occurring in many upland and several lowland tributaries and reaches of the North and South Johnstone Rivers; in upland tributaries of the Herbert River; upper Tully River and the upper Daintree River. Subsequent genetic work (Zhu *et al.* 1998, McGuigan 2000, McGuigan *et al.* 2000, Hurwood and Hughes 2001) suggested that at least some of these occurrences are not *M. eachamensis* but either unusual variants of *M. splendida*, *M. utcheensis* or populations displaying alleles (one member of a pair or series of genes that occupy a specific position on a specific chromosome) of more than one species.

Rainbowfishes from Utchee Creek, a tributary of the South Johnson River, had long been recognised by rainbowfish enthusiasts as being different, although scientifically known as *M. splendida*. The above mentioned research found that most of the specimens studied from this stream were phenotypically indistinguishable from *M. eachamensis*. However, additional research indicated that the 'Utchee Creek' variety was indeed a distinct species, and they were formally described as *Melanotaenia utcheensis*, with populations known from Utchee, Fisher, Rankin and Short Creeks in the North and South Johnstone catchments (McGuigan 2001).

Genetic analysis also revealed *M. eachamensis* occurred in Bromfield Swamp in the North Johnstone River headwaters (McGuigan 2000). Bromfield Swamp occupies a partially breached crater and is very shallow. Bromfield Swamp is an explosion crater, from which water drains from an outlet on the east side. The swamp, which is 500 metres in diameter is approximately 45 metres below the rim of the crater, and was once surrounded by tropical rainforest.

Zhu *et al.* (1998) also found populations that contained a mixture of alleles from *M. eachamensis* and *M. splendida*, in other locations such as an irrigation channel from Tinaroo Dam (Walkamin "eachamensis"), Streets Creek (Kuranda Reds), upper Barron, and other tributaries of the North and South Johnstone Rivers such as Williams Creek and Ithaca Creek. The finding of fish with *M. eachamensis* alleles in irrigation channels of Tinaroo Dam would probably represent a translocation of these species to the upper Barron River catchment. The unusual distribution of *M. eachamensis* alleles demonstrated by Zhu *et al.* (1998) may also suggest that it was translocated to other locations, and raises the possibility that even Lake Eacham may not have been their original habitat.

On the Atherton Tablelands there are apparently at least three species of rainbowfish (*M. splendida*, *M. eachamensis* and *M. utcheensis*) which all live within close proximity of one another. It has been suggested that *M. utcheensis* and *M. eachamensis* were the original inhabitants of the region and *M. splendida* may have invaded relatively recently. It has been suggested that the dispersal of rainbowfishes between the various river systems on the Atherton Tablelands had occurred due to rearrangements of the streams (e.g., river capture) at some stage in the past. The species boundaries of all three species are not well defined and recent evidence suggests that at least some populations have hybridised in the streams of the Cairns-Atherton region.

Rainbowfishes are notoriously easy to hybridise in an aquarium and although there is not a lot of evidence of this in the wild, it may have more to do with the fact that it just hasn't been recorded, rather than it not actually occurring. Male rainbowfishes in captivity are not very choosy when it comes to spawning with females even if the females are a "different" species.

In general, rainbowfishes evolve into different species and subspecies after becoming geographically isolated from others, adapting to their different environments, and changing over time through the process of natural selection. Geographic populations of rainbowfishes have been isolated from each other for perhaps thousands of years. They have gradually evolved physical changes that reflect that adaptation. However, despite the research that has been undertaken to date, the specific status and distribution of *M. eachamensis* still remains unclear.

## Remarks

I obtained about twelve wild-caught specimens of *M. eachamensis* in May 1982. These were placed in a single species aquarium, and in September 1982 they were spawned. During 1983–84, I distributed large numbers of tank-raised young adults to interested aquarists as well as some retail and wholesale outlets. However, like many other rainbowfish keepers, in those early years, the advent of the more colourful New Guinea rainbowfish found their way into my aquariums and my stock of *M. eachamensis* slowly faded away. In 1987, following their reported extinction-in-thewild I once again obtained stock from the original collector, who still had some specimens from the 1982 collection, and was once again spawning this remarkable fish. I continued to maintain a small captive population until February 2000.

Despite the research that has been undertaken to date however, it is my opinion that these "genetic" look-a-likes of *M. eachamensis* don't physically look like the original fish collected from Lake Eacham and maintained by myself for many years. While I would agree that the fish from Dirran Creek are very similar, the rest of the so-called "*Melanotaenia eachamensis*" look nothing like the original Lake Eacham Rainbowfish.

I also have doubts about the validity of some of the "*M. eachamensis*" being maintained in the Australian hobby today. The problem is that many of these "look-a-likes" have and are being distributed and bred under the umbrella name "*eachamensis*" and present captive stocks do not look like the original fish. I suspect that there are very few genuine descendants of the original "Lake Eacham" rainbowfish still in existence. There may be some original stock in Europe and North America if they haven't been contaminated with the "look-a-likes" as they have in the Australian hobby.

Another problem with rainbowfishes kept in captivity is that instead of natural selection, selection is done by the aquarist; because only a relatively small number of fish can be kept, the aquarist tends to select for those which grow best and look best under aquarium conditions. In the long term, the fish being kept may be genetically a long way from the original wild fish, and may even look very different. It's possible that the Lake Eacham form of *M. eachamensis* is no longer a viable population.



Rainbowfishes occupy such a wide variety of habitats that we couldn't reasonably expect evolution to have resulted in a neat, uncomplicated, uniform species. I like to think that rainbowfishes exist in nature as unique populations irrespective of the name they carry. Many things in nature cannot be rigidly and accurately categorised and the Lake Eacham population is worthy of our attention and preservation. Whether or not these "look-a-likes" are truly M. eachamensis, emphasis should be placed on retaining the known pure populations of the Lake Eacham Rainbowfish and protecting these from interbreeding with the related species. Therefore, specific names based on the locality where each is found should be used by rainbowfish enthusiasts to identify each form. Where populations need to be identified, they should be sold and distributed by inclusion of a form or population identifier in brackets following the species name e.g., Melanotaenia eachamensis (Dirran Creek).

Despite the above developments, captive breeding programs can and do have an important impact on the conservation of threatened species, and the preservation of their natural habitats. Such programs must, however, be part of a well coordinated approach involving all interested groups. I guess we can all learn a lesson from this and that is not to translocate any fish from one habitat to another and not to release any unwanted aquarium fish into a natural environment. If you have aquarium fish that you no longer require then please return them to the shop where you purchased them or dispose of them humanly.





Lake Eacham Habitats



## Melanotaenia exquisita

Allen, 1978 Exquisite Rainbowfish

#### **Species Summary**

*Melanotaenia exquisita* have a slender and compressed body. Two dorsal fins, very close together, the first much smaller than the second. Mature males have a higher first dorsal fin, which overlaps the origin of the second dorsal fin when depressed. Females have smaller rounded dorsal and anal fins. May reach a maximum size of 9 cm, but usually less than 8 cm. Adults are olive on the back and silvery white on the lower half. There is a pair of prominent stripes running along the middle of the side with a red stripe just below. A pair of dark zigzag stripes is situated between the red stripe and base of the anal fin. The dorsal, anal, and caudal fins are edged with red, frequently with small black spots. Males are relatively slender compared to that of most other rainbowfishes.

#### **Distribution & Habitat**

*Melanotaenia exquisita* was originally collected in 1977 by Gerald Allen and Geoff Evans from the Edith River about 1 km upstream from Lake Malkyullumbo, Northern Territory. At the time *Melanotaenia exquisita* were believed to occur only in the Northern Territory. However, in 1986 more of these exquisite fishes were discovered in the King George River in the Kimberley region of Western Australia. Then in 1997, another population was discovered in Bindoola Creek, a small stream that flows into the Pentecost River in the Cambridge Gulf in the far north-eastern Western Australia. Since then a number of populations have been found.

Preliminary genetic studies of *Melanotaenia exquisita* from Bindoola Creek have shown some clear differences from other known populations. Further genetic and morphological studies may justify its recognition as a distinct species.

*Melanotaenia exquisita* typically inhabit small, clear, swiftflowing streams, often congregating in rock pools at the base of small waterfalls such as Jim Jim Falls in the South Alligator system and Seventeen Mile Falls in the Katherine system (plus the King George Falls in Western Australia). They also occur in the still waters of Lake Malkyullumbo at the base of Edith Falls.

## Remarks

Another rainbowfish species known in the hobby as "Waterfall Creek Exquisita" comes from above the Gunlom Falls (also known as UDP Falls or Waterfall Creek Falls) in Kakadu National Park. Gunlom Falls is a waterfall on Waterfall Creek about 200 km east-southeast of Darwin in

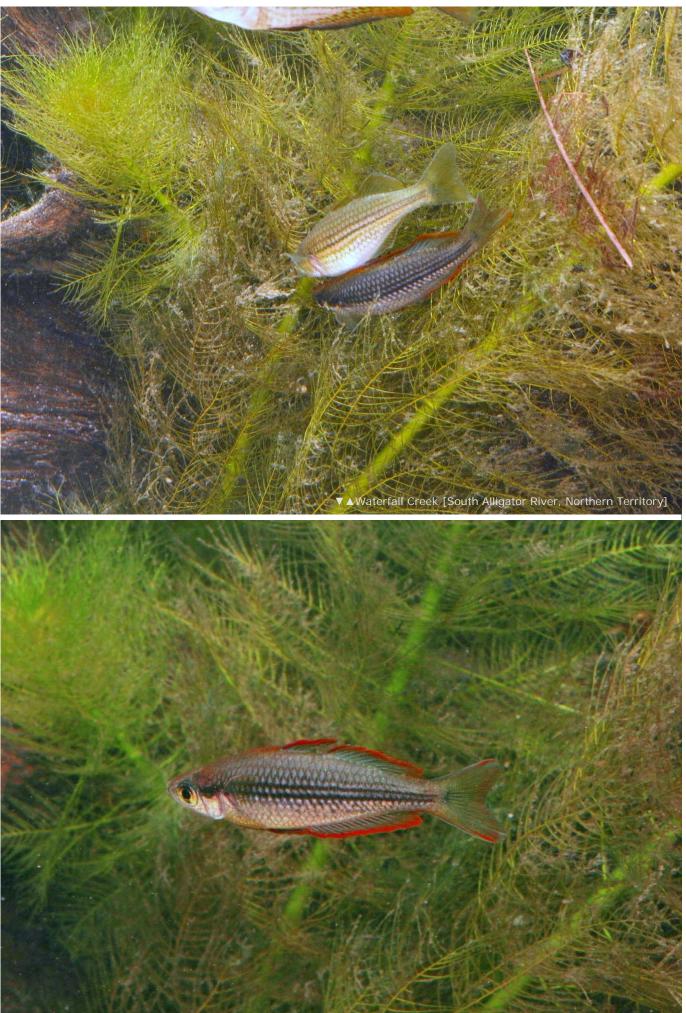




"Sleisbeck" ▲ male ▼ female [South Alligator River, Northern Territory]







Dave Wilson

the Northern Territory. Waterfall Creek flows into the South Alligator River. Permission to collect specimens from this site for whatever purpose is almost impossible to get. However, there are a number of captive populations in existence. They differ from *Melanotaenia exquisita* by having a deeper body and totally different colour. The males show a charcoal black chequer-board pattern over the body with red edging on the dorsal and anal fins. When spawning the nape band is a bright orange-red colour.

Genetic studies (P. J. Unmack 2009, pers. comm.) suggest that the "Waterfall Creek Exquisita" are an introgressed (e.g. hybridised) population of M. exquisita and M. nigrans. Introgression, which seems to be common among some sympatric rainbowfishes in their natural environment, as suggested above, frequently blurs the differentiations of rainbowfishes we recognise as species or populations. Although technically a 'hybrid', this process may contribute to a continuum in speciation and genetic diversity. This may be important to the species' ability to adapt to changing environmental conditions over time. Some populations may become better adapted than others; some may become extinct. However, it is a good idea not to maintain the different varieties together in the same aquarium.







# Melanotaenia fluviatilis

(Castelnau, 1878) Murray River Rainbowfish

Aristeus fluviatilis Castelnau, 1878 *Rhombatractus fluviatilis* Gill, 1894 *Melanotaenia neglecta* Rendahl, 1922 *Nematocentris fluviatilis* Whitley, 1957 *Melanotaenia splendida fluviatilis* Allen, 1980 *Melanotaenia fluviatilis* Crowley, Ivantsoff & Allen, 1986.

## **Species Summary**

*Melanotaenia fluviatilis* were initially collected during the 1870s from the Murrumbidgee River in New South Wales and scientifically described as *Aristeus fluviatilis* by Castelnau in 1878. Until 1986 this species was considered the same as *Melanotaenia duboulayi* (both were known as *Melanotaenia fluviatilis*). Following a review of the rainbowfish family in 1980, they were renamed *Melanotaenia splendida fluviatilis*. However, a study of its early life-history stages resulted in *Melanotaenia duboulayi* from the eastern coastal drainage systems of northern New South Wales and southern Queensland, and *Melanotaenia fluviatilis* from the inland Murray-Darling River system. They are not easily distinguished from *Melanotaenia* 

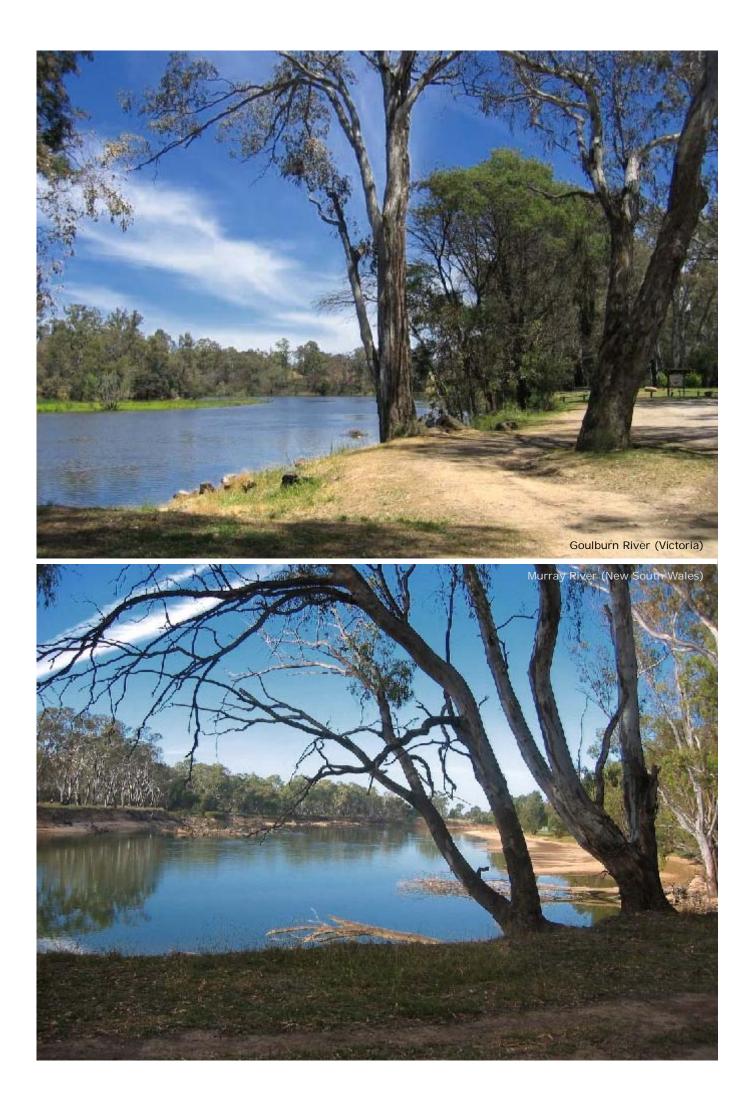
*duboulayi.* Principal variations are body depth, fin counts, and colour pattern. In addition, there are clear differences in egg characteristics and larval development. Southern populations often have a broader head and blunter snout compared to fish from the northern part of the range.

*Melanotaenia fluviatilis* is a small species with a maximum size of 10 cm, but more commonly less than 8 cm. Males are usually much larger and deeper bodied than females. They have two dorsal fins, very close together, the first much smaller than the second. Mature males can usually be identified from the elongation of posterior rays in the second dorsal and anal fins and are more brightly coloured. Females have smaller rounded dorsal and anal fins.

As with most rainbowfishes, the colour is variable depending on location and water conditions. The basic colour is olive, brownish, or slightly turquoise on the back and upper side grading to white on the lower half. A thin reddish stripe is situated between each horizontal scale row, particularly in mature males. There is sometimes a blackish, mid-lateral stripe. Fins are clear to reddish, sometimes with faint spotting. Males may have blackish margins on the dorsal, anal, and anterior margin of the pelvic fins, especially during courtship and spawning. Prior to spawning the male's colour intensifies becoming emerald green, the throat orange and the tailfin red.







#### **Distribution & Habitat**

*Melanotaenia fluviatilis* is the most southerly ranging rainbowfish in Australia and is the only species adapted to low winter temperature (normally around 10 to 15°C). *Melanotaenia fluviatilis* possess a range of temperature and salinity tolerances. However, there is some evidence that numbers are seriously reduced during winter periods, when water temperatures drop below 10°C. Southern populations can survive a few days at 7°C, but are susceptible to bacterial and protozoan infection at these temperatures. Their distribution covers the Murray-Darling River system in Queensland, New South Wales, Victoria, and South Australia. However, specimens collected from the Warrego, Paroo and upper Darling Rivers (and other streams between those tributaries) have been identified as *Melanotaenia splendida* subsp. *tatei* based on unpublished allozyme and mitochondrial DNA data (P. J. Unmack *pers. comm.*).

*Melanotaenia fluviatilis* have been found in the middle and lower sections of the Murray, Murrumbidgee and Macquarie Rivers, and in several tributaries of the Darling River. They are considered common and abundant in the Broken River and near Mildura in Victoria, around the Murray-Darling confluence, and in parts of the Goulburn River in Victoria. This species is also present in the middle to upper parts of the Gwydir River near Bingara, the Namoi River around the Peel River, Caroll Gap-Somerton on the Dumaresq River, and the Bogan River near Bogan Gate. It is moderately common in some areas of its distribution range. However, Victorian and South Australian populations seem to be decreasing in number. This is not surprising; as the Murray River is the most used and abused river system in Australia.

*Melanotaenia fluviatilis* inhabit rivers, streams, billabongs, drainage ditches, reservoirs, overflows, swamps, and ponds with dense aquatic vegetation. Their natural environment is subjected to seasonal variations with water temperature, *p*H, and hardness levels varying considerably. They occupy a diverse variety of habitats, occurring in almost every kind of freshwater habitat, from slow-moving streams, swamps, lakes and clear flowing rivers. However, they prefer slow-flowing or still clear water with dense

aquatic vegetation, in water temperatures between 18–28° Celsius. They are usually found along grassy banks, or around sub-surface vegetation, submerged logs and branches.

#### Biology

Not a lot is known about the biology of Melanotaenia fluviatilis in their natural habitat. Spawning usually occurs from October to January as water temperatures rise. Females produce between 100 and 150 eggs, spawning a number of times daily for several days. Spawning occurs during the early morning or evening just before dark. Each female lays several eggs a day, which are fertilised by the male. Eggs are spherical and colourless and adhere to fine-leaved foliage plants or among the roots of floating vegetation by several thin filaments originating at one point on the egg membrane. The water hardened eggs have a diameter of 0.98-1.08 mm and hatch in 5-9 days after fertilisation at water temperatures between 24 and 29°C. At hatching, larvae 2.5 to 4.2 mm in length have a reduced but still present yolk-sac. The yolk sac is fully absorbed within 3-5 days after hatching.

The newly hatched larvae remain in the upper 1-cm water laver within a few hours and begin feeding within 24 hours. The swim bladder inflates within 10 hours of hatching. During the next 12 days there are few changes in larval morphology. The swim bladder gradually elongates to become cylindrical. At 32 days after hatching, the length of the larval is about 13-15 mm and at 72 days 21-25 mm. Growth rates vary greatly with differences in temperature, feeding rate and densities. Juvenile fish grow quickly and reach maturity in the year following hatching. Sexual maturity occurs at about 4-5 cm for both sexes. Strong sexual dimorphism is present in the species with males typically being larger and brighter in colouration. Melanotaenia fluviatilis is essentially carnivorous, feeding on both aquatic invertebrates associated with its weedy habitat and terrestrial arthropods which may fall onto or alight on the water's surface; however, it is also known to consume algae and fallen plant pollens.





Melanotaenia fredericki (Fowler, 1939) Sorong Rainbowfish

Charisella fredericki Fowler, 1939 Melanotaenia fredericki Allen, 1990

## Species Summary

The basic body colour of Melanotaenia fredericki is mauve with blue reflective scales above the midlateral band. They have broad yellow shading immediately below the midlateral band on the middle of the body (most prominent in juveniles). Mature fish sometimes show the yellow colouration only on the scale edges. The fins are mainly translucent, but the dorsal, anal and caudal may have a hint of yellow or red. Males may reach a maximum size of 12 cm, but females usually less than 10 cm. Males are more brightly coloured, larger, and deeper bodied than females.

M. fredericki was described in 1939 by Henry W. Fowler on the basis of very young specimens (22-28 mm) found in the vicinity of Sainkedoek, on the Vogelkop Peninsula. They were collected during the Denison-Crockett South Pacific Expedition in 1938 from a stream in the Wa(r) Samson River drainage. However, in his book "Rainbowfishes of Australia and New Guinea" in 1982, Gerald Allen wasn't certain whether M. fredericki represented a valid species. He placed



Fowler's Charisella fredericki, as it was originally named, as a synonym of Melanotaenia goldiei. However, adult specimens collected in 1989 confirmed their validity as a distinct species.

## **Distribution & Habitat**

*M. fredericki* is currently found only in a few small creeks in the vicinity of Sorong at the western end of the Vogelkop Peninsula, West Papua. They have been collected from clear slow flowing streams in closed canopy rainforest -pH 6.5-7.5; Temperature 24-28°C, usually around sub-surface vegetation, submerged logs, or branches.

#### Remarks

A number of live specimens were collected by Heiko Bleher in 1992 and distributed in the aquarium hobby. Other live specimens have been collected from the Warsamson River in the Sorong region and are currently being distributed in the ornamental fish trade as M. fredericki.





# Melanotaenia goldiei

(Macleay, 1883) Goldie River Rainbowfish

Aristeus goldiei Macleay, 1883 Nematocentris novae-guineae Ramsay & Ogilby, 1886 Rhombosoma goldiei Ogilby, 1896 Rhombatractus goldiei Ogilby, 1896 Rhombatractus novae-guineae Ogilby, 1896 Melanotaenia dumasi Weber, 1908 Rhombatractus kochii Weber, 1908 Rhombatractus weberi Regan, 1908 Rhombatractus senckenbergianus Weber, 1911 Rhombosoma novae-guineae Regan, 1914 Rhombatractus archboldi Nichols & Raven, 1934 Anisocentrus dumasi Munro, 1958 Melanotaenia goldiei Allen, 1991

## **Species Summary**

*Melanotaenia goldiei* have a distinctive coppery coloured sheen on the upper half of the body with a creamy white colour on the lower half. The mid-lateral stripe is discontinuous, dark blue or blackish, and about 2 scale rows wide. There is a narrow copper or orange-coloured stripe between each scale row on the upper half of the body. Males may reach a maximum size of 10 cm, but females are usually less than 8 cm.



## **Distribution & Habitat**

*M. goldiei* were initially collected from the Goldie River, a major tributary of the Laloki River in southern New Guinea, near Port Moresby in the 1880's. The Laloki River and its major tributaries, the Brown and Goldie Rivers, arise in the lush foothills of the Owen Stanley Ranges in the Central District of Papua New Guinea.

*M. goldiei* is one of the most widely distributed rainbowfishes in southern New Guinea, ranging from Lake Yamur (West Papua) eastward to the Port Moresby region. They are very abundant and one of the most common rainbowfish throughout the region. They have also been found on the Aru Islands. Other river systems where *M. goldiei* has been collected include the Fly, Kemp Welsh, Lakekamu, Lorentz, Ok Tedi, Palmer, Oriomo and Sapoi Rivers as well as the Timika region in West Papua. In 2005, *M. goldiei* were collected in 17 sites in the Fly River catchment - Elevala River, Ok Tedi, upper Fly River, Ok Mart and Ok Menga.



M. goldiei are found in a wide ranges of habitats, including swamps, backwaters, small creeks, and large rivers. They are most abundant in deep pools behind fallen logs or buttress roots of large trees, where they form loose midwater aggregation. They occur most frequently around sub-surface vegetation, submerged logs, or branches in small tributary streams. Typical habitat consists of small, clear; slow-flowing creeks in closed canopy forest over relatively flat terrain. These creeks typically have mud or gravel bottoms and littered with leaves and log debris. Their natural environment is subjected to seasonal variations with water temperature, pH, and hardness levels varying considerably, and they adapt to the particular water conditions and the seasonal changes when they occur. They have been found in company with Melanotaenia ogilbyi, Melanotaenia papuae, Melanotaenia splendida rubrostriata and Melanotaenia sylvatica. Temperature and pH recorded 24.6-33.0°C; pH 7.0-7.8.

#### Remarks

*M. goldiei* was one of the first New Guinea rainbowfishes to enter the aquarium hobby. They were being maintained in the Australian hobby as early as 1958 and perhaps even earlier. Just how many separate collections have occurred over the years is not known, but I know of at least 3 during the early 1970's. What eventually happened to the fish from these importations has been lost in the pages of aquarium history. One collection that has been documented was made by Gerald Allen in 1978. However, as the number of new species arrived from New Guinea, *M. goldiei* fell out of favour with hobbyists and most of the captive stock disappeared.

Another collection was made by Heiko Bleher around the late 1980s, and these were introduced to the European hobby. They are known as the "Tapini" variety. These were collected from the Loloipa River, which is an upper tributary of the Angabanga River which flows to the sea near Bereina, Papua New Guinea.

Allen (1982) suggested that *M. goldiei* and *M. trifasciata* from northern Australia originated from one and the same ancestral species. In the time when New Guinea and Australia were connected by land, this species must have lived in the area where now the Arafura Sea is. After the rising of the sea levels the separated populations have become the two species we know today. However, recent genetic studies (P. J. Unmack 2009, *pers. comm.*) now suggest that the populations from the Fly River may actually be *Melanotaenia trifasciata*.

For much of the past three million years, Australia and New Guinea were a single land mass, with a wide plain across what is now the Arafura Sea. The only high ground on the plain were low hills that are now islands fringing the Kimberley coast and Arnhem Land, the islands in Torres Strait and the low hills that fronted the north-western coastline of the Arafura plain (now the Aru Islands of Indonesia).

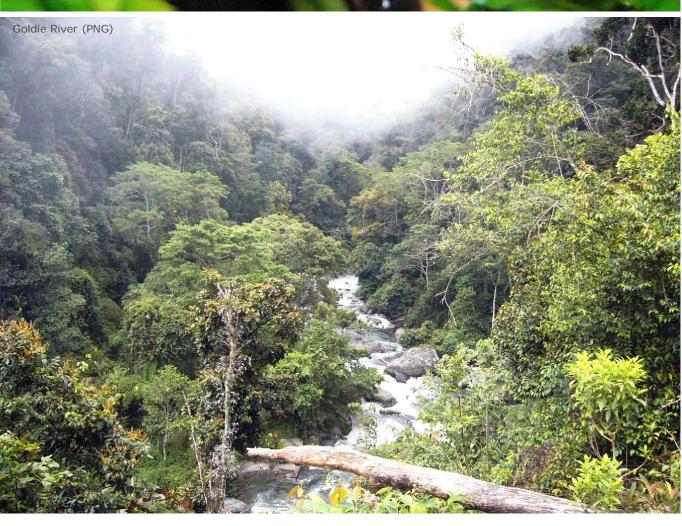
The Aru Islands (also known as Aroe Islands or Kepulauan Aru) lie on the western edge of the shallow seas of Torres Strait, around 7°S and 134°E, and is part of the Maluku province of eastern Indonesia, which lies directly south of the Vogelkop Peninsula, West Papua. New Guinea is some 150 km to the north across a shallow sea; central Arnhem Land in Australia is some 550 km to the south.



Gunther Schmida









## Melanotaenia gracilis Allen, 1978

Slender Rainbowfish

## Species Summary

*Melanotaenia gracilis* were first discovered in 1975 by Barry Hutchins from the Western Australian Museum. They have a rather more slender body than most other rainbowfishes and laterally compressed. The lower half of the body has a wash of bright iridescent lavender. Fins are transparent or slightly pink with a bright red border on the dorsal and anal fins, and red tips on the caudal lobes. Closely related to *Melanotaenia nigrans*, apparently having evolved from a common ancestor. The midlateral stripe of *Melanotaenia gracilis* is not as well defined as that of *Melanotaenia nigrans*. Mature males are usually much larger and deeper bodied than females. Males may reach a maximum size of 10 cm, but are usually less than 8 cm.

## **Distribution & Habitat**

The overall distribution of *Melanotaenia gracilis* is highly restricted, with it known only to occur in the King Edward and Drysdale River systems. Most collections have taken place in the Drysdale River. They have been collected from the Carson River and Morgan Rivers (tributaries of the King Edward River), about 140 km west northwest of Wyndham in the Kimberley region of northern Western Australia. They have only been collected at two Carson River tributary sites and at a single tributary site within the Morgan River. At this site it was however very abundant, with 265 individuals captured. They are generally found congregating around submerged aquatic vegetation, fallen tree branches etc., in clear, slow-flowing tributary streams.

The following creeks and rivers flow into the King Edward River: Hair Creek, Drum Creek, Mainroads Creek, Noolawayoo Creek, Coondillah Creek, Carson River and Parndia Creek. The Carson River (155 km) is a major tributary of the King Edward River. The following creeks and rivers flow into the Carson River: Morgan River, Laurie Creek, Swider Creek and Pronga-Marie Creek. The Morgan River (103 km) merges with the Carson River. The Morgan River flows through Wollangooyoo Pool on its way to joining the Carson River. The following creeks flow into the Morgan River flows through Wollangooyoo Pool on Creek, Gnamoongie Creek, Pangoor Creek and Changoola Creek.

#### Remarks

The first live specimens to be successfully established in the aquarium hobby were collected in 1986 by Ray Leggett and Graham Heidke. Since then there have been numerous collections and they are now reasonably well established.







## Melanotaenia herbertaxelrodi Allen, 1981

Lake Tebera Rainbowfish

## **Species Summary**

Melanotaenia herbertaxelrodi belongs to a group of rainbowfish species, which inhabits the Highland drainage systems of the Kikori and Purari Rivers. This group contains two other species, M. monticola and M. lacustris. Gerald Allen believes that the three species are probably derivatives of the same ancestral stock as M. goldiei, which ranges widely in the lowland and foothill areas of southern New Guinea. M. herbertaxelrodi is most closely related to M. monticola from the upper Purari System near Mendi, about 200 km upstream from the Lake Tebera Basin. Males are mainly bright yellow (sometimes greenish) with a blueblack, mid-lateral line. The dorsal, anal, and caudal fins can be red or yellow. During spawning the male has an intense blue or white stripe from the first dorsal fin extending down over the nape to the tip of the snout, while the whole head can become almost black. The rest of the body is bright yellow with reddish fins.

*M. herbertaxelrodi* may reach a maximum size of 12 cm, but usually less than 10 cm. Males are typically deeper bodied than females and develop a high forehead and an angulated breast profile with increased growth. The body begins to deepen in males after a length of 45–50 mm SL is attained or at about the onset of sexual maturity. In addition,

the middle portion of the first dorsal fin is much longer in males and the posterior outline of the second dorsal fin is more pointed than in females, although this difference is not nearly as apparent as in many other members of the *Melanotaenia* genus.

## **Distribution & Habitat**

*M. herbertaxelrodi* was collected by Gerald Allen and Brian Parkinson in September 1980 from a small clear water stream about 4 km east of Lake Tebera. Lake Tebera, about 410 kilometres northwest of Port Moresby, is situated in the rugged Central Highlands of Papua New Guinea and is part of the Purari River System. Lake Tebera is composed of numerous interconnecting ponds, swamps and springs that occupy a basin that is about 10 kilometres long and 2 kilometres wide. *M. herbertaxelrodi* are usually found around the shoreline margin in tall grasses or sub-surface vegetation.

The search for *M. herbertaxelrodi* came about after Patricia Kailola, then curator of the Kanudi Fisheries Research Lab in Port Moresby, sent a photo to Gerald Allen of an unusual rainbowfish collected by Grant West at Lake Tebera. Although the specimens in the photo were dead and faded, Gerald Allen recognised that it was clearly a new species. After collecting specimens for proper identification, he later named them *M. herbertaxelrodi* in honour of Herbert R. Axelrod who funded the collecting expedition.





## Remarks

The first live specimens to enter the aquarium hobby were collected by Gerald Allen in 1980 and were brought back into Australia where they were later bred and distributed in the hobby.









# Melanotaenia irianjaya

Allen, 1985 Irian Java Rainbowfish

## **Species Summary**

Melanotaenia irianjaya have an overall mauve colouration with silvery reflections. There is a broad, bluish midlateral band, which is most prominent on the rear half of the body. The dorsal and anal fins are reddish with narrow white margins. The caudal fin is reddish and has distinctive black upper and lower margins. This species is unusual in having the middle rays of the dorsal and anal fins longer than the other rays; a feature generally associated with the genus Glossolepis. Males may reach a maximum size of 12 cm, but females are usually less than 10 cm. Unlike most rainbowfishes, males of this species lack the pronounced elongation of the posterior dorsal and anal fin rays. Males are more brightly coloured, larger, and deeper bodied than females.

#### **Distribution & Habitat**

Melanotaenia irianjaya are found primarily in river systems that drain into Bintuni Bay, in the southern Vogelkop Peninsular region. In 1982, they were collected by G. Allen and W. Tins from a tributary of the Kamundan River at Senopi Village on the north side of Bintuni Bay, and also by G. Allen and H. Bleher near the village of Fruata on the Bomberai Peninsula. In 2007 live specimens were collected from the Bintuni East River and the Tisbo River.

Habitats consist of rainforest streams ranging from slightly turbid and slow flowing over flat terrain to clear, moderately fast flowing through hills. Temperature and pH values range from 27-28°C and 7.3-7.8 respectively. The fish are found in areas with relatively few aquatic plants, over gravel or sand bottoms, often in the vicinity of submerged logs.

#### Remarks

M. irianjaya have been available into the aquarium hobby since 1983. In 2007, a number of collecting surveys were conducted by the Papuan National Marine and Fisheries Research, the Academy of Fishery Sorong, and the Institut de recherche pour le développement (IRD) Jakarta in a number of regions in West Papua. In the Bintuni Bay region they collected 332 rainbowfish specimens. Among them were specimens of *M. irianjaya*, which were taken back to Jakarta and the Sorong Fisheries Academy for breeding purposes.







# Melanotaenia iris

Allen, 1987 Strickland Rainbowfish

## **Species Summary**

*Melanotaenia iris* is most closely related to *M. goldiei*, a widely distributed species in southern New Guinea, but differs in colour pattern and having more soft rays in the second dorsal fin (17 to 20 vs. 12 to 17, usually 14 to 16) and more scales covering the suboperculum-preoperculum (about 30 to 40 vs. 15 to 25. The nature of the midlateral stripe also differs between the two species. It is continuous and uniformly broad along the middle of the side in *M. iris*, but in *M. goldiei* it is generally absent or very faint anteriorly over a space covering about 8–10 scales, the stripe then recommences below the soft dorsal fin origin, becoming broadest on the caudal peduncle.

Adult males are bluish on the upper back and white on the lower portion with a vivid dark blue stripe (about 2 scales wide) on the middle of the sides. There is also a series of narrow, red-orange stripes between each scale row on the upper and lower margin of the blue midlateral stripe and one in the middle of the stripe particularly prominent. Median fins dusky blue-grey with white outer margin; pelvic fins white; pectoral fins translucent. Live colours of the female are unknown.

## **Distribution & Habitat**

Inhabits the upper tributaries of the Strickland River system in southwestern Papua New Guinea. The Strickland River is a major tributary of the Fly River.

#### Remarks

Known only from five specimens collected by David Gwyther in 1984 from the Logatyu River, a mountain tributary of the Strickland River near Wankipe, Papua New Guinea. Presently unknown in the aquarium hobby. The species was named iris (Latin: goddess of the rainbow) with reference to the common appellation for the family to which it belongs.





# Melanotaenia japenensis

Allen and Cross, 1980 Japen Rainbowfish

## **Species Summary**

*Melanotaenia japenensis* have an overall mauve colouration with silvery reflections on the back and sides. There is a red-orange horizontal stripe between each scale row on the body. Males have red-orange dorsal, anal, and caudal fins. Growing to a length of around 11 cm, males are usually deeper bodied than females. *M. japenensis* is clearly derived from the same phyletic line which includes *M. affinis* and *M. vanheurni* of northern New Guinea.

These species possess similar colour patterns and have dorsal and anal soft fin ray counts which are relatively high for the genus. However, *M. japenensis* differs from *M. affinis* by having a higher anal ray count (26–28 vs. 18–24), and from *M. vanheurni* by having fewer soft dorsal rays (15–17 vs. 18–21 usually 19). In addition, the male holotype of *M. japenensis* has a deeper body compared to similar sized males of *M. vanheurni*.

## **Distribution & Habitat**

*M. japenensis* were collected near Serui on Japen Island (Yapen Island) on the north coast of West Papua. They were collected in rocky rainforest streams at lower elevations on the southern side of the island. Temperature and *p*H recorded at the collection sites were  $24-28^{\circ}$ C and 7.2-7.8.



*M. japenensis* is apparently restricted to Japen, a long (approximately 160 km), narrow island situated in the gulf (Teluk Sarera) on the north coast which isolates the Vogelkop Peninsula from the remainder of New Guinea. The island represents a continuation of a coastal mountain chain found on the nearby (30 km distance) New Guinea mainland and has a maximum elevation of 1500 m. Presumably speciation of *M. japenensis* has occurred in relatively recent times as a result of the separation of Japen from the mainland due to a post pleistocene rise in sea level.

#### Remarks

*M. japenensis* were first collected in May 1955 by M. Boeseman from the Leiden Museum. David Price who does missionary work on Japen Island collected specimens for his own aquarium, but is still unavailable in the general hobby. Named *japenensis* with reference to the Japen Island, the type locality and only known collection site for this species thus far.





### **Melanotaenia kamaka** Allen and Renyaan, 1996 Kamaka Rainbowfish

## **Species Summary**

*Melanotaenia kamaka* have a silver-blue colouration on the upper back, grading to silver white on lower half; upper half of body of males frequently flecked with silver; body scales with narrow dark outline, more intense on two midlateral scale rows; males can expand melanophores of these two rows, forming blue to blackish midlateral stripe on posterior part of body, including caudal peduncle; blue to blackish patch usually present, especially on mature males, between the upper rear corner of eye and region under the pectoral fin; first dorsal, pelvic, and anal fins whitish; second dorsal and caudal fins translucent with bluish suffusion; pectoral fins translucent; female fin colouration generally more diffuse and more translucent compared to males.

Besides the colour differences mentioned above, females often exhibit a diffuse midlateral stripe, about one and a half scales wide and extending from the eye to the base of the caudal fin. Fin shape differences between sexes are not as apparent as in most other members of the genus, but as in most *Melanotaenia* there is a pronounced difference in body depth. Males increase in body depth with advanced age. They may reach a maximum size of 8 cm, but are usually less than 6 cm.

#### **Distribution & Habitat**

This species is apparently restricted to Lake Kamakawaiar, the largest of three main lakes, and several smaller ones just inland from Triton Bay, West Papua. The Triton lakes are situated on the southern coast of West Papua, immediately east of the Bomberai Peninsula and about 50 km due east of the seaport of Kaimana. The lakes are surrounded by high limestone hills and lie just inland from Triton Bay. There are three main lakes: Kamakawaiar, Lakamora, and Aiwaso. Kamakawaiar (usually referred to as Kamaka) lies less then 5 km from the coast and is separated from the second lake, Lakamora, by a distance of about 7 kilometres. The third lake, Aiwaso, lies only a few hundred metres from Lakamora. The lakes do not appear to have any outlet streams and drainage is presumably subterranean. The following measurements were recorded in July 1995: water temperature 28.9°C; pH 8.0; and conductivity 220 µS/cm.

#### Remarks

Heiko Bleher collected this species in June 1995 together with Paola Pierucci and Patrick de Rham. The species was named kamaka, the name used by inhabitants of Triton Bay for Lake Kamakawaiar, the type locality.









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## Melanotaenia kokasensis

Allen, Unmack and Hadiaty, 2008 Kokas Rainbowfish

## **Species Summary**

Adult male *Melanotaenia kokasensis* have an overall pale bluegrey body colouration with a blackish midlateral band at the level of the upper pectoral-fin base. The midlateral band is more or less solid on the rear half of the body, but incomplete anteriorly where it is composed of darkened posterior scale margins. There are narrow orange stripes between each scale row on the upper half of body. Most of the scales on the upper two-thirds of the side have narrow greyish margins.

The upper portion of head is greyish while the lower half is white. A poorly defined stripe, about equal to the pupil in width runs from the rear edge of the eye to immediately above the pectoral-fin base where it merges with the midlateral band. The lower body is whitish with a broad, oblong bluish patch extending from just below the pectoral-fin base to above the middle of the anal fin.

The dorsal fins are mainly greyish-blue with a narrow white margin on the second dorsal fin. The caudal fin is whitish with a slightly dusky grey basal half and faint blackish dorsal and ventral margins. The anal fin is dusky yellow; pelvic fins bright yellow; pectoral fins translucent with small black spot on upper base and smaller silvery-white spot just above. The colour pattern of females is similar to that of the males except they are generally less vivid and the pelvic fins are whitish. Unlike most members of *Melanotaenia*, males and females are difficult to differentiate on the basis of body depth or other external features.

This species was named after the village of Kokas, which is the major landmark in the area.

## **Distribution & Habitat**

This species is currently known only from the type locality; a small stream flowing along a limestone creek bed through primary forest. The stream plunges down a steep 20 metre high ramp next to the sea into a mangrove-lined inlet near the village of Kokas on the northern Fakfak Peninsula. Specimens were located about one kilometre upstream, in a circular pool with an approximate diameter of 15–20 metres with a maximum depth of about 0.5 metre. This pool was situated only about 20 metres downstream from a series of limestone fissures that appears to be the stream's underground origin.

## Remarks

*Melanotaenia kokasensis* were collected from a small creek above a waterfall near Kokas, northern Fakfak Peninsula in West Papua by G. R. Allen and M. Ammer in 2008. This species is not currently available in the aquarium hobby.





## Melanotaenia lacustris Munro, 1964

Lake Kutubu Rainbowfish

## **Species Summary**

Depending on water conditions, captive environment and diet, *Melanotaenia lacustris* can display an array of different colours and patterns ranging from cobalt blue, steel blue, aquamarine through to lighter and darker shades of turquoise. When spawning the nape area changes to a bright orange or gold colouration. Males are more brightly coloured, larger, and with a body depth of 4–5 cm, much deeper bodied than females. They may reach a maximum size of 12 cm, but usually less than 10 cm.

Melanotaenia lacustris is closely related to Melanotaenia mubiensis. Large adults of the two species have very similar colouration. However, they differ in several features, including body depth, eye size, and modal fin ray counts. Adults of *M. mubiensis* are much more slender than those of *M. lacustris*; *M. mubiensis* had an average depth as percent of the SL of 38.4 compared to an average of 47.2 for *M. lacustris*. The eye diameter of *M. lacustris* is larger than the snout length, but in *M. mubiensis* it is shorter or equal to the snout length. Although the two species have overlapping counts for dorsal, anal, and pectoral fin rays, there are significant modal differences. *M.* 

*mubiensis* most frequently has 14 or 15 dorsal rays, 21 or 22 anal rays, and 14 pectoral rays compared to usual counts of 12 or 13, 18 or 19, and 15 respectively for *M. lacustris*.

## **Distribution & Habitat**

*Melanotaenia lacustris* have been collected in Lake Kutubu and the Soro River, which is the only outlet stream of the lake. The Soro River eventually flows into the Kikori River system. In late 1983 Gerald Allen, John Paska, and Barry Crockford collected around 40 live specimens. Only 4 fish (1  $\bigcirc$  and 3  $\bigcirc$ ) survived the journey back to Australia and a week after arriving in Melbourne the only male became infected with hook worm and subsequently died. The aquarium hobby had to wait another 3 years before Heiko Bleher was able to collect further live specimens. He returned in 1988 together with Gerald Allen and once more was able to bring back live specimens.

Lake Kutubu is a scenically beautiful crystal clear lake situated about 40 kilometres from Mendi, the main town of the Southern Highlands Province in central Papua New Guinea. Lake Kutubu was originally formed when debris and ash originating from a volcano, blocked a valley. It is the 2nd largest lake in PNG and its largest perched lake. Measuring approximately 19 km by 4 km at its widest point, it has a maximum depth of about 70 m. Water conditions recorded at the lake were a temperature range of 21 to  $25^{\circ}$ C and a *p*H of 8.5–9.0.







An exceptionally clear lake where, in contrast to most of PNG's inland waters. The Lake plays a significant role in the maintenance of biodiversity of the Kikori River basin and beyond. The lake's extraordinary level of fish endemicity (10 of the 14 fish species found within the Kikori drainage are endemic to the lake itself) exceeds that of any other lake in the entire New Guinea-Australian region. The Kikori drainage and the surrounding primary rainforest also support high levels of endemism and rare terrestrial fauna. Lake Kutubu provides the sole spawning, nursery and feeding grounds for the 10 species of endemic fish.

The water quality of the mainstream rivers of the Tagari-Hegigio and Lake Kutubu-Digimu-Mubi sub-basins are typical of other mainstream rivers in Papua New Guinea that are near neutral to mildly alkaline (*p*H 7.4 to 8.2) and calcium-bicarbonate dominated. These properties are indicative of water draining a limestone catchment area. The lower calcium concentration, alkalinity and hardness of the Ai'io River, which drains to the upper Hegigio River, probably reflect the predominantly volcanic and sedimentary terrain at this location. Water hardness in all rivers except the Ai'io River (30 mg/L CaCO<sub>3</sub>) is moderate to hard (60–180 mg/L CaCO<sub>3</sub>). Conductivity values are generally similar in all streams, with median values ranging between 167 and 267  $\mu$ S/cm.



#### Remarks

Australian Patrol Officer T. Terrell first collected this species in Lake Kutubu during 1955. He sent preserved specimens to Australian ichthyologist Ian Munro, who in 1964 described them as *Melanotaenia lacustris*. Ian Munro (1919–1994) worked with the CSIRO Division of Fisheries Research and was an early pioneer in the identification of Australian and New Guinea rainbowfishes. Not only was he a highly regarded ichthyologist but an accomplished aquarist as well and maintained many Australian and New Guinea rainbowfish species. During the 1960's he was maintaining New Guinea rainbowfish species such as *Melanotaenia papuae*, *M. sexlineata* and *M. goldiei* in captivity. Although at the time *M. papuae* and *M. sexlineata* had not been scientifically described. He later went on to publish "The Fishes of New Guinea" in 1967.





## Melanotaenia lakamora

Allen and Renyaan, 1996 Lakamora Rainbowfish

## **Species Summary**

*Melanotaenia lakamora* is a very attractive rainbowfish. The body colour of adult males is generally an overall mauve colouration except for silvery white on the breast and lower half of head. Four lateral scale rows on middle of the body are separated by bright orange stripes. They display a broad, blackish to dark blue, mid-lateral stripe, which is most intense from the eye to the pectoral fin region and on the caudal peduncle, one scale row wide anteriorly and occupying two scale rows posteriorly. The scales of the body have narrow dark margins, most evident on the lower half, particularly above the anal fin where several zigzag lines may be apparent.

The dorsal and anal fins are bright red. The pelvic fins are slightly orange to translucent while the caudal and pectoral fins are mainly translucent. Female specimens from Lake Lakamora are bronze on the upper half and whitish below with blackish mid-lateral stripe about 1–2 scales wide. They have narrow orange stripes along the upper and lower edge of the mid-lateral stripe. Fins are clear to translucent, except the second dorsal and anal fins, which have a pale orange colouration. Growing to a size of around 6 cm, the males are easily distinguished from females by their brighter colours and longer and more elongated dorsal fin rays.

During spawning males become intensely red with a white to light blue forehead stripe. A male specimen collected from Lake Aiwaso by Gerry Allen was pale mauve on the upper half and silvery white below with golden scale margins.

#### **Distribution & Habitat**

*Melanotaenia lakamora* have been collected from Lake Lakamora and Lake Aiwaso in the remote southern region of West Papua, immediately east of the Bomberai Peninsula and about 50 km due east of the seaport of Kaimana. The lakes are surrounded by steep forested hills and situated just inland from Triton Bay. There are three main lakes: Kamakawaiar, Lakamora, and Aiwaso collectively known as the Triton Lakes. Lake Aiwaso is roughly circular with a diameter of about 2.5 km and lies a few hundred metres from Lake Lakamora and separated by a 100 metre high ridge. Lake Lakamora is approximately 6–7 km long and 1–3 km wide.

#### Remarks

This species was named *lakamora*, with reference to Lake Lakamora, the type locality. Live specimens were collected from Lake Lakamora by Heiko Bleher for the aquarium hobby in 1995.





Dirk Godlinski



## Melanotaenia maccullochi

Ogilby, 1915 McCulloch's Rainbowfish

## **Species Summary**

*Melanotaenia maccullochi* were described in 1915 by J. D. Ogilby from two specimens collected from the Barron River, near Cairns in north Queensland, by Mr. A. Anderson. They were named after the ichthyologist, Allan Riverston McCulloch (1885-1925). It is therefore, according to recognised nomenclature rules, pronounced McCulloch - eye, not "mac - cul - lo'kee".

*Melanotaenia maccullochi* is another rainbowfish species that varies across its wide distributional range. Several geographically isolated populations are found in northern Queensland. Several distinct colour forms are known, which show marked variation in the intensity of the dark body stripes and markings on the dorsal and anal fins as well as differences in the colour of the 'spawning' stripe on the nape of males. This coloured nape is flashed on and off during spawning activities and may be white, yellow, orange or red.

Current genetic data separate *Melanotaenia maccullochi* into three groups, Burtons Creek, Etty Bay and Cape York populations. Further genetic and morphological studies may justify recognition of two or three separate species. Male specimens of the variety found between Cairns and Innisfail are easily recognised by the silvery-white or yellowish body colour and 6–8 reddish-brown stripes on sides. The dorsal and anal fins are orange-red with a lower black margin running along the body line. The caudal fin has a fan of orange-red colouration. Females are much less colourful, though some do show a hint of the males' coloration. Females of this variety tend to grow larger and have deeper bodies than males.

The variety found in the drainage division of the Jardine River are characterised by a series of fine black stripes on the sides, with black submarginal bands and white to yellowish margins on the dorsal and anal fins. Females generally have the stripes less defined. They are also a lot smaller than the other varieties, both in length and body depth. The population from the latter area is similar to those that occur in the southwestern lowlands of Papua New Guinea. The varieties found north of Cairns but south of the Jardine River are intermediate. However, the stripe pattern is plain and they do not show the orange-red colouration. An unusual blue coloured form has been collected in the Hope Vale region; a remote region situated 46 kilometres north of Cooktown

In 1988 a new colour variety was collected from a small shallow stream known as Burton Creek. Burton Creek is a spring-fed tributary in the Finniss River catchment. This variety has clear to yellowish dorsal and anal fins with bluish





edges above a black sub-marginal band. The body colour is silver to yellowish with a dark mid-lateral stripe and grows to a much smaller size than the other varieties. More recently (2007) another population was found in Tolmer Creek, a tributary of the Reynolds River in the Northern Territory. These are similar to the Burton Creek population.

Differences between the various populations are considerable and in all probability the different populations will be separated into distinct species at some later date. Therefore, for the serious rainbowfish breeder, it is very important to maintain each geographical population separately in captivity.

## **Distribution & Habitat**

Melanotaenia maccullochi occur as a number of isolated populations in southern New Guinea and northern Australia. In Australia, several isolated populations are known to exist in Queensland and the Northern Territory. The distribution in Queensland includes the coastal plains between Cairns and Innisfail. In this region they have been collected from the Barron, Mulgrave (Behana Creek), Russell (Harvey Creek), Johnstone and Moresby Rivers, Maria Creek, Hull River and the Murray/Tully Rivers. They are also found from the Daintree River north through Cooktown to the McIvor River and streams in the Hope Vale region (Black Creek). Another area where they are found is Cape York Peninsula, primarily in the Jardine River and its tributaries. They also occur at Cape Flattery and the Olive River and probably occur elsewhere along the east coast of Cape York Peninsula that has suitable habitat. Small isolated populations have also been found in two locations in the Northern Territory. The known New Guinean distribution encompasses the lower and middle sections of the Fly River westward to the Bensbach River.

Wild populations are still abundant in New Guinea and Cape York Peninsula, but the more southerly populations along the Queensland coast have declined due to habitat destruction. The extensive development of coastal plains has contributed to the demise of this species. It is now confined to a relatively few widely scattered locations and has long been absent in the Barron River, the site of its first capture. The Murray River and its floodplain lagoons represent the remaining habitat of the Cairns colour form.

*Melanotaenia maccullochi* are generally found in lowland swamps and small streams, usually in clear, moderately flowing streams, grassy wetland swamps and tannic stained ponds in sandy coastal floodplains. Often with ample cover in the form of log debris or aquatic vegetation. The water in these natural habitats is usually very soft and often tannin stained. A temperature range of 19° to 32°C and *p*H values of 5.5 to 7.0 have been recorded in their natural habitats.

## Biology

Very little is known about the biology of *Melanotaenia maccullochi* in their natural environment. Most information is mainly based on aquarium observations. They may reach a maximum size of 6 cm, but usually less than 4 cm.

*Melanotaenia maccullochi* are most likely aseasonal spawners, breeding continuously at intervals throughout the year. However, a peak in reproductive activity is usually during the early-wet season, from October to December. Strong sexual dimorphism is present in the species with males typically being brighter in colouration. Before spawning, a bright spawning' stripe is evident in the males. It runs from the tip of the mouth to the first dorsal fin on the dorsal surface of the fish.

Females produce between 20–30 eggs each day for several days. Eggs are attached by adhesive threads or tendrils to a range of submerged physical structures, including gravel substrates, woody debris, root masses, aquatic vegetation and submerged marginal (riparian) vegetation, which hide them from predators. The eggs are subject to desiccation if the water level drops or to dispersal if there is a flood. It will take around 8–9 days at 28° Celsius for the first young to appear. Larvae achieve a length of around 12 mm by 60 days and 2.5–3.0 cm in five months, when they become sexually mature. *M. maccullochi* is an opportunistic omnivore. The main food items are aquatic insects, algae and terrestrial insects. Their diet varies in relation to the habitat they occupy.

*Melanotaenia maccullochi* readily breeds in captivity. They generally spawn during the early morning hours, preceded by intense spawning activity of the male. The male presses against the side of the female and accompanied by heavy trembling of both fishes; eggs are expelled directly among the plants. The eggs are fairly large  $(1.5 \pm 0.5 \text{ mm in diameter})$ , light amber to yellowish in colour and hang by a fine thread. The fry emerges after 7 to 8 days, first hanging mostly near the surface, feeding on protozoans or dust-fine prepared food. When properly fed and maintained, the fry grow rapidly and become sexually mature at around four months of age.

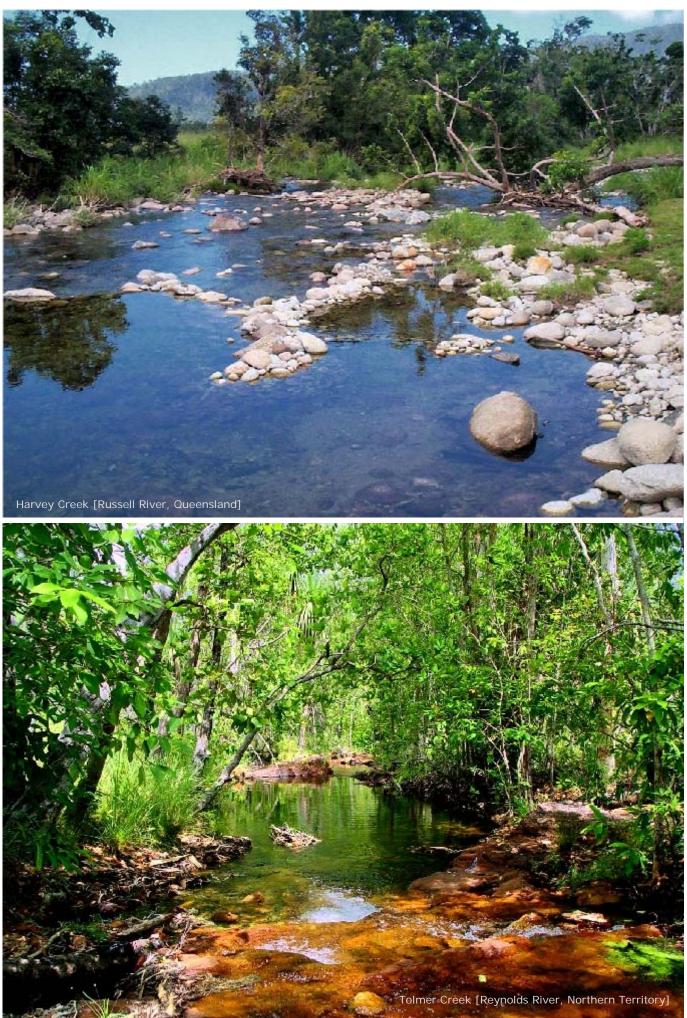
## Remarks

*Melanotaenia maccullochi* is one of the smaller species of rainbowfishes and have been a popular aquarium fish for many years. They were first introduced to the international aquarium hobby in 1934, when Amandus Rudel, a founding member of the Aquarium & Terrarium Society of Queensland, sent 12 specimens, collected by him near Cairns, to Fritz Mayer in Hamburg, Germany. Four arrived alive and developed into 2 pairs. They were one of the most popular aquarium fish from Australia. In the German aquarium magazine "Wochenschrift für Aquarien und Terrarienkunde" in May 1935, Fritz Mayer gave the first account of their breeding, which was translated by F. H. Stoye in Innes' "The Aquarium" in December 1936.

Through my correspondence with friends in all parts of the world, I have been able to instigate new imports. In this way I became acquainted with Mr. A. Rudel of Brisbane, Australia, who notified me December, 1934, that he was sending me twelve Melanotaenia maccullochi, collected by him near Cairns, northeastern Australia. Four arrived alive and developed into 2 pairs.

~ Fritz Mayer, Hamburg (1935)









Gunther Schmida







#### Melanotaenia maylandi Allen, 1982 Mayland's Rainbowfish

## **Species Summary**

Adult males of *Melanotaenia maylandi* are generally olive green or brownish dorsally and silvery white on the lower half. The upper back and sides often reflect bluish or mauve hues and there is a series of narrow oranges lines on the sides between each horizontal row of scales. There is also a diffused midlateral band extending from the upper corner of the opercula margin to the middle of the caudal fin base, often consisting of large blotches. The fins are translucent to light blue-grey except for a yellow anal fin. Males are more brightly coloured, larger, and deeper bodied than females. They may reach a maximum size of 10 cm. The species is named in honour of Hans Mayland a well-known German writer, photographer, and aquarist.

#### **Distribution & Habitat**

So far *Melanotaenia maylandi* have only been collected from a small creek about 2 km upstream from Danau Bira (Lake Holmes) in the lower Mamberamo system of West Papua. Lake Holmes is situated in the Mamberamo region of West Papua. It is a complex of three interconnected lakes lying at an altitude of about 430 metres above sea level and



set in the foothills of the van Wees Mountains, approximately 290 kilometres west of Jayapura, the capital city of West Papua. The lakes lie within a radius of 6–7 kilometres with the main lake having a length of approximately 4.5 kilometres and maximum width of about 2 kilometres. The lakes are drained by a small stream, which flows into the Mamberamo River at a point approximately 15 kilometres directly to the north. The lake and surrounding creeks are inhabited by 11 fish species, including one other rainbowfish, *Chilatherina bleheri*.

#### Remarks

This species was discovered by Heiko Bleher and Gerald Allen during a visit to West Papua in 1982. No live specimens have been collected for the aquarium hobby.





# Melanotaenia misoolensis Allen, 1982

Misool Rainbowfish

### **Species Summary**

*Melanotaenia misoolensis* was described from 23 specimens collected from Misool Island off the western extremity of New Guinea. Adult males are bluish with a bronze or golden sheen, and silvery white on the lower half. The upper back and sides often reflect bronze or golden sheen and there is a series of narrow oranges lines on the sides between each horizontal row of scales. There is also a diffused midlateral band extending from the upper corner of the opercula margin to the middle of the caudal fin base, often consisting of blotches. The fins are translucent to light yellow. Males grow to a length of around 6 cm and are usually deeper bodied than females.

They are closely related to *Melanotaenia catherinae* which is endemic to Waigeo Island, a large island lying approximately 160 km north of Misool. Both species are similar in colour; however, the mid-lateral stripe of *M. catherinae* is significantly wider, having a maximum width of about three scales compared with  $1\frac{1}{2}$  scales for *M. misoolensis*. Moreover, the midlateral stripe of *M. misoolensis* is nearly covered entirely by the pectoral fin, whereas it is broadly exposed (at least one scale row) above the pectoral fin of *M. catherinae*. In addition, the latter species lacks the dusky spot on the fin membrane behind the last dorsal spine and has a dusky soft dorsal fin which is often blackish in adult males. *M. misoolensis*, in contrast, has a dusky spot behind the last dorsal spine and the soft dorsal fin is yellowish-orange. The only meristic difference noted is related to counts for the soft anal rays. *M. misoolensis* usually has 22 to 25 rays compared with 19 to 21 rays for *M. catherinae*.

### **Distribution & Habitat**

*M. misoolensis* is currently only known from Misool Island. Misool Island is the second largest (approximately 90 x 38 km) of the four Raja Ampat Islands. It covers an area of about 2034 km<sup>2</sup> and is separated from the mainland by a distance of 32 kilometres. Its geographical remoteness from continental Papua and rugged, deeply bisected and heavily forested, predominantly limestone karst terrain make this one of the wildest and most visually stunning places in the entire Raja Ampat archipelago. To the north, a coastline of dense and deep mangroves shelter some villages located on the rare beaches fringed with coconut palms. To the east, a labyrinth of toadstool-shaped limestone islets and pinnacles, deeply undercut below the high-tide water surface and covered in luxuriant vegetation, spreads out into a turquoise sea.



*M. misoolensis* have been found in moderately fast-flowing clear-water streams running through primary rainforest. Other rainbowfishes have been collected from several other islands in the Raja Ampat group of islands: *M. batanta* (Batanta Island); *M. catherinae* (Batanta and Waigeo Islands); *M. fredericki* (Salawati Island) and *M. synergos* (Batanta Island).

### Remarks

In October 1948, a Dutchman by the name of Maurits Lieftinck collected some rainbowfishes from a tributary of the Wai Tama River near Fakal Village on Misool Island. The collection remained unstudied in the Zoological Museum of the University of Amsterdam in the Netherlands until officially described by Gerald R. Allen in 1982. The species was named *misoolensis* in reference to the type locality.

In 2000 Heiko Bleher collected what he described as two different species. They are currently known in the hobby as *Melanotaenia misoolensis* "Kasim" and *Melanotaenia misoolensis* "Ifaupan". However, genetic analyse has failed to find any difference between the two different varieties. *M. misoolensis* were also collected by Gerald Allen from the Wai Tama River in 2002.



Male ▲ Female ▼ "Ifaupan" variety. This variety was collected by Heiko Bleher on Misool island in 2000, and distributed into the aquarium hobby in 2001.







### Melanotaenia monticola Allen, 1980

Mountain Rainbowfish

### **Species Summary**

Melanotaenia monticola males generally have a lilac-blue to greenish coloured wash over the body, fading to silvery white on the chest and abdomen. Scales are edged with a coppery gold colouration. They have a very prominent black mid-lateral stripe extending from the eye to the caudal fin base. Females are similarly coloured but not quite as bright. When spawning, the colouration of the males becomes more intense. The upper part of the head and body become very dark and a vivid orange coloured nape band is flashed on and off on top of the head. They may reach a maximum size of 10 cm. Melanotaenia monticola is similar in general appearance to Melanotaenia mubiensis. The two species have been collected together. Besides differing in colour pattern they also have differences in soft dorsal and anal fin rays; M. monticola has 15 to 17 dorsal rays and 18 to 21 (usually 19 or 20) anal rays. M. mubiensis most frequently has 14 or 15 dorsal rays and 21 or 22 anal rays.

# **Distribution & Habitat**

*Melanotaenia monticola* are found in a relatively small area of the Southern Highlands of Papua New Guinea between Mendi and Lake Kutubu. They have been collected from small headwaters tributaries in the Purari River system, including Omei Creek, a tributary of the Ka River, 15-km south of Mendi, and streams near Pimaga (about 13-km southeast of Lake Kutubu). They generally inhabit the slower flowing regions of swift flowing streams. They are usually found along grassy banks, or around sub-surface vegetation, submerged logs, and branches. Water temperature recorded from their natural habitat was 18°C and *p*H 7.6. However, temperatures have been known to drop to 16°C.

#### Remarks

*Melanotaenia monticola* were initially collected in September 1979 by Gerald Allen and Brian Parkinson from Omei Creek. Allen gave the species the scientific name of *'monticola'* (Latin: mountain dwellers) with reference to the mountainous terrain of the type locality.

Live specimens were collected by Barry Crockford from Omei Creek and introduced to the Australian hobby in 1983. During the mid 1990s, eggs were collected from a small stream near Lake Kutubu and brought back into Australia where they were subsequently hatched, reared and distributed in the hobby. This is another rainbowfish that has been in the aquarium hobby for a long time but has never been widely available.





## Melanotaenia mubiensis Allen, 1996 Mubi Rainbowfish

# **Species Summary**

Melanotaenia mubiensis are blue-green on the upper half of head and back, frequently with golden sheen anteriorly, lower side whitish. Dark blue midlateral band extending from rear edge of eye to base of caudal fin, about one scale row wide anteriorly and two scales wide on caudal peduncle. 6-7 pale orange stripes between each horizontal scale row on upper half of body. Yellowish stripe, one scale wide, immediately below dark midlateral band, from pectoral fin base to level of middle anal rays. Pupil sized orange spot on upper part of operculum. Iris of eye golden-yellow. First dorsal fin pale green or bluish. Second dorsal and anal fins dusky blackish, except bluish basally. Caudal fin bluish to translucent, upper and lower edge narrowly dusky. Pelvis fins translucent with dusky anterior edge. Pectoral fins mainly translucent. Female colouration generally less intense and all fins mainly translucent or bluish. Males are generally deeper bodied and have more elongated, somewhat pointed shape posteriorly on the soft dorsal and anal fin rays.

*Melanotaenia mubiensis* is most closely related to *Melanotaenia lacustris* from Lake Kutubu. Large adults of the two species have very similar colouration. However, they differ in several features, including body depth, eye size, and modal fin ray counts. Adults of *M. mubiensis* are much more slender than those of *M. lacustris*; the four largest male types of *M. mubiensis* had an average depth as percent of the SL of 38.4

compared to an average of 47.2 for *M. lacustris*. The eye diameter of *M. lacustris* is larger than the snout length, but in *M. mubiensis* it is shorter or equal to the snout length. Although the two species have overlapping counts for dorsal, anal, and pectoral fin rays, there are significant modal differences. *M. mubiensis* most frequently has 14 or 15 dorsal rays, 21 or 22 anal rays, and 14 pectoral rays compared to usual counts of 12 or 13, 18 or 19, and 15 respectively for *M. lacustris*. *M. mubiensis* is also similar in general appearance to *M. monticola*, which occurs in the middle Kikori and adjacent Purari River system. The two species have been collected together. Besides differing in colour pattern they also have differences in soft dorsal and anal fin rays; *M. monticola* has 15 to 17 dorsal rays and 18 to 21 (usually 19 or 20) anal rays.

# **Distribution & Habitat**

*Melanotaenia mubiensis* was collected from a relatively small section of the middle Kikori drainage system, spanning a distance of approximately 20 km, between elevations of about 380 and 400 metres above mean sea level. All sites were tributaries of the Mubi River, one of the primary mountain tributaries of the Kikori, and the outlet for Lake Kutubu, which lies approximately 70 km farther upstream from the collecting sites. The habitat consists of narrow, crystal clear streams in closed-canopy forest, flowing through limestone hills. The holotype was collected from a spectacular series of sinkholes linked by short tunnels to the main channel of the Mubi River.

### Remarks

This species is named '*mubiensis*' with reference to the general locality where the type specimens were collected. Currently no live specimens have been collected for the aquarium hobby.





# Melanotaenia nigrans

(Richardson, 1843) Blackbanded Rainbowfish

Atherina nigrans Richardson, 1843 Atherinichthys nigrans Gunther, 1861 Nematocentris nigra Gunther, 1861 Melanotaenia nigrans Gill, 1863 Zantecla pusilla Castelnau, 1873 Nematocentris pusilla Macleay, 1882 Melanotaenia pusilla Ogilby, 1896

# **Species Summary**

*Melanotaenia nigrans* is the type species of the genus *Melanotaenia*. They were collected by John Gilbert in 1840, from the King River, near Victoria Settlement in the Northern Territory. John Gilbert later perished somewhere in the Australian wilderness with the famous German explorer, Ludwig Leichhardt. A single specimen ended up in the British Museum of Natural History in London where John Richardson described it in 1843 as a new species of hardyhead named *Atherina nigrans*. The differences between *Atherina nigrans* and the real hardyheads were sufficient enough for the American Thomas Gill to create the genus *Melanotaenia* for this lone species in 1862, still within the family Atherinidae. The genus name being

inspired by the typical black mid-lateral band. The next step was the creation of a subfamily Melanotaeniinae by Gill in 1894 to stress the differences with the hardyheads even more. It took another 70 years however, before Ian Munro elevated them to full family status of Melanotaeniidae in 1964. A full generic classification of the rainbowfishes followed in 1980 by Gerald Allen from the Western Australian Museum.

Melanotaenia nigrans can be recognised by a rather slender body than most other rainbowfishes. They may reach a maximum size of 12 cm, but are usually less than 7 cm. Colouration includes a continuous distinct black band in the mid-lateral position. Above the lateral line the colouration is generally an olive-grey, brownish colouration and silverywhite below. Colour variations can be found in the different geographically located populations. Specimens from the Kimberley region have several lines of dots below the lateral line and red in the fins. Blue colouration in the body and fins is also found in some populations. Colour variability in rainbowfishes has been a source of confusion to both aquarists and taxonomists studying their life history. Colour appears to vary from population to population as well as within a population, particularly during different stages of the fishes' lifespan. This colour variability is related to age, sex, stress, habitat conditions and spawning.





Bruce Hansen





Male and female rainbowfishes usually have different colours and this adds further difficulties to species recognition. In males, the spines of the first dorsal are usually extended and may lie well past the origin of the second dorsal when not erect. The posterior rays of the second dorsal and anal fins are extended caudally and may extend past the origin of the caudal fin. In females, the first dorsal spines are short, not reaching the origin of the second dorsal. The posterior rays of the anal and second dorsal fin are not extended. The spines and outer rays of the ventral fins of some males are also extended and may reach past the vent and the origin of the anal fin.

### **Distribution & Habitat**

*Melanotaenia nigrans* has a discontinuous distribution across northern Australia, from the Kimberley region in Western Australia, across the northern part of the Northern Territory to Cape York Peninsula in northern Queensland, including a number of offshore islands such as Groote Eylandt in the Gulf of Carpentaria and some islands in the Torres Strait. Discontinuous distribution of fish species appears to be a feature of much of the northern Australian fish fauna. A species found in one river system may not necessarily exist in an adjoining system. Most specimens have been collected from the lower reaches of streams within about 50 km of the coast; however, they also occur in several upland areas including above waterfalls barriers. They have been collected in the upper South Alligator River some 130 km upstream.

In Western Australia, *Melanotaenia nigrans* have so far only been collected in Dominic Creek during 1990 and Pago Creek in 1997; however, they probably occur elsewhere. Dominic and Pago Creeks are small isolated streams between the Drysdale and King Edward Rivers. Pago Creek in the next stream north of Dominic Creek, but it is probably not the correct name and is most likely an unnamed stream. It was just called Pago Creek because it is near Pago Mission (Drysdale River Mission) site which was abandoned in 1939. A new mission was established at Kalumburu, about 30 km south of Pago. Unlike other *M. nigrans* varieties, the Western Australian specimens have a row of reddish-orange dots below the black stripe and may yet prove to be genetically different.

*Melanotaenia nigrans* have been found in a variety of freshwater environments but seem to prefer slow-flowing clear water streams, billabongs, and swamps with abundant aquatic vegetation. A temperature range of 19–35°C has been recorded in their natural environment; pH 5.0 to 8.1; hardness and alkalinity levels are usually below 50 mg/L CaCO<sub>3</sub> and conductivity 4 to 180 µS/cm. *M. nigrans* is most frequently found in clear waters with sandy substrates, followed by rocks, leaves and mud. They are usually found around sub-surface vegetation, submerged logs, or branches. They are often found in streams with *M. trifasciata*, *M. australis* and/or *M. splendida inornata*.

#### Biology

In their natural environment *Melanotaenia nigrans* is an omnivore feeding opportunistically across substrates and in surface waters, with possibly less emphasis on mid-water areas. The main food items are aquatic insects, algae and terrestrial insects. The diet varies in relation to the habitat they occupy. In the mainchannel waterbodies they eat mainly aquatic insects, with small amounts of terrestrial insects, plant material and algae. In perennial streams, algae and terrestrial plant material are less important, while aquatic insects and, to a lesser extent, oligochaetes and microcrustaceans, are consumed. The diet in the lowland sandy creekbeds had much larger algal and terrestrial insect components. Specimens examined from the floodplains feed mainly on aquatic arachnids and aquatic insects, and a small amount of algae.

Spawning is possibly continuous, with a few eggs laid at a time, or opportunistic whenever conditions are favourable. Small (less than 20 mm) juvenile fish have been collected in all seasons. Therefore it is difficult to define their breeding season. However, a peak in reproductive activity was recorded during the early-wet season (December-March). *Melanotaenia nigrans* appears to breed in small streams that contain deep shaded pools with roots and submerged vegetation around the edges.

Spawned eggs are adhesive, negatively buoyant in freshwater and average 1.00-1.08 mm in diameter, are usually clear to light amber in colour and hang by a fine thread. Usually one to three eggs are deposited at a time, during which time 50–70 eggs can be produced. In one study of the ovaries of *M. nigrans*, the number of eggs ranged from 220 to 500 (mean = 344); egg diameters were not measured. These fish were not mature, so the numbers only indicate developing eggs within the ovary, not how many might actually be shed during spawning.

Survival of eggs is reduced by predation activity of the parents. Many of the eggs are eaten before and after they attach to the waterplants or other objects.

### Remarks

From the very beginning of its introduction to the aquarium hobby, until around the mid 1960s, *Melanotaenia duboulayi* was mistakenly identified in both Australian and International hobby publications as *Melanotaenia nigrans*. The real *M. nigrans* never entered the International hobby until around 1976, when specimens were sent to Europe. Even today it is still being incorrectly identified on some Internet web sites. Some sites also incorrectly refer the name '*nigrans*' to *Melanotaenia australis*.





# Melanotaenia ogilbyi

Weber, 1910 Ogilby's Rainbowfish

*Nematocentris ogilbyi* Munro, 1967 *Melanotaenia ogilbyi* Weber, 1910

### **Species Summary**

*Melanotaenia ogilbyi* is a poorly known species which is known only on the basis of 7 specimens collected from pandanus swamps on the lower Lorentz River in western New Guinea. These were collected during the Dutch New Guinea Expedition of 1907 and no further specimens have been collected.

*Melanotaenia ogilbyi* have a bluish body colouration on the sides and back, becoming whitish ventrally. Each horizontal scale row on the blue portion of body is separated by narrow dark stripe. Fins are bluish to translucent, anterior edge of first dorsal fin and outer portions of second dorsal and anal fins dusky in males. Pelvis and pectoral fins translucent. *Melanotaenia ogilbyi* may reach a maximum size of 10 cm, but usually less than 8 cm.

This species belongs to the *Maccullochi spp.* group of rainbowfishes. The main differences between this species and its nearest relatives *M. papuae*, *M. maccullochi*, and *M. sexlineata* are related to colour pattern.

## **Distribution & Habitat**

Found in tributaries of the Lorentz River, and streams north of Timika, West Papua. This species was first collected from the Noord-Fluss (North River) by the Dutch explorer Hendrikus Albertus Lorentz during an expedition in 1907. The Noord-Fluss River was later renamed the Lorentz River after Dr. Lorentz. Lorentz participated in three expeditions to Dutch New Guinea, the present-day West Papua (western) portion of the island of New Guinea. The first expedition was in 1903, led by A. Wichmann. Lorentz led expeditions in 1907 and 1909-1910.

The Lorentz River is one of the large slow-flowing rivers of the southern lowlands of New Guinea draining into the Arafura Sea near Agats. The river passes through several major wetland habitats including freshwater swamp forest, peat swamp forest and mangrove forest.

In 1995 Gerald Allen collected specimens in the vicinity of Timika. *Melanotaenia ogilbyi* are often found with *Melanotaenia goldiei* and *Pseudomugil novaeguineae*.

#### Remarks

This species is not currently available in the international aquarium hobby, although they have been maintained locally.





## Melanotaenia oktediensis Allen and Cross, 1980

Oktedi Rainbowfish

### **Species Summary**

*Melanotaenia oktediensis* were initially collected by C.R. Boyden from the Ok Tedi River at Tabubil, Papua New Guinea during the Cambridge Expedition in 1974. In 1975, Dr. Tyson Roberts collected them from the lower portion of Karamonge Creek, a tributary of the Ok Tedi River during an ichthyological survey of the Fly River and mistakenly identified them as *Melanotaenia vanheurni*, a species found in the Mamberamo basin, northern New Guinea. After examining specimens in the Zoologisch Museum, Amsterdam and the American Museum of Natural History, New York, Allen and Cross recognised them as a distinct species and named them *M. oktediensis* after the Ok Tedi River. *M. oktediensis* have a body colouration that is coppery brown above a prominent mid-lateral line and pale mauve to white below. They may reach a maximum size of 12 cm, but usually less than 10 cm.

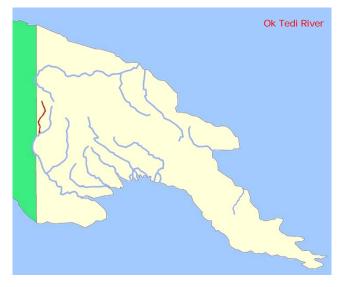
### **Distribution & Habitat**

*M. oktediensis* have been found in tributaries of the Ok Tedi River (Ok Menga, Ok Ma, Karamonge Creek), which is one of the main headwater streams of the upper Fly River system. They are rather uncommon in the main river, which is mostly populated by *Melanotaenia splendida rubrostriata*.

The tributaries of the Ok Tedi River are cool, clear, relatively fast flowing rainforest streams with rocky bottoms. Water conditions reported from this habitat are temperature  $17-24^{\circ}$  Celsius, *p*H 7.3–8.7, hardness 20 to 80 ppm and a high dissolved oxygen content of 6.9–10.0 mg/L.

### Remarks

Live specimens were introduced to the Australian hobby in 1982 and were spread to Europe and Northern America, but they have never been readily available.







# Melanotaenia papuae

Allen, 1981 Papuan Rainbowfish

### **Species Summary**

The overall body colouration of *Melanotaenia papuae* is generally olive-green above the mid-lateral line and silvery white below. A prominent red spot is frequently present on the upper part of operculum. A pair of prominent black lines is present at the upper and lower edge of the lateral line and they continue down the lower half of the body. On the posterior half of the body the colours of these lines become reddish and/or purplish in males and yellow to pale orange in females. The colours and markings of the females are generally less brilliant than those of the males. Named *M. papuae* after its type locality Papua, the southern portion of Papua New Guinea. *M. papuae* may reach a maximum size of 8 cm, but usually less than 6 cm SL.

#### **Distribution & Habitat**

The full extent of its geographic range remains undetermined. Most collections have been within a 35 km radius of Port Moresby, Papua New Guinea. They are generally found in rainforest and coastal freshwater streams where they are often found together with *Melanotaenia goldiei*. Temperature and *p*H reported from these habitats were  $25-33^{\circ}$  Celsius and *p*H 7.3–7.8. From the collections around the Port Moresby District,

Gerry Allen found that *Melanotaenia goldiei* prefers more inland streams which are faster flowing, less turbid, and slightly cooler whereas *M. papuae* is more often found closer to the coast in quieter conditions. However, in some locations such as the Sogeri Plateau, they are found together, although at this locality *M. goldiei* was far more abundant.

#### Remarks

*Melanotaenia papuae* were one of the earlier New Guinea Rainbowfishes to be maintained in the aquarium hobby and were available in Australia as early as 1961. In 1964, Ian Munro catalogued some specimens from Port Moresby as paratypes of *Melanotaenia sexlineata* in "Additions to the fish fauna of New Guinea". However, there was some confusion regarding their correct identification as among Munro's paratypes were some specimens later identified as *Melanotaenia papuae* by Gerry Allen in 1981. *Melanotaenia papuae* were collected from tributaries of the Laloki, Brown and Goldie Rivers in 1970 and 1971. Gerald Allen and Brian Parkinson collected about 30 live specimens in 1978 from a small creek at Mount Diamond, about 15 miles west of Port Moresby. Specimens were also collected from the tributaries of the Laloki River in 1981.

Some of these specimens were brought back to Australia and small numbers were distributed in the hobby. They were a very popular species but these days they seem to have been forgotten and are rarely seen.





## Melanotaenia parkinsoni Allen, 1980

Parkinson's Rainbowfish

### **Species Summary**

*Melanotaenia parkinsoni* have an overall silvery body colour with a rosy chest and narrow orange stripes between the scale rows. The fins of adult males are bright orange with dark edges. Mature males can be magnificent and usually have sweeping blotchy orange colouration on the posterior area of the body, giving an overall patchy appearance. Females are rather plain in comparison. Some males also develop large extended dorsal and anal fins with a ragged appearance. Another colour variety exists with bright yellow stripes or blotches instead of orange. Both colour forms have been collected from the Kemp Welsh River. Sometimes the stripes or blotches fuse to form an overall colouration, which covers the entire posterior half of the body. May reach a maximum size of 15 cm, but usually less than 12 cm.

#### **Distribution & Habitat**

*Melanotaenia parkinsoni* was first collected in October 1978 by Gerry Allen and were found along the southern coast of eastern Papua New Guinea between the Kemp Welsh River and Milne Bay. They were collected in only a few interspersed locations, but in all probability, could be widespread within this region.



Gerry Allen collected specimens from two small tributaries of the Kemp Welsh River a short distance inland from the coast and about 75 kilometres southeast of Port Moresby, and from a small stream about 3 kilometres west of Alotau at the extreme eastern tip of mainland Papua New Guinea. Most of the original specimens collected were taken from a small stream that was mainly dry except for the occasional isolated pool. The stream was situated in grassy plains habitat with patchy rainforest immediately adjacent to the creek. The temperature and *p*H range recorded from this habitat was 27– 30° Celsius and 7.6–7.8.

#### Remarks

Named in honour of Brian Parkinson, a regular companion of Allen's on numerous collecting trips to Papua New Guinea. Live specimens were brought back to Australia in 1978 by Gerald Allen and distributed in the aquarium hobby. Heiko Bleher collected a yellowish coloured form in the 1990s and distributed them in the European hobby. Males have a yellowish body and a pinkish breast.





Norbert Grunwald



This form was originally collected by Heiko Bleher in the late 1990s from south-eastern Papua New Guinea and have been maintained by Marcel Dielen in Belgium. They are known in the European hobby as either *M. parkinsoni* cf. "Orient" or *M. parkinsoni* cf. "South East". ▲ Male ▼ Female





## Melanotaenia parva Allen, 1990

Lake Kurumoi Rainbowfish

### **Species Summary**

Gerry Allen reported that there appeared to be two colour forms of *Melanotaenia parva* in Lake Kurumoi; males being either bluish to mauve with a black midlateral band or silver with red speckling and narrow red lines between each scale row on the side of the body. Fins of both varieties were red. Young males have a rosy-mauve body colour that turns more and more bright orange-red as they grow. The new species was reported to be very small (hence, the species name "parva", meaning small in Latin). However, *Melanotaenia parva* may reach a maximum size of 9–10 cm.

### **Distribution & Habitat**

*Melanotaenia parva* is currently known only from Lake Kurumoi, a small and isolated lake situated on the isthmus that links the Vogelkop Peninsula with the rest of New Guinea. Lake Kurumoi is part of the Yakati River system. They were collected along the shoreline of the lake amongst dense aquatic vegetation.

### Remarks

In March and April 1989 Gerald Allen collected a number of rainbows at various localities in the Vogelkop Peninsula. Among these specimens were *Melanotaenia angfa* from the Yakati River and *Melanotaenia parva* from Lake Kurumoi. Live specimens of *M. parva* were collected by Heiko Bleher in 1999 and introduced to the aquarium hobby.

In 2007 surveys were conducted by the Papuan National Marine and Fisheries Research, the Academy of Fishery Sorong, and the Institute of Research for Development of France in five bioregions of West Papua. Fifteen species of rainbowfishes were collected during these expeditions. Based on the collections in the Bintuni Bay region 332 rainbowfish specimens were collected. Among them were *M. irianjaya*, *M. parva* and *M. fredericki*. Five species were taken live to Jakarta for breeding while the rest were sent to the Sorong Fisheries Academy. These included *M. parva* and *M. irianjaya* plus possibly three new species. *M. angfa* were not found!

A number of freshwater lakes were identified as high priority for conservation by Conservation International in 1999. This was because they are important areas of fish and crayfish endemicity. These are Danau Bira (Lake Holmes), Lake Sentani, Lake Kamaka, Paniai Lakes, Ayamaru Lakes, Lake Kurumoi, Lake Yamur, Lake Lakamora and Lake Aiwaso. Specific rainbowfishes that are considered threatened in West Papua are: *Chilatherina bleheri, Chilatherina sentaniensis, Glossolepis incisus, Melanotaenia arfakensis, Melanotaenia boesemani* and *Melanotaenia parva* (Conservation International 2002).







# Melanotaenia pierucciae

Allen and Renyaan, 1996 Pierucci's Rainbowfish

### **Species Summary**

*Melanotaenia pierucciae* have a body colour of mauve or purplish on the upper back with a bronze sheen, white or very pale mauve on the lower half (except a large violet patch may be evident just behind pectoral fin). The body scales have a narrow dark outline, which is more intense on the ventral half, particularly those above anal fin where scales often have greatly expanded black margins. There is a broad, blackish to dark blue, mid-lateral stripe between the eye and base of caudal fin, occupying about two horizontal scale rows, except interrupted on middle of side for about 6–7 vertical scale rows (scales in this area have a bronze sheen). First dorsal fin white; second dorsal fin bluish; anal fin dusky grey to whitish; caudal and pectoral fins translucent; dorsal and ventral edge of caudal fin narrowly black on basal half. Female fin colouration generally less intense and more translucent compared to males.

Males have a more intense colour pattern, particularly during spawning and display a whitish-green forehead stripe. The species exhibits fin shape differences, typical for the genus, in which males have a longer first dorsal fin and the posterior profiles of the second dorsal and anal fins, are somewhat elongated and pointed. Males have a deeper body as well and may reach a maximum size of 8 cm, but usually less than 6 cm.

## **Distribution & Habitat**

This species is known only from Werfyang Creek, which flows into the north-western end of Lake Kamakawaiar. The habitat lies about 1–2 km upstream from the lake and is separated from it by a scenic 20 m high waterfall. The fish was common in the main creek (about 4–5 m wide and 1–2 m deep) as well as a small tributary, both flowing through dense rainforest. The water was crystal clear and flowing rapidly in Werfyang Creek, but slowly in the small tributary.

The Triton lakes are situated on the southern coast of West Papua, immediately east of the Bomberai Peninsula and about 50 km due east of the seaport of Kaimana. The lakes are surrounded by high limestone hills and lie just inland from Triton Bay. There are three main lakes: Kamakawaiar, Lakamora, and Aiwaso. Kamakawaiar (usually referred to as Kamaka) lies less then 5 km from the coast and is separated from the second lake, Lakamora, by a distance of about 7 kilometres. The third lake, Aiwaso, lies only a few hundred metres from Lakamora. The lakes do not appear to have any outlet streams and drainage is presumably subterranean.

### Remarks

Heiko Bleher collected these species in June 1995 together with Paola Pierucci and Patrick de Rham. The species is named in honour of Miss Paola Pierucci, who together with Heiko Bleher discovered the species.







# Melanotaenia pimaensis

Allen, 1981 Pima River Rainbowfish

#### **Species Summary**

*Melanotaenia pimaensis* have a body colour of olive to brown dorsally; silvery blue on sides with narrow orange lines between each scale row. Adults have a prominent blackish midlateral band. May reach a maximum size of 9 cm, but usually less than 6 cm.

## **Distribution & Habitat**

First discovered in 1980 from the Pima River (Oima River on some maps) at the junction with Tua River, Purari River system, Papua New Guinea. In 1991 further collections were made in the Pio River. They were collected from small slow-flowing tributaries in shallow depths of less than one metre. The streams were relatively open and exposed to sunlight, although bordered by rainforest in some areas. Temperature ranged from 19°C in the deeper flowing sections to 25°C in the exposed shallows. A *p*H of 7.8 was recorded. About half of the specimens were collected from moderately flowing turbid water and the remainder from a crystal-clear backwater with minimal flow. One stream was inhabited by a hardyhead (*Craterocephalus*) and another rainbowfish, *Chilatherina campsi*.

#### Remarks

Named "pimaensis" in reference to the Pima River type locality. This species was first collected by Brian Parkinson and Gerald Allen in 1980. Live specimens were collected and brought back to Australia, but they failed to become established in the aquarium hobby.



# Melanotaenia praecox

(Weber and de Beaufort, 1922) Neon Rainbowfish

#### **Species Summary**

*Melanotaenia praecox* are bright neon blue with red dorsal, anal, and caudal fins. May reach a maximum size of 8 cm, but usually less than 6 cm SL. Heiko Bleher reported in Aqua Geõgraphia, "... males have red-edged fins while the fins of females are pure yellow". However, my original females had red fins and succeeding generations produced red-finned females, although at times they can appear faintly orange coloured. There are however, aquarium stocks that have yellow-finned females.

#### **Distribution & Habitat**

*Melanotaenia praecox* was initially collected by the Dutch naturalist W. C. van Heurn in 1910 from a tributary of the *Mamberamo River* in West Papua. They have been collected from the Mamberamo and Wapoga River systems. Gerry Allen collected specimens in 1991 from two small localities near the airstrips at Dabra and Iritoi on the edge of the Mamberamo Plains. Gerry Allen (1998) also found them in small creeks and swampy ponds near Siewa, in the Tirawiwa River system, a remote area of northern West Papua about 200 km west of the Mamberamo River basin. These specimens differ from the Mamberamo representatives in having a red stripe between each scale row and males do not get so deep-bodied.

#### Remarks

*Melanotaenia praecox* were originally introduced to the aquarium hobby by Charles Nishihira around 1991 who had obtained wild-caught specimens from a local aquarist in Jayapura. Heiko Bleher collected wild-caught specimens in 1993. Further live specimens were collected by Gary Lange and Johannes Graf in 2008 and taken back to Europe and the United States. Fish from these collections have been bred and distributed in the aquarium hobby.

Breeding trials with *Melanotaenia praecox* were conducted over a six-month period involving six replicates on viability regarding to number of eggs, fertilisation rate, hatching rate, length of incubation period, and survival rate in a seven days rearing period. While observations of growth rate, survival rate, and male percentage were conducted until 6 months old in three replicates. The results reported the average of number of eggs was 27 eggs/spawning, fertilisation rate was 92.93%; hatching rate was 98.18%; length of incubation period was 8 days (7–9), and survival rate in seven days rearing period was 89.45%, respectively. The growth rate up to 6 months rearing period was 3 cm, while the survival rate was 94 (92–96)%, and the male percentage was 42.58%.





▲ The above specimen was collected near the village of Pagai [Taritatu River]. The Tariku River (previously known as the Rouffaer River) in the west flows eastward and the Taritatu River (previously known as the Idenburg River) in the east flows roughly westward. They meet in the Meervlakte Basin to form the main Mamberamo River.









Hans Booij





# Melanotaenia pygmaea

Allen, 1978 Pygmy Rainbowfish

#### **Species Summary**

*Melanotaenia pygmaea* males display a brilliant colouration consisting of a metallic sky-blue back, a blackish midlateral stripe, and pale yellow fins. Males are more brightly coloured, larger, and much deeper bodied than females. Spawning males' display a yellowish body colour below the lateral line and a brightly coloured rustic-red band running from the first dorsal fin to the upper lip and extending down the breast. Males may reach a maximum size of 7 cm, but females are usually less than 5 cm SL.

#### **Distribution & Habitat**

*Melanotaenia pygmaea* was first discovered by Gerald Allen in 1974 in the tributaries of the Prince Regent River in the Kimberley region of Western Australia. The Prince Regent River is currently the only known habitat of *M. pygmaea*. They have been collected from only two small tributaries; Cascade Creek and Youwanjela Creek, where they were found around sub-surface vegetation, submerged logs, or branches. Most collections have been from Cascade Creek, situated approximately 20 km upstream. The Prince Regent River is situated in northeast Western Australia and flows into the Indian Ocean. The river rises 50 kilometres from the coast at an elevation of about 800 metres and drops through a rugged gorge. At the head of the river lies a broad plateau averaging over 700 metres above sea level. The coastline is deeply indented by a number of drowned river valleys. The following creeks flow into the Prince Regent River: Pitta Creek, Gundarara Creek, Womarama Creek, Youwanjela Creek, Cascade Creek and Quail Creek. The upper reaches of the river and creeks are mostly seasonal, with some permanent pools varying in depth up to several metres during the dry season.

#### Remarks

In 1992 and 1994, live collections were made and descendants from these collections now form the basis of the current stock available in the aquarium hobby today.







# Melanotaenia rubripinnis

Allen and Renyaan, 1998 Red-finned Rainbowfish

## **Species Summary**

*Melanotaenia rubripinnis* have a body colouration that is red on the back, mainly white on lower half of body except for blue smudge above anterior part of anal fin. A black stripe runs from the rear edge of the eye to the pectoral fin base, continuing as a blue-black mid-lateral stripe to the base of caudal fin. The mid-lateral stripe is more or less uniform in width, bordered by a narrow blue stripe above and broader yellow stripe below. The dorsal, anal and caudal fins are red-orange. The pectoral and pelvic fins are translucent. Specimens from more open habitats are brown above and white below with a black mid-lateral stripe that is narrowly bordered above and below by a light blue stripe.

Occasional specimens from mountain streams are uniformly bluish except for a white breast region and black stripe between the eye and pectoral-fin base. Males have a more intense colour pattern, especially specimens from dense lowland rainforest streams. May reach a maximum size of 12 cm, but usually less than 10 cm SL.

Melanotaenia rubripinnis belongs to the 'affinis speciesgroup' of northern New Guinea, which includes M. affinis, M. japenensis, M. maylandi and M. vanheurni, and is most closely related to M. vanheurni from the Mamberamo River system of West Papua. However, it differs in colour and modal number of soft dorsal rays (usually less than 18 in *M. rubripinnis* and more than 18 in *M. vanheurni*). In addition, *M. rubripinnis* has fewer cheek scales (range 17–26, average 19.9 vs. range 19–36, average 29.2).

### **Distribution & Habitat**

Melanotaenia rubripinnis is currently known only from the Wapoga River system of northern New Guinea. It was relatively common in a variety of habitats including tanninstained creeks in lowland rainforest, larger streams in more open situations, and mountain tributaries to an elevation of about 400 metres above sea level. It is found in quiet shaded pools, as well as sunlit sections of larger streams and relatively fast-flowing mountain streams. They are found together with *Chilatherina alleni* and *Glossolepis leggetti*. *Melanotaenia rubripinnis* and *Chilatherina alleni* generally co-occur in the same streams and are also sometimes found with *Glossolepis leggetti* in lowlands immediately adjacent to foothills.

#### Remarks

*Melanotaenia rubripinnis* was described from 51 specimens collected in 1998. It was named "*rubripinnis*" (Latin: with red fins), with reference to the characteristic fin colouration. Currently, no live specimens have been collected for the aquarium hobby.





# Melanotaenia sexlineata

(Munro, 1964) Fly River Rainbowfish

*Nematocentris sexlineatus* Munro, 1964 *Melanotaenia sexlineata* Allen, 1980

### Species Summary

Melanotaenia sexlineata are a very attractive species. They have a lemon-greenish body colouration with 5-8 narrow black stripes, with a darker mid-lateral band. This species belongs to the "Maccullochi Species Group" of rainbowfishes. The main differences between this species and its nearest relatives, M. papuae, M. maccullochi, and M. ogilbyi are related to colour pattern. They have a deeper body than M. papuae or M. maccullochi and males often show red coloured lips giving the appearance that they are wearing lipstick. Females are a subdued version of the male with lesser-defined markings. There appears to be a number of different colour forms. Heiko Bleher collected a different colour variety with a blue coloured back and orange fins from the upper Fly River catchment, apparently somewhere along the Kiunga-Tabubil Road in 2003. In 2007, Mark Allen collected specimens with an iridescent turquoise and golden sheen. M. sexlineata may reach a maximum size of 8 cm, but usually less than 7 cm SL.

# **Distribution & Habitat**

*Melanotaenia sexlineata* are presently only known from the Fly River and its tributaries. Most specimens have been collected from creeks immediately north of Kiunga along the Kiunga-Tabubil Road. The full extent of its distribution is unknown. They have been found in small, shallow tannic stained streams. Readings of pH 6.1–7.4 and 25°C have been reported from some collection sites.

In October-December 1975, Tyson R. Roberts in his fish survey of the Fly River in Papua New Guinea (Roberts, 1978) collected "*M. sexlineata*" from 2 locations:

(1) Small tributaries and mainstream upper Fly River 1-2 km upstream from mouth of Elevala River.

(2) Lake Herbert Hoover (Lake Bosset), Wam River (which drains Lake Herbert Hoover), and swampy lagoons along the main stream of the middle Fly River.

The eight specimens from (1) above, plus three additional ones comprise the type specimens (Munro, 1964). The seven additional specimens reported from Lake Bosset (2) were actually *M. maccullochi*. In addition, seven paratypes of *M. sexlineata* from the Port Moresby district were re-indentified as *M. papuae*.





In 1982 Maunsell and Partners collected *M. sexlineata* from the Membok village, Binge River, 10 km from confluence with middle Fly River. Then in 2005, a fish survey by the Ok Tedi Mining Company collected them from 4 sites in the middle (2) and upper (2) Fly River.

#### Remarks

*Melanotaenia sexlineata* were initially discovered in 1937 by Stuart Campbell in an upper tributary of the Fly River, Papua New Guinea. However, they were not scientifically described until 1964 when Australian ichthyologist, Ian Munro named them *Nematocentris sexlineatus*. In a later review of the rainbowfish group (Allen, 1980) the name was changed to *Melanotaenia sexlineata*.

In October 1978, Gerald Allen and Brian Parkinson collected what they thought were *M. sexlineata* from a small creek at Mount Diamond, about 25 km west of Port Moresby. The stream consisted of a series of small disconnected pools only a few centimetres deep, due to the dry season. They collected about 30 specimens, which Allen found out later were actually *M. papuae*. They also collected more (*M. papuae*) in the Laloki River. These were initially distributed in the Australian hobby as *Melanotaenia sexlineata*. However, further research on these and newly collected specimens by Allen resulted in the recognition of the new species *Melanotaenia papuae*. Then in

1982 Gerald Allen brought live specimens of *M. sexlineata* to Australia that he collected from the upper Fly River where they were subsequently bred and distributed in the Australian aquarium hobby. Heiko Bleher collected *M. sexlineata* in the early 1980s and took them to Europe. Since then there has been a number of live collections and *M. sexlineata* is relatively freely available in the aquarium hobby.











# Melanotaenia solata

Taylor, 1964 Northern Rainbowfish

# **Species Summary**

This species was described by William R. Taylor in 1964 from specimens collected in 1948 from Groote Eylandt, Bickerton Island, and a creek near Yirrkala. The name "solata" is from solatus Latin, meaning sunburned. It is given to this species of rainbowfish in reference to the characteristic golden yellow life colours. Taylor described them as a species of Melanotaenia with a rather slender, compressed body; with complete dentition, with poorest developed in upper jaw; with a rather faint brown lateral body band and with numerous, characteristic, golden yellow life stripes through brown band as well as along the body. Large adults with diffuse dark band and about ten brilliant golden longitudinal stripes on each side; basal half of caudal fin bright yellow; bronze bar behind eye crossing preopercle and diffusing downward on opercle; belly and bases of second dorsal and anal fin pinkish; the inter-radial membranes paler outward; interradial membranes of first dorsal fin red. In specimens from Yirrkala, the dorsal and anal fins red; caudal fin yellowish orange; pelvic fins deep red; lower side bluish silvery; scale centres on side generally golden bronze; they form about five longitudinal rows, the lowermost of which is approximately on a level with the ventral surface of the caudal peduncle.

Following a scientific review of the rainbowfish family by Gerald R. Allen (1980) it was considered that Melanotaenia solata fell within the range of Nematocentris australis with regard to colour pattern, morphometrics and meristics, and in this review these two species were considered as one and were placed in the large "splendida" group as a sub-species, and named Melanotaenia splendida australis. However, earlier Allen (1978) remarked that Melanotaenia australis and Melanotaenia splendida inornata were so closely related that he was tempted to consider the latter a subspecies of australis. Morphologically or meristically there is little that tells them apart, the biggest difference is that Melanotaenia splendida inornata tends to be deeper bodied and seems to grow a little larger. Later, Allen et al. (2002) distinguished Melanotaenia solata from Melanotaenia australis on the basis of the genetic results of McGuigan et al. (2000).

"The differentiation of *M. s. australis* lineages in this study could (i) indicate the presence of two distinct species, (ii) be the result of introgression of the Northern Territory population with sympatric *M. nigrans* or *M. exquisita*, or (iii) reflect retention of ancestral polymorphisms. This third possibility is unlikely because the polymorphisms would need to be present in the ancestor of clades A, B and C. To distinguish between hypotheses (i) and (ii), *M. s. australis*, *M. nigrans* and *M. exquisita* would need to be characterised





molecularly (mtDNA and nuclear) and morphologically from across their geographical range. This would also help to determine the cause of the polyphyly in cytochrome b of Northern Territory *M. s. australis*. If hypothesis (i) is true, then the name *M. solata* (Taylor 1964) could be applied to the Northern Territory populations following re-description. Irrespective of the situation in the Northern Territory, West Australian populations of *M. s. australis* should be accorded species status." ~ McGuigan *et al.* (2000).

"However, the status of *M. solata* is questionable based primarily on the mtDNA analysis of the unusual population from upper South Alligator River which is introgressed with *M. nigrans* (Zhu *et al.*, 1994; McGuigan *et al.*, 2000). Therefore, it is probably better recognised as a synonym of *M. s. inornata* until further work is conducted, especially given the recognition that hybridization may be the cause of at least some of the odd morphotypes observed." (P. J. Unmack 2009, *pers. comm.*)

#### **Distribution & Habitat**

*Melanotaenia solata* are confined primarily to Arnhem Land, Northern Territory between the South Alligator and Walker rivers. They are also found on the larger offshore islands of the Gulf of Carpentaria including Groote Eyland and Bickerton Island. They are a stream dwelling rainbowfish mainly found around sub-surface vegetation, submerged logs, or branches in small tributary streams, but can also occur in swamps and lagoons. They generally form small groups at or near the surface of deeper pools in stream habitats, especially where there is aquatic vegetation. Their natural environment is subjected to seasonal variations with water temperature, pH, and hardness levels varying considerably. There is often a large fluctuation in water conditions between the dry and wet seasons.

#### Remarks

A rainbowfish fitting the description by Allen *et al.* (2002) of *Melanotaenia solata* has been reported from the Howard River system near Darwin (Pidgeon, 2003). In the aquarium hobby another rainbowfish from the Blackmore River in the Northern Territory is also often called *Melanotaenia solata*.

However, despite the research that has been undertaken to date, the specific status and distribution of Melanotaenia solata still remains unclear. Colour variability in rainbowfishes has been a source of confusion to both aquarists and taxonomists studying their life history. Populations of almost every river system they occupy have their own distinctive body colour and pattern. Colour can also vary considerably within stream populations in the same river system. Rainbowfishes at one end of a river system can look very different from rainbowfishes at the other end of the river system. This colour variability is often related to habitat conditions. Consequently, until more scientific research has been completed on the M. solata complex, specific names based on the locality where each variety is found is best used by rainbowfish enthusiasts to identify the different varieties, e.g., Melanotaenia sp. (Kambolgie Creek, South Alligator River).





# Melanotaenia splendida

(Peters, 1866) Eastern Rainbowfish

### **Species Summary**

*Melanotaenia splendida* is by far the most widespread of any rainbowfish species, occurring across western and central southern New Guinea and northern Australia from the Adelaide River in the Northern Territory to Deepwater Creek a small coastal stream located between the cities of Bundaberg and Gladstone on the east coast of Queensland. They also occur throughout most rivers in central Australia as well as the Paroo and Warrego Rivers in Murray-Darling system. The "splendida" rainbowfishes are currently a widely distributed group comprising four subspecies:

Melanotaenia splendida inornata Melanotaenia splendida rubrostriata Melanotaenia splendida splendida Melanotaenia splendida tatei

*Melanotaenia splendida* were originally collected from the Fitzroy River in central Queensland and scientifically described as *Nemacentrus splendida* in 1866. Gerald Allen's revision of the family Melanotaeniidae in 1980 places them under their current name. The different subspecies of *M. splendida* are not easily identified in relation to each other as they display a great variation of colours and markings. Principal visual differences are body depth and colour pattern, which is variable depending on location and natural environment. At the same time, body form within each subspecies is variably and appears to be related to habitat conditions, which can sometimes make correct identification difficult.

Populations of almost every river system they occupy have their own distinctive body colour and pattern. Colour variability in rainbowfishes has been a source of confusion to both aquarists and taxonomists studying their life history. Colour appears to vary from population to population as well as within a population, particularly during different stages of the fishes' lifespan. This colour variability is related to age, sex, stress, habitat conditions and spawning. Geographic distribution is very helpful; if you know where they were collected you can generally make a confident identification. Consequently, specific names usually based on the locality where each is found are used by rainbowfish enthusiasts to identify each variety. Where populations need to be identified, they are often done by inclusion of a form or population identifier in brackets following the species name e.g., Melanotaenia splendida (Burdekin River).

Whether or not *Melanotaenia splendida* is truly a distinct species or subspecies complex is a matter of on-going debate. Ever since Carolus Linnaeus founded the modern system of classifying species in the mid-18th century, taxonomists have argued over just what exactly species and subspecies are. In general, fishes evolve into different species and subspecies after becoming geographically isolated from others, adapting to their different environments, and changing over time through the process of natural selection. Geographic populations of *M. splendida* have been isolated from each other for perhaps thousands of years. They have gradually evolved physical adaptations that reflect their habitat. Some biologists classify *M. splendida* as separate subspecies because they are visibly different. Others say they are genetically the same as other *M. splendida* subspecies and differ only because of environmental circumstances. The traditional view of subspecies is morphological variants distinguishable at the level of the population where 75% or more of the individuals of the populations of one subspecies can be distinguished from those of other subspecies.

## Biology & Ecology

Not a lot is known about the biology or ecology of *M. splendida* in their natural environments. Most information is mainly based on aquarium observations. In captivity they can reach a maximum size of 12–15 cm, but are usually less than 8 cm. Males are more brightly coloured, larger, and much deeper bodied than females. Generally, the larger males can usually be identified from the elongation of posterior rays in the second dorsal and anal fins. Females and juveniles have plain silvery bodies and fins that are either translucent or only faintly coloured compared to the brighter colours of males. Sexual maturity occurs at about 3–4 cm for both sexes.

The main components of their natural diet are algae, aquatic insects, terrestrial insects and microcrustaceans. The algal component consists mainly of green filamentous species. A variety of aquatic insects are eaten; the main identifiable species being chironomid larvae and pupae, and coleopterans. The main terrestrial insects were formicids (ants) and the main microcrustaceans were cladocerans. Traces of hydrophytes, oligochaetes, gastropods, arachnids, macrocrustaceans, teleosts, terrestrial plants, detritus and inorganic material were also found in the stomachs. In the pools and riffles that enter the floodplain in the wet season they feed mainly on non-aquatic insect forms such as winged diptera and ants.

In their natural environment *M. splendida* has a prolonged spawning period with a peak of spawning activity in pre-flood and flood periods, although individuals in spawning condition and juveniles may be found throughout the year. Spawning during the wet season (November to April), when the inundation of streams and floodplains ensures an expanded habitat (in area and diversity) and a greater array and abundance of food. In contrast, spawning peaks during the dry season (May to October) ensures that larvae are produced during a period of relatively stable environmental conditions. This strategy increases the chances of some eggs surviving. Increased stream flow may result in conditions unfavourable for reproduction (i. e., physical removal of eggs, larvae and spawning substrate).

In the main, *M. splendida* will breed when environmental conditions ensure maximum fertilisation and larval survival. They usually spawn small numbers of eggs over a large area in slow-flowing waters and the backwaters of flooded areas. The presence of extensive spawning substrate enables them to 'spread the risk' from predators. The eggs are attached by adhesive threads to aquatic plants and other objects in the water, which hide them from predators.



The eggs, however, are subject to desiccation if the water level drops or to dispersal if there is a flood.

*Melanotaenia splendida* subsp. *inornata* are reported to migrate upstream at the onset of the breeding season, which corresponds to the start of water flow after the dry season. They spawn for an extended period during the early-wet season, and their gonads are developing for the next year's spawning by the late-wet-early-dry season. In a number of gonads examined, the number of eggs ranged from 70 to 370.

Under aquarium conditions, pre-spawning behaviour usually occurs in the morning and may continue for up to an hour before spawning takes place. During this period the colours in both sexes become more intense, but to a lesser extent in the female. In males, the edges of the fins and the chequering of the caudal and second dorsal fins become black. The nape may darken to black in some but not all fish. The midlateral stripe is prominent, extending from the origin of the pectoral fin to the caudal peduncle. Near the tail, two shorter dark lines appear above and below the midlateral stripe. The normal orange-yellow longitudinal stripes become a more intense colour in both sexes. The males have an iridescent purple sheen in light. The operculum has a glowing red spot in both sexes. The pectorals do not become coloured in either males or females. In the females, the midlateral stripe darkens before spawning but the stripe is not as long as in males. The chequered appearance of the second dorsal fin becomes more noticeable and the colour of all the fins darkens except in the case of the pectorals.

Chasing follows immediately after the colour change. The males chase the females, swim below them and brush their vent area with erect dorsal fins or butt them in the vent region or in the area of the pectoral fins. The males frequently tremble as they swim below the females. They display erected fins as they swim beside or at right angles in front of the females. During the pre-spawning chasing and display, the males can become quite aggressive and nip the females if the latter do not show interest in the display. In the final phase of pre-spawning behaviour, the pair swim with their bodies parallel, sinking and rising and then remain in one place with heads touching and their bodies vibrating rapidly. Eggs and sperm are then expelled amongst the plants or spawning medium.

Large females (>50 mm TL) produce more than 100 eggs per day at the peak of their spawning. Smaller females (30-35 mm TL), which were only just sexually mature shed fewer eggs, 20-30 per day and do not spawn each day. Accurate counting of the eggs is difficult as the male disperses the eggs rapidly by the swishing of his tail. Two females were once observed to lay more than 1700 eggs within a single one-week spawning period. The number of eggs shed by a single female is directly related to the size of the female with large females spawning from 40-250 eggs. Females usually only spawn once each day; however, males will often spawn with more than one female in one day. In captivity, with limited area and artificial substrate, females may spawn all their eggs at the same time. The eggs of *M. splendida* at fertilisation are similar in appearance to those of other rainbowfishes. All are spherical, with a number of adhesive filaments, 3-8 mm in length, arising from a small area of the chorion at the animal pole. Spawned eggs, which range in size from 0.93 to 1.24 mm in diameter, are adhesive, negatively buoyant in freshwater and are usually clear to light amber in colour. The eggs hatch after an incubation period of four to nine days depending on temperature. Temperature is one of the major factors that influences the embryonic period for rainbowfishes. Average embryonic period is about 5 days at 28°C.

The average larval length of M. *splendida* at hatching ranges from 2 to 4 mm, which is similar to other rainbowfish species. Hatched larvae are well developed and competent swimmers. Growth rates of the larvae are initially slow, with little variation until around 7 to 14 days. Growth is directly related to the initial absorption of the yolk sac and the provided larval diet. After that period growth rates increased. As the larvae increased in age, the variation in length between individuals also increased.

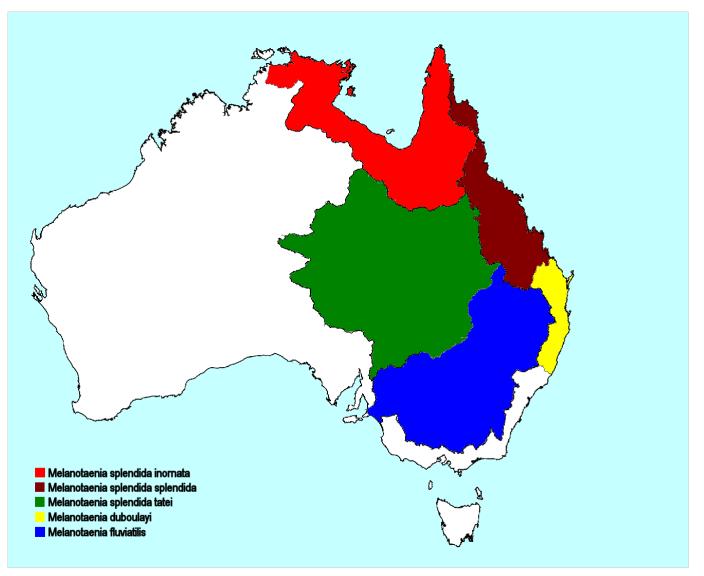
The continued growth and development of the fry will vary from one hobbyist to another and is largely conditional upon captive conditions such as temperature, water quality, and feeding regime. Under aquarium conditions increased temperature generally results in higher growth rates.  $28 \pm 1^{\circ}$ C is considered the most effective and safe temperature for optimum growth rate. At this temperature range, *M. splendida* are relatively fast growing with sexual differences beginning to appear between 9 and 12 weeks after hatching.

Food is an important factor affecting growth, especially in the early larval stages. Research has found that diet strongly affects not only fecundity but also the biochemical make-up of eggs and sperm as well as the growth rate and survival of larvae. The preferred size of food for larval fishes increases as mouth size and feeding competency increase. Providing natural 'green-water' (phytoplankton) with resident zooplankton as food for the newly hatched fish has several advantages. The larvae are easily able to switch to different sized food, a feature not present when feeding foods such as rotifers or brineshrimp. Green water also enables the zooplankton to feed on resident algae and microbes, thus retaining their nutritional value for greater periods of time. In addition, a varied diet may affect the growth of rainbowfishes positively.

### Remarks

Because of the great variation in colours and body forms, *Melanotaenia splendida* should be bred within their own localised populations. Regardless of their various colour patterns, at this point of time, they are all believed to belong to the same species and are capable and willing to breed together if permitted to do so. The serious hobbyist intent on maintaining pure lines must keep each population in separate aquariums. Unless this is done, members of the different subspecies or populations will interbreed and complicate future breeding programs and identification.





# Melanotaenia splendida — Distribution Map

*Melanotaenia splendida* subsp. *inornata* inhabit the river systems of the Northern Territory and Queensland, which flow into the Arafura Sea and Gulf of Carpentaria – from the Adelaide River to Cape York Peninsula, extending down the east coast to around the Lockhart and Stewart Rivers.

*Melanotaenia splendida* subsp. *rubrostriata* are widely distributed in southern New Guinea between Etna Bay in West Papua and the Central Province of Papua New Guinea. The Kikori River was the previous eastern limit of distribution, but recent surveys indicate that they are more widespread; having been collected in the Sapoi River in the Lakekamu Basin. The Sapoi River drainage is located approximately 150 km northwest of Port Moresby. They have also been found on the Aru Islands. However, their full distribution is unknown.

*Melanotaenia splendida* subsp. *splendida* are found in streams east of the Great Dividing Range along the coast of Queensland from Deepwater Creek north to Scrubby Creek, just south of the Lockhart River. *Melanotaenia splendida* subsp. *tatei* is widespread and abundant in the larger rivers of the Lake Eyre Basin and the Western Plateau of the Northern Territory. This species has only recently been identified from the Murray-Darling Basin, where it is recorded from the arid rivers in the north-western basin. It is found only in the Paroo and Warrego rivers, and hybrids with Murray-Darling rainbowfish have been identified in the lowermost Warrego River and the Darling River from around the Bogan River down to at least Menindee.

*Melanotaenia duboulayi* inhabits the coastal drainages east of the Great Dividing Range from the Hastings River on the mid northern coast of New South Wales to Baffle Creek in southern Queensland.

*Melanotaenia fluviatilis* is the most southerly ranging rainbowfish in Australia. Their distribution covers the Murray-Darling River system in Queensland, New South Wales, Victoria, and South Australia.





# Melanotaenia splendida subsp. inornata

(Castelnau, 1875) Chequered Rainbowfish

*Aida inornata* Castelnau, 1875 *Aristeus cavifrons* Macleay, 1882 *Rhombatractus cavifrons* Ogilbyi, 1896 *Aidapora carteri* Whitley, 1935 *Melanotaenia maculata* Allen, 1978 *Melanotaenia splendida inornata* Allen, 1980

### **Species Summary**

*Melanotaenia splendida* subsp. *inornata* were first described in 1875 as *Aida inornata*. During the 1950–60's they were very popular with native fish keepers in Australia. They were then scientifically known as *Aidapora carteri* and commonly known as "Carter's Sunfish". They did not become known in the international hobby until around the mid-1970s when they went on displayed at the Berlin Aquarium in Germany. At that point of time they were known as *Melanotaenia maculata* (Allen, 1978). Gerald Allen's revision of the family Melanotaeniidae in 1980 places them under their current name.

Generally the body colour is olivaceous to yellowish with white breast. Scales on side of body with purplish sheen. Mid-lateral stripe deep yellowish anteriorly, and bluishgreen or brownish-green on caudal peduncle. Other body stripes yellow or red. An orange or yellow spot on opercula. Dorsal, caudal and anal fins red and yellow chequered or orange-yellow with bright red spots on their membranes, with faint black edge.

### **Distribution & Habitat**

Melanotaenia splendida subsp. inornata inhabit the river systems of the Northern Territory and Queensland, which flow into the Arafura Sea and Gulf of Carpentaria from Darwin to Cape York Peninsula, extending down the east coast to around the Lockhart and Stewart Rivers. The Adelaide River is the furthest west that *M. s. inornata* has been recorded. They are frequently found in company with other rainbowfish species.

*M. s. inornata* are tropical fish and are found in almost every kind of freshwater habitat, from slow-moving streams, wetland swamps, lagoons and clear flowing rivers. They are generally found in waters with moderately thick vegetation. A temperature range of 10–40° Celsius; *p*H 4.6–8.5 and conductivity 2–220  $\mu$ S/cm, has been recorded in their natural environment.





This wide range of water conditions matches the wide distribution of the species. However, survival rates for *M. s. inornata* are known to decline sharply when the water temperature is high and will often die at temperatures above  $36^{\circ}$ C. Such increases in temperature are common in tropical waterbodies of Australia during the late dry season.

















Flying Fox Creek [Roper River, Northern Territory]





# Melanotaenia splendida subsp. rubrostriata

(Ramsay and Ogilby, 1886) Red-striped Rainbowfish

Nematocentris rubrostriatus Ramsay & Ogilby, 1886 Aristeus loriae Perugia, 1894 Rhombatractus loriae Ogilby, 1896 Rhombatractus rubrostriatus Ogilby, 1896 Rhombatractus patoti Weber, 1907 Melanotaenia maculata Weber, 1908 Melanotaenia dumasi Weber, 1913 Melanotaenia rubrosriatus Weber, 1913 Nematocentris rubrosriatus Weber, 1913 Anisocentrus rubrostriatus Regan, 1914 Amneris rubrostriata Whitley, 1935 Nematocentris maculata Munro, 1967 Melanotaenia splendida rubrostriata Allen, 1980

# **Species Summary**

*Melanotaenia splendida rubrostriata* have a basic body colouration of overall pale bluish-green, grading to white on the lower sides. Each horizontal scale row is separated by a narrow orange to pink stripe. The membranes between the rays of the second dorsal and anal fin are red. *Melanotaenia splendida rubrostriata* may reach a maximum size (TL) of 16 cm, but usually less than 12 cm, with a body depth of 6–8 cm. Males are more brightly coloured, larger, and much deeper bodied than females.

# **Distribution & Habitat**

Melanotaenia splendida rubrostriata were initially collected during the 1880s from the Strickland River, Papua New Guinea. They are widely distributed in southern New Guinea between Etna Bay in West Papua and the Central Province of Papua New Guinea. The Kikori River was the previous eastern limit of distribution, but recent surveys indicate that they are more widespread; having been collected in the Sapoi River in the Lakekamu Basin. The Sapoi River drainage is located approximately 150 km northwest of Port Moresby. They have also been found on the Aru Islands. Melanotaenia splendida rubrostriata inhabit freshwater creeks and rivers along lowland coastal plains. They are usually found around sub-surface vegetation, submerged logs, or branches. Temperature and pH recorded in their natural habitats range from 24-33°C and 5.6-7.5.

### Remarks

*Melanotaenia splendida rubrostriata* were one of the earlier New Guinea rainbowfishes to be introduced to the aquarium hobby. They first appeared in the Australian hobby around 1959. Live specimens were also collected by Gerald Allen during the period 1978-1982. This was another rainbowfish that fell out of favour with hobbyists as the number of new species arrived from New Guinea, and much of the captive stock disappeared.







Preliminary genetic studies (P. J. Unmack 2009, *pers. comm.*) have revealed significant genetic variation between *Melanotaenia* splendida rubrostriata and other geographically distinct populations of *Melanotaenia splendida* subspecies in northern Australia that warrant taxonomical separation at the species level.







# Melanotaenia splendida subsp. splendida

(Peters, 1866) Eastern Rainbowfish

Nematocentris splendida Peters, 1866 Strabo nigrofasciatus Kner & Steindachner, 1867 Aristeus fitzroyensis Castelnau, 1878 Aristeus rufescens Macleay, 1881 Melanotaenia nigrofasciata Ogilby, 1896 Rhombatractus fitzroyensis Ogilby, 1896 Rhombatractus rufescens Ogilby, 1896 Melanotaenia splendida splendida Allen, 1980

# **Species Summary**

*Melanotaenia splendida* subsp. *splendida* were originally collected from the Fitzroy River in central Queensland and scientifically described as *Nemacentrus splendida* in 1866. The basic body colouration is overall pale bluish-green, olivaceous to yellowish, grading to white on the lower sides. Each horizontal scale row is separated by a narrow orange to reddish stripe. The scales on the side of the body usually have a bluish-green, yellowish-red or purplish sheen. The mid-lateral stripe can be faded black to deep yellowish anteriorly, and bluish-green or brownish-green on the caudal peduncle. Other body stripes can be yellow, green, blue or red. There is usually an orange or yellow spot on the opercula. The dorsal, caudal and anal fins can be red and yellow chequered or orange-yellow with bright red spots on their membranes, with faint black edges. Other forms can have a blue-green body with yellow-green fins, with dark flecks and a dark border. However, colour is extremely variable and will depend upon the mood of the fish, water conditions and diet. Females and juveniles have plain silvery bodies and fins that are either translucent or only faintly coloured compared to the brighter colours of males.

Genetic studies beginning in the mid 1990's (Zhu *et al.* 1994) revealed the existence of significant genetic variation between populations of *M. s. splendida* that occur in the upland streams of north Queensland. In particular, these studies highlighted the degree of isolation of upland populations from the lowland populations. Subsequent genetic research (McGuigan et al. 2000) suggested that at least some of these species are unusual variants of *Melanotaenia splendida* - or populations displaying geness that have traits of more than one species. As a direct result of some of this research, the Utchee Creek Rainbowfish (*Melanotaenia utcheensis*) was described as a new species in 2000, with populations known from Utchee, Fisher, Rankin and Short Creeks in the North and South Johnstone River catchments (McGuigan 2001).







Wallaby Creek [Annan River, Queensland]



- Martinese



Rainbowfish from upstream sections of the Burdekin River have long been considered to be a distinct species by rainbowfish enthusiasts, and are known in the hobby as the Burdekin Rainbowfish (Running River or "zigzag" form). This form is believed to also be present in other tributaries draining the Paluma Range, notably the Fanning River.

There are other informally recognised forms of "*splendida*" such as the Davies Creek Rainbowfish, Kuranda Reds and Mena Creek Rainbowfish. However, despite the research that has been undertaken to date, the specific status and distribution of *M. s. splendida* still remains unclear.

### **Distribution & Habitat**

*M. s. splendida* are found in streams east of the Great Dividing Range along the coast of Queensland from Deepwater Creek a small coastal stream located between the cities of Bundaberg and Gladstone extending up the east coast to around the Lockhart and Stewart Rivers on Cape York Peninsula. Scrubby Creek, just south of the Lockhart River, appears to be the northernmost location for this species.

*M. s. splendida* are usually abundant in almost every kind of freshwater habitat, from slow-moving streams, swamps, lakes and clear flowing rivers. They are most abundant in open reaches of zero to low flow containing abundant instream vegetation and cover, and an intact riparian zone. They are less abundant in riffle/rapid habitats.

Abundance varies significantly over the seasons; being greatest after the wet season (which enhances recruitment) but will decline greatly as flow decreases during drought condition.

*M. s. splendida* are frequently found in company with *M. maccullochi, M. trifasciata, Cairnsichthys rhombosomoides,* and *Pseudomugil* species. Their natural environment is subjected to seasonal variations with water temperature (12–36°C), pH (5.0–9.2), and hardness levels varying considerably. This wide range of water conditions matches the wide distribution of the species.

*Melanotaenia duboulayi* inhabits the coastal drainages of the east coast of northern New South Wales from the Macleay River region to Baffle Creek north of the Bundaberg region in Queensland. It may be that these two species live sympatrically in some locations. Natural hybrids of *M. splendida* and *M. duboulayi* have been reported from Deepwater and Mullett Creeks.

### Remarks

*M. s. splendida* were introduced to the international hobby in an article by Paul V. Loiselle in the March, 1970 edition of "The Aquarium" magazine under the name *Nematocentris splendida*.

















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# Melanotaenia splendida subsp. tatei

(Zietz, 1896) Desert Rainbowfish

*Nematocentris tatei* Zietz, 1896 *Nematocentris winneckei* Zietz, 1896 *Rhombatractus winneckei* Ogilby, 1896 *Melanotaenia splendida tatei* Allen, 1980

### Species Summary

*Melanotaenia splendida* subsp. *tatei* is a small, laterally compressed fish. They may reach a maximum size of 10 cm, but usually less than 8 cm SL. The eyes are large and positioned towards the top of the head, and the mouth is moderately large, oblique and upturned. There are two dorsal fins separated by a small gap, with the first shortbased and the second long-based. There is a long-based anal fin and the tail is moderately forked. Two colour forms exist; in one form males have a purple body with yellow-green fins, with dark flecks and a dark border. The other form has a blue-green body with similar colouration on their fins. During spawning the belly of the male turns bright pink. Colour varies depending upon the mood of the fish, water conditions and diet.



Females and juveniles have plain silvery bodies with clear fins. Males are usually more brightly coloured with pale stripes along the sides, larger, and much deeper bodied than females.

*M. s. tatei* was originally named *Nematocentris tatei* by Zietz in 1896 after Ralph Tate (1840-1901), a geologist and botanist who was on the 1894 Horn Expedition when this species was first collected.

### **Distribution & Habitat**

M. s. tatei is widespread and abundant in the larger rivers of the Lake Eyre Basin and the Western Plateau of the Northern Territory. This species has only recently been identified from the Murray-Darling Basin, where it is recorded from the arid rivers in the north-western basin.



It is found only in the Paroo and Warrego rivers, and hybrids with Murray-Darling rainbowfish have been identified in the lowermost Warrego River and the Darling River from around the Bogan River down to at least Menindee. Any rainbowfish captured in or near the Darling River need to be carefully examined, as confusion or hybridisation with Murray-Darling rainbowfish is likely (P. J. Unmack *pers. comm.*).

*M. s. tatei* inhabit semi-permanent streams, springs, artesian bores (wells), and lagoons. During prolonged droughts the fish are confined to isolated pools. Their natural environment is subjected to seasonal variations with water temperatures ranging between 24°C during the dry season and up to 33°C in the wet season. The water is generally alkaline (pH 7.2–8.0); however, they are occasionally found in acidic conditions (pH 6.5–6.9). They are usually found around sub-surface vegetation, submerged logs, or branches. Living in a hot and arid environment *M. s. tatei* have broad physiological tolerances. Spawning typically takes place during the warmer months when temperatures are above 20°C, or whenever ample rain falls.

Balcombe *et al.*, 2005 found that terrestrial fauna was a major food group consumed by *Melanotaenia splendida tatei*. Their diet consisted of terrestrial insects (67.4%), other terrestrial invertebrates (10.2%), algae (16.8%), and aquatic insects (5.6%).

In addition to the consumption of some aquatic insects and algae, this species fed chiefly upon terrestrial arthropods, many of which were flying insects (e.g., ants, wasps and dipterans). Other items included aquatic dipterans, coleopteran larvae and zooplankton. The aquatic dipterans were mostly chironomid larvae, while the zooplankton prey consisted chiefly of conchostracans and cladocerans. Terrestrial foods included isopods, scolopendridid centipedes, or a variety of alighting insects such as dipterans, hymenopterans and coleopterans. Up to 100% terrestrial insects was consumed during the dry season.

## Remarks

This species has never generated much interest among Australian aquarists, except for a few dedicated rainbowfish enthusiasts and is still relatively uncommon in the international hobby.







# Melanotaenia sylvatica

Allen, 1997 Forest Rainbowfish

# **Species Summary**

*Melanotaenia sylvatica* was described from 26 specimens collected in 1996 during a Conservation International fauna survey in the upper Lakekamu basin of southeastern Papua New Guinea. The Lakekamu basin covers 2500 km<sup>2</sup> of pristine lowland alluvial rainforest, surrounded in the east, west and north by mountains. The Lakekamu basin is an undisturbed region with minimal human impact. The collection site is located approximately 150 km northwest of Port Moresby on the southern slope of the Central Dividing Range. Most of the basin is an extensive lowland alluvial plain over which the Lakekamu River and its tributaries meander. The collection sites were situated about 15–20 km above the Lakekamu River junction and 90–100 km upstream from the sea in the Sapoi River and its forest tributaries, close to the transition from lowland to mountainous terrain.

Melanotaenia sylvatica belongs to a species complex known as the "maccullochi group" (Allen 1981), which includes M. caerulea (Kikori River, New Guinea), M. maccullochi (northern Australia and Fly River, New Guinea), M. ogilbyi (Lorentz River, New Guinea), M. papuae (vicinity of Port Moresby, Papua New Guinea), and M. sexlineata (Fly River, New Guinea). The group is characterised by a relatively small maximum size, similar shape, and a relatively low number of dorsal, anal, and pectoral rays (7–11, 14–19, and 11–14 respectively), as well as a low number of cheek and predorsal scales (10–16 and 13–17 respectively). Although the members of the group have similar live colour patterns, each is clearly distinct (Allen 1995). Live colours are most similar to *Melanotaenia caerulea*, but it lacks the pronounced neon blue that covers much of the body. There are also differences in modal fin-ray counts between these two species. Preserved specimens, which show a distinct blackish midlateral stripe, closely resemble *M. ogilbyi*, but the two species have different live colours and there is a modal difference in the number of soft anal rays. The known geographic distributions of this pair are separated by a distance of approximately 900 km.

*Melanotaenia sylvatica* have body colour of yellowish-bronze with narrow brownish-orange stripes between each scale row. Greenish brown on upper back; midlateral row of scales on side grey to blackish, connected to similar coloured stripe extending from edge of eye; lower half of head and body silvery-white, usually with variable duskiness associated with edge of scales on side of abdomen; dorsal and anal fins yellow to translucent bronze, grey near outer margin with fine white border; caudal fin clear or slightly grey; pelvis fins pale yellow; pectorals clear or with slight yellow tint. May reach a maximum size of 6 cm, but usually less than 5 cm.



Typical of most members of the genus; males are generally deeper bodied and have a more elongate, somewhat pointed shape posteriorly on the soft dorsal and anal fins. In addition, the depressed first dorsal fin of adult males overlaps the second dorsal fin in males, but falls short of this point or barely reaches it in females. The body depth (as percentage of the standard length) of 13 males, 31.5-55.0 mm SL, ranged from 30.2-35.5 with an average of 32.7; that of 13 females, 36.6-50.6 mm S L , was 27.8-31.3 with an average of 29.5. The smallest gravid female examined was 36.6 mm SL. The smallest male exhibiting secondary sexual characteristics (elongated first dorsal fin and pointed shape posteriorly of anal and second dorsal fins) was 31.5 mm SL. Judging from the growth rates of closely related members of the "maccullochi group" sexual maturity is reached before the end of their first year.

# **Distribution & Habitat**

Tributaries of the Lakekamu and Sapoi River drainage are located approximately 150-km northwest of Port Moresby on the southern slope of the Central Dividing Range, Papua New Guinea. Typical habitat consists of small (1–3 metre wide), clear, slow-flowing creeks in closed canopy forest over relatively flat terrain, but also found in side channels and quiet pools. These creeks typically have mud or gravel bottoms and are littered with leaves and log debris. One species of submerged aquatic plant, *Hydrostemma motleyi* (Nymphaeaceae), was common in many of the streams. The fish was most abundant in 0.5–1.0 metre deep pools behind fallen logs or buttress roots of large trees. In addition to the primary forest habitat, *M. sylvatica* also occurs in the main Sapoi River below an altitude of about 50 metres. Above this altitude, the river undergoes a relatively quick transition from a slow-flowing lowland stream to a mountain torrent. The riverine habitat of *M. sylvatica* consists of deeper (to 3 metres), sand or gravel bottom pools, often behind log jams, either in shaded positions or in full sunlight. Two other species of rainbowfish, *M. goldiei* (abundant) and *M. rubrostriatus* (rare) share this habitat. Temperatures recorded in the habitats ranged from 20° to 29°C. Aquatic and terrestrial insects and various aquatic larval insects feature prominently in the diet of the rainbowfishes.

The fish fauna of the upper Lakekamu Basin is broadly typical of freshwater localities in New Guinea. It consists of approximately 23 species in 18 genera and 14 families and is dominated by catfishes, rainbowfishes, gobies and gudgeons. The majority of species recorded are distributed widely either across the southern portion of New Guinea or the combined northern Australia-southern New Guinea region. The rivers of the upper Basin are still pristine and essentially uncontaminated by introduced species.

# Remarks

The species is named "*sylvatica*" (Latin: "of the forest") with reference to its typical forest habitat. They are currently not available in the aquarium hobby.







# Melanotaenia synergos

Allen & Unmack, 2008 Batanta Island Rainbowfish

### **Species Summary**

Adult males have a bluish (turquoise) wash above a usually discontinuous mid-lateral stripe and a whitish belly region. They have a gold (yellowish) wash of colour above and below the lateral line near the caudal peduncle. The second dorsal and anal fins are a silver-grey-blue colour. Females are similarly coloured but not as intense and their dorsal/anal fins are uncoloured. They are very similar in colouration, body shape etc., to Melanotaenia catherinae and can easily be confused with this species. The two species share similar meristic and morphological features as well as general colour pattern similarities. However, they differ in modal counts for pectoralfin rays and lateral scales. They also exhibit slight colour pattern differences related to the width of the dark midlateral stripe, which is generally narrower in Melanotaenia synergos, covering one and a half scale rows for most of its length versus 2 to 3 scale rows for Melanotaenia catherinae. This species should not be confused with Melanotaenia batanta, another rainbowfish found on Batanta Island.

Males are distinguished from females by their brighter colours and longer and more elongated fin rays. Growing to a length of around 10 to 12 cm, and a body depth of 3.5 to 4.0 cm, males are usually much larger and deeper bodied than females. I found this species to be reasonably tolerant of the presence of newly hatched fry in their aquarium.

### **Distribution & Habitat**

*Melanotaenia synergos* is currently only known from Batanta Island, which lies immediately west of the West Papuan mainland. Batanta is a small island approximately 55 kilometres long and 30–35 kilometres wide and is home to three separate species of rainbowfishes. Much of the Island is covered with dense rainforest. They are mainly found around submerged logs, or branches in clear rainforest streams, in water temperatures between 18–28° Celsius. However, their natural environment is subjected to seasonal variations with water temperature, pH, and hardness levels varying considerably.

### Remarks

This species was first collected in 1992 by Heiko Bleher while exploring the freshwaters of Batanta Island. Gerry Allen tentatively identified the species as *Melanotaenia misoolensis*, which he described in 1982 from a collection held in the Zoological Museum of the University of Amsterdam in the Netherlands. However, as it turns out they were an undescribed species.









**Melanotaenia trifasciata** (Rendahl, 1922) Regal Rainbowfish

*Rhombosoma trifasciata* Rendahl, 1922 *Melanotaenia trifasciata* Allen, 1980

### Species Summary

*Melanotaenia trifasciata* was collected in June 1894 (Port Darwin) by Knut Dahl, a Norwegian Zoologist. The species description was published in 1922 by Hialmar Rendahl, on the basis of one single specimen from the Mary River, in the Northern Territory. Why he proposed the name trifasciata, meaning literally "three-banded", is not very obvious when you look at live specimens. However, it becomes clear when you bear in mind that Rendahl only knew this one museum specimen caught more than 20 years earlier. He described the colours as follows:

"The ground-colour of the fish is (in spirits) a light brown. Along the sides of the body, occupying the adjacent two-thirds of the scales, there is a broad blackish brown bar on the 5th and 6th longitudinal rows of the scales. ... On both sides, this dark bar is (except on the head) bordered by a very obvious light (in alcohol whitish), bar, the ventral of which is the broadest and about half the width of the dark one." *M. trifasciata* are commonly known as the Banded Rainbowfish. However, I think they should be called the "*Regal Rainbowfish*" because they are without doubt the most majestic rainbowfishes you will ever see. As with many rainbowfishes, the colouration of *M. trifasciata* is variable depending on location, water conditions and diet. Populations from almost every river system where they are found have their own distinctive body colour. Consequently, specific names usually based on the locality where each is found are used by rainbowfish enthusiasts to identify each variety. They can be recognised by a very deep body, usually deeper than 1/3 of their body length, and an often discontinuous black mid-lateral band. Males are more brightly coloured, larger, and much deeper bodied than females. Specimens found in a number of rivers in Queensland are more streamlined and do not have the deeper body shape of their counterparts from other river systems.

*M. trifasciata* may reach a maximum size of 15 cm, but are usually less than 12 cm, with a body depth of 6–8 cm. Males are deeper bodied than females and the overall colour pattern of males is more intense. The vertical fins of females are either translucent or only faintly coloured compared to the brighter colours of males. These features become more obvious with increased growth.





# **Distribution & Habitat**

*Melanotaenia trifasciata* have a discontinuous distribution across northern Australia, from the Mary River in the Northern Territory, throughout Arnhem Land to Cape York Peninsula. In north Queensland they are found as far south as Gap Creek, north of the Bloomfield River. In 1989, some specimens were collected on Melville Island, the only offshore record thus far. Recent genetic studies suggest that *M. trifasciata* may also inhabit the Fly River and the Aru Islands (P. J. Unmack 2009, *pers. comm.*).

Melanotaenia trifasciata are a tropical fish, and occur in almost every kind of freshwater habitat, from slow-moving streams, wetland swamps, lagoons and clear flowing rivers. They are most common in flowing waters or in streams where water flow is present for much of the year. M. trifasciata are most frequently found in clear waters with sandy substrates, followed by rocks, leaves and mud. In these habitats they are commonly found around sub-surface vegetation, submerged logs, or branches. Depth of waters in which M. trifasciata are mostly found range from 30 to 200 cm. They are often found inhabiting the same streams with M. nigrans, M. maccullochi and M. s. inornata. Their natural environment is subjected to seasonal variations with water temperatures ranging between 24°C during the dry season and up to 33°C in the wet season. The water is generally alkaline pH 7.2–8.0; however, they are occasionally found in acidic conditions pH 6.5-6.9.

Rainbowfishes have evolved to survive under a wide range of environmental conditions, and those conditions and the habitats created can change dramatically over time. Part of the reason for this is that Australian freshwater fish have adapted to live in variable and unpredictable environments, and so have not developed the strong habitat associations that are characteristic of highly specialised fish in more predictable river systems in other countries. Not surprisingly, the largest and healthiest populations of a species will generally be found where the conditions are closest to optimal for all the water quality factors.

## Biology

*M. trifasciata* characteristically display a considerable range of growth rates, depending on conditions such as food, space, numbers, competition and water temperature. In tropical waters, which have prevailing high temperatures, fish generally grow faster, mature younger, and have a shorter life span than fish in temperate waters.

*M. trifasciata* are aseasonal spawners, breeding continuously at intervals throughout the year. Therefore it is difficult to define their breeding season. However, a peak in reproductive activity is usually during the early-wet season. The breeding season must coincide with the conditions that offer the greatest amount of protection for the eggs, and food and shelter for the newly hatched young. The duration and timing of reproductive activity are thus two critical components for their continual survival.

Females produce between 200 and 500 eggs, spawning a number of times daily for several days or opportunistically whenever conditions are favourable. Large females usually produce more than 50 eggs per day for several days. Smaller females, which are only just sexually mature, shed fewer eggs, 20–30 per day and spawning does not occur daily. The eggs are attached by adhesive threads or tendrils to aquatic plants or artificial substitutes. The eggs are large (average  $1.5 \pm 0.5$  mm in diameter); clear to light amber in colour. After spawning, the female will leave, while the male remains displaying to passing females and thus defending his territory and the fertilised eggs. Depending on temperature hatching will occur 6–7 days after spawning.

*M. trifasciata* is an omnivore feeding opportunistically in the surface and mid-water regions. In sunny conditions shoals of juveniles occurred near the water surface, but larger fish tend to occur in the mid-water region near submerged vegetation, often utilising aquatic plants as a refuge and food source. Under cloudy conditions, however, fish of all sizes preferred deeper water.





The main food items are aquatic insects, algae and terrestrial insects such as green ants (*Oecophylla smaragdina*), which presumably fall on to the water surface from overhanging vegetation.

The diet varies in relation to the habitat they occupy. In the mainchannel waterbodies they eat mainly aquatic insects, with small amounts of terrestrial insects, plant material and algae. In perennial streams, algae and terrestrial plant material are less important, while aquatic insects and, to a lesser extent, oligochaetes and micro-crustaceans, are consumed. The diet in the lowland sandy creekbeds has much larger algal and terrestrial insect components.

Specimens examined from the floodplains feed mainly on aquatic arachnids and aquatic insects, and a small amount of algae. Planktonic invertebrates (mostly zooplankton) are importance in their early life history stages. The availability of appropriate zooplankton is an important determinant of mortality levels endured by larval fish populations and thus is an important determinant of recruitment into the adult population.

### Remarks

Due to the diversity of range and habitat, there are many colour variations in the *trifasciata* group. One of the most appealing is the 'Goyder River' variety found in the Goyder River in Arnhem Land. This species was first introduced to the aquarium hobby back in the early 1970's.

Wally Muller, a well-known aquarium dealer in those days obtained a few during his visit to the Northern Territory to collect aquarium specimens. He obtained permits from the Fisheries and Wildlife Department, and was able to bring back some fine specimens to Brisbane where he had his retail business.

When first introduced to the aquarium hobby in Brisbane, the Goyder River rainbowfish created quite a deal of excitement. At a public aquarium exhibition, where they made their first public appearance, the cover glass was glued to the aquarium proper so that no one would be able to steal them.





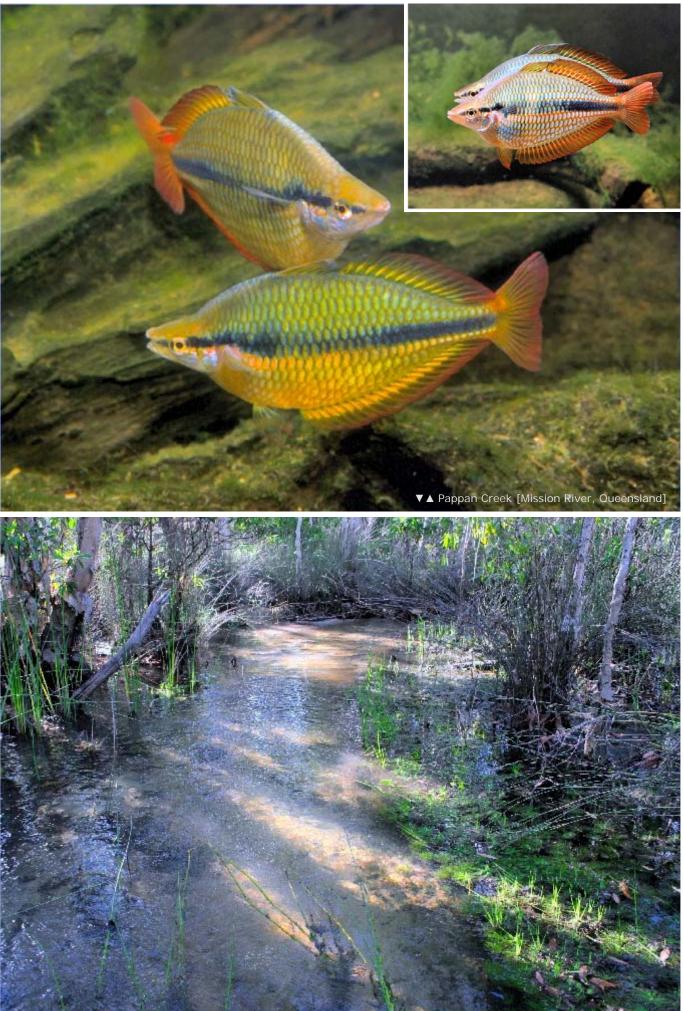












Leo O'Reilly



















# Melanotaenia utcheensis

McGuigan, 2001 Utchee Creek Rainbowfish

#### **Species Summary**

Melanotaenia utcheensis has a distinctive colour pattern with a blue-black mid-lateral band and orange margins on the vertical scale rows. It is morphologically distinct from the broadly sympatric *M. eachamensis* and *M. s. splendida*, as well as from its sister species from southern Queensland/northern New South Wales, *M. duboulayi*. In particular, *M. utcheensis* has more first dorsal spines and fewer vertical scale rows and anal rays than *M. s. splendida*, and fewer soft second dorsal rays and more pectoral rays than either *M. eachamensis* or *M. duboulayi*. *M. utcheensis* is also generally smaller than either *M. s. splendida* or *M. eachamensis* and intermediate between them in eye diameter, predorsal length, head depth and body depth.

#### **Distribution & Habitat**

*Melanotaenia utcheensis* have currently only been collected from the Utchee, Fisher, Rankin and Short Creeks in the North and South Johnstone River catchments in north Queensland. They are found in sites with moderate to high water flow over cobbles and boulders. The Johnstone River flows into the Coral Sea near the north Queensland town of Innisfail. The river branches about 5 km from the mouth into the North and South Johnstone Rivers, both of which have their sources on the Atherton Tablelands. Rankin and Fisher Creeks flow northeast into the lower North Johnstone River. Short Creek and an unnamed creek are in the upper North Johnstone a little bit upstream of Gillies and Dirran Creek and they enter from the opposite side of the river. Utchee Creek feeds into the South Johnstone River on the coastal plain upstream of Innisfail.

#### Remarks

I obtained wild-caught specimens of *Melanotaenia utcheensis* in September 1983, and maintained a small captive population until at least 1989. Although, at the time they were considered just a variety of *M. s. splendida*. Genetic studies beginning in the mid 1990's revealed the existence of significant genetic variation between populations of *Melanotaenia splendida* that occur in the upland streams of north Queensland. In particular, these studies highlighted the degree of isolation of upland populations from the lowland populations. Subsequent genetic research suggested that at least some of these species are unusual variants of *Melanotaenia splendida* - or populations displaying genes that have traits of more than one species. As a direct result of some of this research, *Melanotaenia utcheensis* was described as a new species in 2000.







# Melanotaenia vanheurni

(Weber and de Beaufort, 1922) Van Heurn's Rainbowfish

*Rhombatractus vanheurni* Weber & de Beaufort, 1922 *Melanotaenia vanheurni* Allen, 1980

#### **Species Summary**

*Melanotaenia vanheurni* are brown or olive on the back and yellow on the lower sides. There is a prominent blue-black, midlateral band with a broad pale yellow to white line along its upper and lower margin. Spawning males display a pulsing glow of golden yellow on top of the head. They may reach a maximum size of 20 cm, but usually less than 15 cm. It is the largest member of the rainbowfish family.

#### Remarks

*Melanotaenia vanheurni* were first collected in the Mamberamo Valley by W. C. van Heurn in 1920 during the Dutch Northern New Guinea Expedition (Mamberamo Expedition) of 1920–1921. It was not seen again until David Price collected it 70 years later.

Live specimens were reportedly imported into Germany during 1992 by Heiko Bleher. However, the status of this population in the hobby today is uncertain. Further live specimens were collected in 2008 near Faowi Village, located at the upper reaches of the Tariku River. The Tariku River flows from Sudirman Mountains in the west to the east and combines with Taritatu River in the middle of Mamberamo River Catchment.

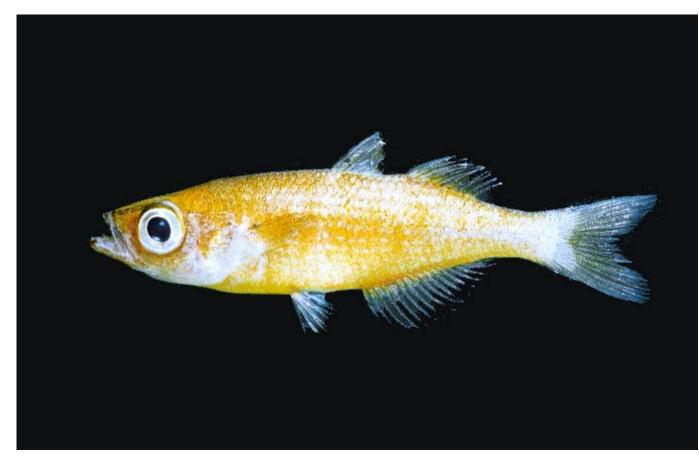
#### **Distribution & Habitat**

Mamberamo River system of northern West Papua. They have been collected from small clear water streams flowing through rainforest mainly close to foothills around the periphery of the Mamberamo Plains.

They are abundant in slow to relatively swift flowing streams with rock and sandy bottoms. It shares this habitat with *Chilatherina fasciata*. Temperature and *p*H values range between 25– $28^{\circ}$  Celsius and 7.1–7.5.







### Pelangia mbutaensis Allen, 1998

Lake Mbuta Rainbowfish

#### Species Summary

The Pelangia genus is the sister group of Glossolepis, judging from external appearance and osteological features, particularly with regards to dentition, and morphology of the premaxillary, pelvic girdle, and pectoral fin (Allen 1980). However, it differs from Glossolepis with regards to several important features including a lack of teeth on the vomer and palatines, a larger mouth (jaws extend to below front of eye, but fails to reach eye in Glossolepis), a larger eye (horizontal diameter exceeds caudal peduncle depth; equal to or less than depth in Glossolepis), reduced crenulations on the scale margins, and fewer anal rays (14-18, usually 16 compared with 18-22 in Glossolepis). There are also differences in the positions of the fins. The anal fin origin is closer to the caudal fin base than to the snout tip, which is the opposite situation compared to Glossolepis. Moreover, the origin of the first dorsal fin is well ahead of the anal fin origin, but in Glossolepis it is either even with the anal fin origin or behind it. Finally, the second dorsal fin origin is approximately level with the third soft anal ray in Pelangia, but in Glossolepis it is usually level with the middle rays.

*Pelangia mbutaensis* was described on the basis of 52 specimens collected in 1997 in the Mbuta Basin near Etna Bay, West Papua. They have a yellowish-tan body colour grading to silvery-white on the lower half of sides. Larger males have scattered yellow flecks along the middle of the side. Fins are mainly translucent except for dusky grey pigmentation on the first dorsal fin. They may reach a maximum size of 6 cm, but usually less than 5 cm.

The difference between sexes is far less evident in this species than in most rainbowfishes. Males have a slightly longer first dorsal fin, which slightly overlaps the second dorsal fin origin when depressed. By contrast, the depressed first dorsal fin of females fails to reach the dorsal fin origin or barely reaches it. In addition, males generally have a deeper body than females. The average body depth as percentage of the SL for eight mature males, 45.2-55.5 mm SL (average = 49.3 mm SL) was 31.3 compared with an average of 28.0 for six females, 43.0-51.4 mm SL (average = 47.9 mm SL).

#### **Distribution & Habitat**

Lake Mbuta Basin, lying approximately 8 km inland from Etna Bay, West Papua is invariably represented on published maps as a lake, but is actually a swampy basin surrounded by low mountains. Although a lake was probably present in former times there is no indication of recent inundation. The basin, which is roughly circular and measures up to 7-8 km in width, is mainly covered by swamp overgrown with 3-4 m tall grass with numerous small ponds, creeks, and at least one small river. It is not known if there is surface drainage from the basin to Etna Bay (about 8 km away) or whether the drainage is subterranean, as is the case for several other small lakes in the area. P. mbutaensis were collected in a small creek approximately 2-3 metres in width, with depths to about 2 metres. The collection was made over a 50 metre long section immediately above its confluence with a small turbid river. The water was very clear, but darkly stained (tea-coloured), with relatively fast flow through forest that formed a nearly closed canopy. The bottom consisted mainly of mud with occasional rocks and log debris with sparse aquatic vegetation. A water temperature of 25.8°C and pH of 6.4 were recorded. Other rainbowfish inhabitants included Melanotaenia goldiei. P. mbutaensis was relatively common, but outnumbered by M. goldiei by a 3:1 ratio.



# Hainbowfishes

There is a number of other informally recognised species of rainbowfishes being maintained in the aquarium hobby that are still awaiting formal scientific description. Clearly, there will also be many more new species found in New Guinea, as most areas remain poorly collected. There is also a need for more careful study of the many widespread species, as it is highly likely that such study will lead to a significant increase in the number of recognised species. For example, variation in morphology within the *Melanotaenia* genus is high, with species differing from one another though small variations in colour, morphology and meristics, each with highly restricted, allopatric distributions. Much could be gained from careful analysis of the many morphological characters already at hand, such as the colouration characters noted for many of the rainbowfish "varieties". Colouration characters, however, when not supported by other characters, have generally been dismissed by ichthyologists working on rainbowfishes from Australia.

The study of species questions and hybridisation has been greatly facilitated by the development of genetic studies, leading to the identification of presumptive new species as natural hybrids or captive hybrids from the aquarium hobby.

Rainbowfishes that may undergo species separation after further genetic studies include:

Chilatherina fasciata; Iriatherina werneri; Melanotaenia australis; Melanotaenia exquisita; Melanotaenia goldiei; Melanotaenia maccullochi; Melanotaenia splendida; Melanotaenia trifasciata; Melanotaenia utcheensis (South Johnstone River); Pseudomugil signifer; Pseudomugil tenellus and Pseudomugil paludicola.

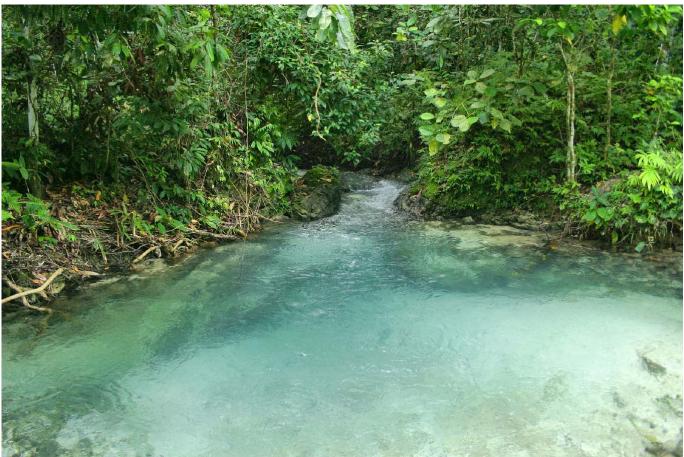
(P. J. Unmack 2009, pers. comm.)



# Melanotaenia sp. (Bonggo Village)

#### **Species Summary**

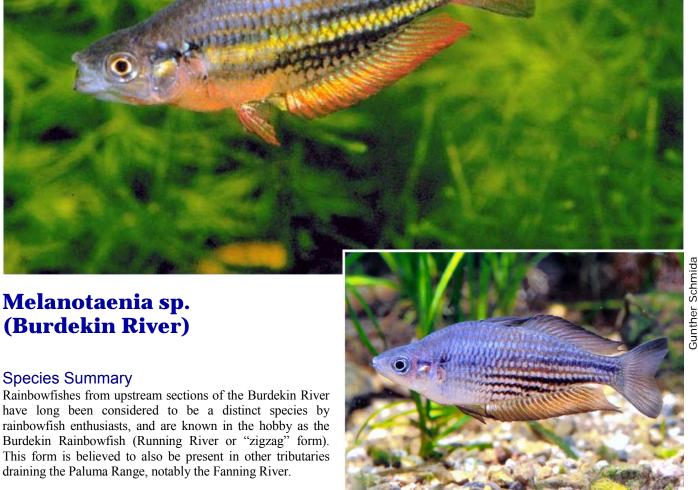
This species was collected by Gary Lange in 2006 from a small stream near the village of "Bonggo" located near Jayapura in West Papua. The small stream flowed under the gravel road and the fish were collected where the water was shallow and a lot easier to catch. Preliminary genetic studies suggest that it is an undescribed species (P. J. Unmack 2009, *pers. comm.*). This species is currently available in the hobby.



**Melanotaenia sp. (Katherine River)** This species was collected in 2009 from the headwaters of the Katherine River in the Northern Territory, located on the top of the Arnhem Land Escarpment.







#### **Distribution & Habitat**

The Burdekin River, the fifth largest in Australia (Australia's largest in terms of peak discharge), is located in north-eastern Queensland. The catchment is the second largest on the east coast of Queensland (after the Fitzroy), covering a total area of 130 500 km<sup>2</sup>. The catchment comprises four distinct subcatchments.

Running River is located near Hidden Valley, 40 km west of Paluma and covers an area of approximately 300 km<sup>2</sup>. The river passes through open eucalypt forest, and forms a steep gorge south of Hidden Valley. This runs for approximate 10 km. The river starts at an elevation of 660 metres and ends at



an elevation of 313 metres merging with the Burdekin River. Deception Creek is a major eastern tributary of Running River and drains a particularly rugged section of the Coane Range.

The Fanning River has two main arms arising within the Herveys Range, west of Townsville, then falling through a gorge before flattening out in savannah rangelands. The west arm within and above the gorge contains a number of permanent waterholes, though the presence of water in the lower half of the river is limited. The upper portions of the river are within a military training area, thus limiting their land use and water conditions there is generally considered to be very good.







# Melanotaenia sp. (Suswa Village)

#### **Species Summary**

*Melanotaenia* sp. "Suswa Village" were originally collected from the Auk River at Suswa Village (approximately 0°56'S, 132°15'E) by G.R. Allen and H. Bleher in 1982 and formed part of the original description of *Melanotaenia irianjaya* in 1985. However, genetic studies clearly show that the specimens collected from Suswa Village were a different species (P. J. Unmack 2009, *pers. comm.*)

They look very similar to *Melanotaenia irianjaya* and were originally distributed in the hobby as *Melanotaenia irianjaya* "Suswa Village". They have a distinctive colour pattern with two dark blotches on the side of the body. These blotches become visible at around 3 cm in size, and increase with age and size. The mauve base colour and the black and white edging of the second dorsal fin are concise characteristics that don't occur on any other rainbowfish species in this combination (Graf 2009).

This species was being maintained locally and were eventually taken to the United States where they were bred and distributed into the aquarium hobby.







The Aru Islands (also known as Aroe Islands or Kepulauan Aru) lie on the western edge of the shallow seas of Torres Strait, around 7°S and 134°E. New Guinea is some 150 km to the north and Arnhem Land in Australia is some 550 km to the south. There are six low-lying islands of significant size -Kola, Wokam, Kobroor, Maikoor, Koba and Trangan - and many smaller ones, comprising about 180 islands in total. The islands lie very close to one another. The seven largest are separated only by narrow channels and are effectively one land mass. The archipelago stretches about 180 km north to south, and is 80 km east to west at its widest, with a total area of about 8563 km<sup>2</sup>. It has a low dissected terrain including chains of low hills with the highest point only some 240 metres above sea level, and extensive areas of coastal and inland swamp. The sea around the islands is shallow, generally less than 20 metres in depth. The islands lie on the Australia-New Guinea continental shelf, and were connected to Australia and New Guinea by land when sea levels were lower during the ice ages.

Seasonal streams are common and some flowing water occurs from springs. Lakes are rare except for a few small karst hollows on Kobroor and Trangan Islands. Permanent and seasonal swamps are also found on Trangan Island. Springs occur along the coast and interior gorges. The savannah formation has strong relationships with southern New Guinea and northern Australia, especially Cape York Peninsula. Three rainbowfishes have been reported from the Aru Islands:

#### Melanotaenia goldiei

Melanotaenia trifasciata (Rhombatractus senckenbergianus) Melanotaenia splendida rubrostriata

#### Remarks

Heiko Bleher collected a number of different rainbowfishes from the Aru Islands. He named them as follows:

Melanotaenia sp.1 – Aru I (Loramar River, Korobor) Melanotaenia sp.2 – Aru II (Ngadamdi, Korobor) Melanotaenia sp.3 – Aru IV (Sin River, Trangan – This is the type locality of *Rhombatractus senckenbergianus*.)

Preliminary genetic study (P. J. Unmack 2009, *pers. comm.*) suggests that the rainbowfishes collected by Heiko Bleher as *Melanotaenia* sp.3 (Aru IV) are actually *Melanotaenia trifasciata*. It is possible however; that these fish are the same that Weber described as *Rhombatractus senckenbergianus*. If that is the case then all *M. trifasciata* would become *M. senckenbergianus* because *Rhombatractus senckenbergianus* Weber, 1910(11) predates *Rhombosoma trifasciata* Rendahl, 1922. However the type specimens would have to be re-examined before any changes were considered.





In reviewing the description of *Rhombatractus goldiei* in 1922, Weber & de Beaufort made the following statement: "Specimens from the Aru-Islands (*Rhombatractus senckenbergianus*) seem to be a little more elongate. Height 3.5-3.75, more than 4-4.5 in length with caudal. As the largest specimen known from the Aru-Islands is 92 mm. and considering the well known variability in the height of these fishes, we don't think the difference sufficient to keep them apart."



▲ ▼ Melanotaenia sp. Aru 3 [Sin River, Trangan Island]









#### **Pseudomugil connieae** (Allen, 1981) Popondetta Blue Eye

*Popondetta connieae* Allen, 1981 *Popondichthys connieae* Allen, 1987 *Pseudomugil connieae* Saeed, Ivantsoff & Allen, 1989

#### **Species Summary**

When *Pseudomugil connieae* were initially discovered they were mistakenly identified as *Pseudomugil furcatus*, a species described by Nichols in 1955. However, when Gerald Allen realised that they were a new species he called them *Popondetta connieae* after his wife Connie (Lagos) Allen. When it was discovered that the genus *Popondetta* already existed, he renamed them *Popondichthys connieae*. However, in 1989 they were placed in the genus *Pseudomugil*, where they remain today.

Males are easily distinguished from females by their brighter colours and longer and more elongated dorsal fin. The body colour is yellow-green in both males and females. The dorsal and anal fins of the males have a broad white or yellow outer margin and black band across the middle. The outer region of the first dorsal fin is yellow. Females are similarly coloured but not as intense, and have much smaller fins, which lack the detailed markings of the males. They have a moderately compressed and elongated body and grow to a length of around 4 to 5 cm.

#### **Distribution & Habitat**

*Pseudomugil connieae* were initially collected from a number of small creeks in the vicinity of Popondetta, situated on the northern side of the central dividing range, eastern Papua New Guinea. They are common in the vicinity of Popondetta and have been collected from a number of localities within a 25 km radius. They are generally found in small, clear, relatively swift-flowing freshwater streams. Temperature and *p*H in these streams recorded at the time of collection ranged from 24–27°C and 7.7–7.9.

#### Remarks

This species was originally collected by Gerald Allen and Brian Parkinson in 1978. They collected approximately 200 specimens. However, mortalities were high and only eight specimens survived the journey back to Australia. These were shipped to Sydney and picked up by Gunther Schmida and acclimatised in his tanks for several weeks before the last lag of their journey to Perth. Small numbers were eventually bred and circulated in the Australian hobby.



*Pseudomugil connieae* have never been readily available in the aquarium hobby and this small initial group formed the base of all populations in Australia, which I might add has now almost disappeared. Heiko Bleher collected live specimens in 1982 and these were bred and distributed in Europe and form much of the available stock currently in Europe. I first obtained stock in 1992 and bred them in January 1993. Some of these were sent to Europe in 1994, which at the time there wasn't many available in Europe.

*Pseudomugil connieae* are a magnificent blue eye and are much sought after by aquarium hobbyists. It is very important to provide regular partial water changes as I have found this species doesn't like old, acidic water. The key for successfully maintaining *Pseudomugil connieae* in captivity is excellent water conditions. This can be easily provided with regular partial water changes. They will display their best colouration when maintained in a densely-planted, partly shaded aquarium. However, this is not a requirement for their successful maintenance in captivity. I have bred and raised them in captivity with the following water conditions: Temperature 19–31°C, *p*H 7.6–8.2, Hardness 90–150 mg/L, Alkalinity 40–65 mg/L and Conductivity 369–663  $\mu$ S/cm. Eggs adhere to water plants and hatching occurs around 15 days at a temperature of 25° Celsius.

The stomach content of several wild-caught specimens indicated a diet consisting primarily of minute crustaceans and insect larvae with small amount of algal matter.

# Comparisons of *Pseudomugil connieae* and *Pseudomugil furcatus* :

The two species are easily distinguished on the basis of colour and there are significant differences in modal counts for the second dorsal and anal fins. The most apparent differences involve fin colouration of mature males. The dorsal and anal fins of P. furcatus are mainly transparent to slightly dusky with a relatively narrow outer margin of yellow. Those of P. connieae have broad, whitish outer margins with a bold black band across the middle of each fin; the outer portion of the first dorsal fin is yellow as in P. furcatus. However, the caudal fin of P. furcatus has a pale yellow lobes with thin black dorsal and ventral margins, whereas that of P. connieae has whitish lobes, a dusty central portion, and lacks dark margins. The pelvic fins of P. connieae are largely dusky or blackish and the pectoral fins are whitish on the upper edge. By contrast the pelvics of male *P. furcatus* are yellow and this same colour is present on the upper edge of the pectoral fins. The females of P. connieae are basically similar to males except the dark bands in the middle of the dorsal and anal fins are less distinct and narrower, the outer edge of the second dorsal is broadly yellow (as in *P. furcatus*), the caudal fin lobes are yellowish and the pelvic and pectorals are uniformly transparent.







## **Pseudomugil cyanodorsalis**

Allen & Sarti, 1983 Neon Blue Eye

#### **Species Summary**

Pseudomugil cyanodorsalis is a very colourful species and deserves a much better common name than 'Blueback Blue Eye' as suggested by Gerald Allen. The upper half of the males' body is metallic blue (similar to the Neon Tetra) and peppered with fine grainy melanophores. The lower half of the males' body is translucent to yellowish white. A single thin dark mid-lateral line runs from the base of the pectoral fin to the caudal fin. The first dorsal fin is translucent with an outer blackish border, and a small vellowish patch at the base near the last spine. The elongated anterior rays and outer edge of the second dorsal and anal fins are black. The remainder of the fin is creamy yellow or whitish. The caudal and pectoral fins are translucent or slightly yellowish with black outer edges. The body of the female is a semi-transparent silver-grey colour with translucent fins and white abdomen. Maximum size is around 3.5 cm. Pseudomugil cyanodorsalis were originally available in the Australian hobby in 1982 but failed to become established. Another wild-collection for the aquarium hobby was made in 1986 and today, they are widely distributed in the aquarium hobby all around the world.

#### **Distribution & Habitat**

*Pseudomugil cyanodorsalis* was first collected by Helen Larson (Northern Territory Museum) in 1981 near Darwin. A year later Gerald Allen (Western Australian Museum) found them in Crab Creek, 15 km east of Broome in Western Australia. In 1983, there were scientifically described by Allen & Sarti. They have been collected from around Broom and Wyndham in northern Western Australia. In the Northern Territory they have been recorded in coastal catchments around Darwin and the Mary River. They have been collected from Melville Island. In Queensland they have been collected from the Norman River in the Gulf of Carpentaria. They are probably widely distributed in estuarine and coastal freshwater habitats across northern Australia and southern New Guinea. *Pseudomugil cyanodorsalis* are sympatric with *Pseudomugil inconspicuus*, and are found together in at least one locality (Woods Inlet) near Darwin.

*Pseudomugil cyanodorsalis* are euryhaline and tolerate a wide range of ecological conditions. Although more commonly found in small brackish estuarine creeks, they also inhabit pure freshwater habitats, especially during the wet season. During the wet season, freshwater flowing into these habitats dilutes the waters to fresh. Water thus varies from saline through brackish to fresh. However, habitat preference appears to be mangrove-lined muddy brackish creeks, where they are commonly found in large numbers. They have been found in hypersaline waters (28–40 ppt) and at temperatures of 22–39°C.







Dave Wilson



## **Pseudomugil furcatus**

Nichols, 1955 Forktail Blue-eye

#### **Species Summary**

*Pseudomugil furcatus* is a small species growing to a length of about 5 or 6 cm. They have two dorsal fins, separated by a small gap, the first much smaller than the second. The body colour is yellow-green in both males and females. The dorsal and anal fins of the males are transparent with narrow yellow margins. The pelvic and pectoral fins are often tinged with red. The caudal fin lobes are yellowish with black dorsal and ventral margins. Females caudal fin lobes are yellowish while the pectoral and pelvic fins are transparent. The outer part of the second dorsal fin is yellowish. The body scales have a slight dark edge. They differ from most other forms of Pseudomugil from Australia and New Guinea, in having the caudal fin longer and more deeply forked. Males are easily distinguished from females by their brighter colours and longer and more elongated dorsal fin.

This species was originally named *Pseudomugil furcatus* in 1955 by John Treadwell Nichols, curator of recent fishes at the American Museum of Natural History. They were collected by Hobart M. Van Deusen during the Forth Archbold Expedition to New Guinea on August 24, 1953. In a review of the family Melanotaeniidae in 1980, they were separated from the

*Pseudomugil* genus and placed in a new genus *Popondetta*, and the name was changed to *Popondetta furcata*. It was then later discovered that the genus name '*Popondetta*' was previously used and in 1987 they underwent another name change and were then called *Popondichthys furcatus*. Two years were to pass and following a review of the *Pseudomugil* genus in 1989, the blue-eye group, including *Popondichthys furcatus*, were placed in their own family Pseudomugilidae and they were returned to their original name of *Pseudomugil furcatus*.

#### **Distribution & Habitat**

*Pseudomugil furcatus* were originally collected from Peria Creek, a tributary of the Kwagira (Kwagila) River, in Papua New Guinea. They have also been collected from Safia, in the Musa River valley where they are relatively common in small, clear rainforest streams. They have a range in Papua New Guinea between Dyke Ackland and Collingwood Bays. They are generally found in small, clear, relatively swift-flowing freshwater streams with abundant aquatic vegetation. Water conditions reported from their natural habitats are: Temperature 24–28.5°C; pH 7.0–8.0 and Hardness 90–180 ppm.

#### Remarks

Live specimens were collected in 1981 by Gerald Allen and Barry Crockford and were returned to Australia whereupon they were later bred and established in the aquarium hobby.



# Comparisons of *Pseudomugil connieae* and *Pseudomugil furcatus* :

The two species are easily distinguished on the basis of colour and there are significant differences in modal counts for the second dorsal and anal fins. The most apparent differences involve fin colouration of mature males. The dorsal and anal fins of *P. furcatus* are mainly transparent to slightly dusky with a relatively narrow outer margin of yellow. Those of P. connieae have broad, whitish outer margins with a bold black band across the middle of each fin; the outer portion of the first dorsal fin is yellow as in P. furcatus. However, the caudal fin of P. furcatus has a pale yellow lobes with thin black dorsal and ventral margins, whereas that of P. connieae has whitish lobes, a dusty central portion, and lacks dark margins. The pelvic fins of *P. connieae* are largely dusky or blackish and the pectoral fins are whitish on the upper edge. By contrast the pelvics of male *P. furcatus* are yellow and this same colour is present on the upper edge of the pectoral fins. The females of P. connieae are basically similar to males except the dark bands in the middle of the dorsal and anal fins are less distinct and narrower, the outer edge of the second dorsal is broadly yellow (as in *P. furcatus*), the caudal fin lobes are yellowish and the pelvic and pectorals are uniformly transparent.













# Pseudomugil gertrudae

Weber, 1911 Spotted Blue Eye

#### **Species Summary**

Pseudomugil gertrudae is a small freshwater fish growing to a size of around 30 mm and is endemic to Australia and New Guinea. They have a moderately compressed and elongated body that is a semi-transparent silvery-blue colour, sometimes having an overall wash of golden-yellow. They have two dorsal fins, very close together, the first much smaller than the second. The tips of the pectoral fins can be bright yellow, orange or orange-red, other fins often edged with white. The dorsal, anal and tail fins can be clear to white, silvery-grey or yellow with rounded or oblong dark spots scattered all over. Several rows of body scales are edged in black forming an attractive latticework pattern over the body. Females generally have a deeper body than the males whilst the adult males have larger dorsal, anal and pelvic fins, with extended filaments on the first dorsal and pelvic fins. Males also exhibit more intense spotting on the body and fins. This species has a patchy distribution where it occurs and as such, there is considerably variation between the different populations in colouration and body size, as well as fin size and shape.

The variety from Weipa is one of the most impressive forms and can be found in Melaleuca swamps besides the road leading into town. A similar form can be found in Pappan Creek. Pappan Creek flows into the Mission River. The forms typical of those found at the top of Cape York in areas such as the Jardine River (and its associated swamps) and Burster Creek have large round fins and multiple small spots. In Arnhem Land a nice form with golden body colours occurs in Goanna Lagoon while not far away in the Giddy River the fish have larger fins with larger spots but no golden body colour. Specimens collected in the Darwin region typically have orange pectoral fins. There are many other forms of this beautiful little fish and new ones are regularly being discovered. Pseudomugil gertrudae from the Aru Islands are larger and have an overall a golden colour (fins and body). To date there has been no research published on the genetic or physical characteristics of the various populations.

*Pseudomugil gertrudae* is very similar to the endemic New Guinea species, *Pseudomugil paskai*. Both species are characterised by rows of permanent spots covering the fins. The only differences being the colour and shape of the fins. *Pseudomugil gertrudae* were originally collected from Terangan Island (one of the Aru Islands), which lies directly south of the Vogelkop Peninsula in western New Guinea. They were described by Max Wilhelm Carl Weber, Professor of Zoology at the University of Amsterdam in 1911 and named "*gertrudae*" after the wife of Dr. Hugo Merton, a German naturalist who travelled through the Aru Islands between October 1907 and August 1908.





#### **Distribution & Habitat**

In Australia, *Pseudomugil gertrudae* has been found in scattered localities in river systems which flow into the Timor Sea and Gulf of Carpentaria - from Darwin through Kakadu and Arnhem Land to Cape York Peninsula, including the offshore islands of Bathurst, Melville and Groote Eylandt, plus some of the islands in the Torres Strait. They are widespread throughout Cape York Peninsula, extending down the eastern coastal plains to around the Innisfail - Tully region.

Their known distribution within Australia includes the Adelaide, Alligator, Blyth, Buckingham, Cadell, Cato, Daly, Finniss, Liverpool, and the Moyle River systems in the Northern Territory; including the smaller catchments around Darwin. In Queensland they have been found in the Barron, Coen, Dulhunty, Embley, Endeavour, Jacky Jacky, Jardine, Johnstone, McIvor, Moresby, Mulgrave, Murray, Olive, Russell, Tully and Wenlock Rivers, plus the smaller coastal streams. They have also been found in oligotrophic sand dune lakes in the Cape Flattery and Shelburne Bay region. The known New Guinean distribution includes the Aru Islands, the Pahoturi, Fly (Elevala River, Lake Bosset) and Bensbach (Torassi) river systems. They probably occur elsewhere along the southern coast of New Guinea that has suitable habitat.

*Pseudomugil gertrudae* are found in small creeks, lagoons, billabongs, swampy marshes and rainforest streams, often associated with dense aquatic vegetation, woody debris and leaf litter. They are almost exclusively found in vegetated lagoons and backwaters with clear water where they can be seen swimming in the shallow waters along the margins. Floating species of waterplants or bottom rooted emergents with floating leaves occur in most, if not all, of their natural habitats. Substrates are usually mud or silt, and there is an abundance of water plants growing to the surface around the margins. Sometimes they may have water plants growing in the deeper water in the middle. Lagoons often have a thick layer of leaf litter around the margins. They are seldom found in turbid lagoons, even when there are abundant water plants.

The water in some habitats is often intensely discoloured by tannic acids leached from decaying vegetation. These 'blackwater' habitats are generally acidic, with *p*H levels from 3.9 to 6.8, have low conductivity (dissolved ions), and vary in their dissolved organic matter, ionic composition, and colour. Alkalinity and hardness levels are very low. Factors contributing to these variations are age, formation, layers of low permeability and peats, proximity to the sea, surrounding vegetation, and the extent to which leaf litter accumulates and decays in the water. However, habitat conditions can vary substantially and *Pseudomugil gertrudae* have been collected from natural habitats within the following range of water conditions:

Temperature: 12–34° Celsius *p*H 3.68–9.4 Conductivity: 12–646 µS/cm Hardness 0–320 ppm Alkalinity 2–180 ppm

#### Biology

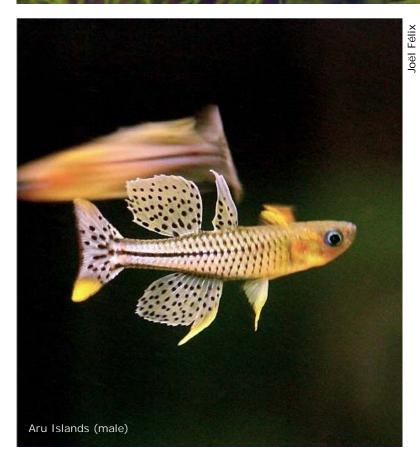
Very little is known about the biology or ecology of this species in their natural environment. Most information is mainly based on aquarium observations. *Pseudomugil gertrudae* are a relatively short lived species in the wild and most die in their first year, even if conditions are ideal. Mortality becomes more predictable after the first two years of life. Natural factors affecting them include disease, parasites, predation and competition for food and space. Females may only spawn once, usually at around one year of age, rarely living to spawn a second season. Males often live around two years. In captivity, life expectancy can increase up to four years if appropriate aquarium conditions are provided.

In their natural environment spawning usually commences during the early-wet season from October to December, which often causes an increase in plankton and other foods. The young are spawned when food is plentiful and when aquatic plant communities are most dense, affording them protection from predation. They are an egg-scatterer, generally spawning amongst aquatic plants and grasses. In captivity pre-spawning behaviour is initiated adjacent to the chosen spawning site by the male actively pursuing a female. Males displayed during the daylight hours with the peak of activity occurring in the late morning and early afternoon when water temperatures are maintained around 24-28°C. When actively pursuing a female the male display from a side on position, with spread dorsal and anal fins, while at the same time raising and lowering his pectoral fins. During this procedure the colour intensifies in both sexes with the spots on the body and fins becoming very dark.

Eggs have adhesive filaments that attach to aquatic plants or amongst the strands of the spawning mops, and sometimes even in the substrate. Spawning has been observed in ponds with eggs being recovered from the roots of floating duckweed. Spawning often continues throughout the day, with each female releasing up to 10 or 12 eggs. Spawned eggs are relatively large, adhesive, negatively buoyant in freshwater and average  $1.3 \pm 0.5$  mm in diameter, and are usually clear to light amber in colour.

Eggs are best left attached to the spawning medium to minimise handling stress and removed from the spawning tank and placed in another aquarium for incubation and larval rearing. Maintain a constant temperature  $\pm$  1°C and gently aeration. Hatching will begin after an incubation period of around four to nine days depending on temperature. Temperature is one of the major factors that influences the embryonic period of blue-eyes. Although Pseudomugil gertrudae are only small their newly hatched larvae are rather large. The hatching size of the larvae is around 3-4 mm. Hatched larvae are well developed and competent swimmers. Upon hatching the larvae swim at the surface of the water, generally within the upper 1-cm water layer. The mouth is well developed and functional, and they begin feeding within hours of hatching. They can be fed finely powdered dry foods, newly-hatched brine shrimp, copepods, phytoplankton and microworm. Pseudomugil gertrudae grow fairly rapidly and reach maturity in about three months; at a size of around 15 to 20 mm in length.





*Pseudomugil gertrudae* have a generalised diet in their natural habitat consuming aquatic prey items such as small crustaceans, various aquatic insects and invertebrates, tadpoles, algae, diatoms and small quantities of terrestrial insects such as flies. In captivity, they can be fed live or frozen foods such as daphnia, copepods, mosquito larvae or brine shrimp. Microworm and other small worms are also an excellent food. Flake food or small bite-sized pellets can also be fed with success however for best results some supplementary feeding with live or frozen food is required.





#### **Pseudomugil inconspicuus** Roberts, 1978

Inconspicuous Blue Eye

#### **Species Summary**

Pseudomugil inconspicuus is a small slender-bodied species, usually not exceeding 35 mm in length. They have two dorsal fins, very close together, the first much smaller than the second. The body colouration is translucent bluish with some scattered melanophores and clear to slightly yellowish fins. A uniformly thin, uninterrupted, longitudinal line of black pigment extends from just above the origin of the first pectoral fin ray to the base of the caudal fin. This line is enhanced with metallic blue reflective scales above and below. P. inconspicuus show only slight sexual dimorphism involving the dorsal and anal fins only. Mature males have a slightly larger first dorsal fin than females (sometimes with a short filamentous extension). The second dorsal fin often has a short filamentous extension as well. P. inconspicuus does not seem particularly closely related to any other described Pseudomugil. They were scientifically described by Tyson R. Roberts in 1978 from specimens collected from a small mangrove-lined tributary of Guiavi Creek at the mouth of the Fly River, Papua New Guinea in 1975.

In their natural environment spawning usually commences during the early-wet season from October to January. They are a planktivorous species and adapt well to freshwater environments.

#### **Distribution & Habitat**

*Pseudomugil inconspicuus* are probably widely distributed in estuarine and coastal freshwater habitats across northern Australia and southern New Guinea, but have escaped notice due to their small size and largely inaccessible habitats. They are known from several locations in New Guinea and probably extend from the Kikori River to the Vogelkop Peninsula. They have been collected in the Fly River, Bintuni Bay, Timika region, Aru Islands and Bristow Island, near Daru. In Australia they have been found in scattered localities around Darwin and Kakadu regions in the Northern Territory. They have also been collected from Jacky-Jacky Creek on Cape York Peninsula.

*Pseudomugil inconspicuus* are euryhaline and tolerate a wide range of ecological conditions. Although more commonly found in small brackish estuarine creeks, they also inhabit pure freshwater habitats, especially during the wet season. During the wet season, freshwater flowing into these habitats dilutes the waters to fresh. Water thus varies from saline through brackish to fresh. However, habitat preference appears to be mangrove-lined muddy brackish creeks, where they are commonly found in large numbers. They have been found in hypersaline waters (28–40 ppt) and at temperatures of 22–39° C. They have been observed sheltering among submerged roots or inundated leaves and branches, often in muddy waters. They are frequently seen swimming in midwater rather than near the surface. They are sympatric with *Pseudomugil cyanodorsalis* over part of their range.

#### Remarks

*Pseudomugil inconspicuus* is currently rare in the aquarium hobby and as far as I know, have never been bred in captivity. So far, they have proved rather delicate when being collected and difficult to maintain in captivity. Captured specimens carry high parasite loads and seem to waste away slowly.









# Pseudomugil ivantsoffi

Allen and Renyaan, 1999 Ivantsoff Blue Eye

#### **Species Summary**

The head and body of male Pseudomugil ivantsoffi is semitransparent, often with a bluish or reddish hue. Iris of eve intensely blue. The opercle, abdomen and swim bladder region are silvery. The upper half of first dorsal and anterior half of second dorsal fins, and edge of anal fin are bright red. The dorsal and ventral contour posterior to second dorsal and anal fins edged with similar red; the upper and lower third of caudal fin also red. The pelvic fins are pinkish. The edges of some scales on abdomen with narrow black margins. Females overall semitransparent, lacking bright red shades of male; opercle, abdomen and swim bladder region silvery; some scales on anterior half of body with faint, fine black margins; fins mainly translucent to faintly yellowish, the outer tips of the first and second dorsal fins yellow. This is a small species, slender and laterally compressed, growing to a length of around 3 cm SL. Named "ivantsoffi" in honour of Walter Ivantsoff, of Macquarie University, Sydney, Australia, in recognition of his valuable contributions to the knowledge of atherinoid taxonomy.

This fish was initially identified as *Pseudomugil reticulatus*, based on the close resemblance of females from the Timika area to the single known example of that species collected about two kilometres east of Ayamaru (Ajamaru) Lake in the centre of the Vogelkop Peninsula. However, collections in 1999 near the type locality of *Pseudomugil reticulatus*, which lies some 900 km northwest of Timika, revealed that the two populations are distinctive.

#### **Distribution & Habitat**

*Pseudomugil ivantsoffi* have been found in the tributaries of the Ajkwa, Iwaka and Kopi Rivers in the Timika-Tembagapura region of West Papua. The habitats consist of small (1-2 metre wide) shallow, slow-flowing streams in dense rainforest. Water is generally clear, but some are tannin-stained, with sparse aquatic vegetation. Bottom conditions included sand, gravel, cobble, and rocks. Water temperature and *p*H values ranged from 24–28 °C and *p*H 6.7–7.8 respectively. Other fishes collected from these habitats include *Pseudomugil pellucidus*, *P. novaeguineae, Melanotaenia goldiei, M. splendida rubrostriata*, and *M. ogilbyi*.

The Timika region includes the following rivers: Kamora River (Kamora, Tuaba, Wataikwa, Iwaka) Wania River, Tipuka River, Ajkwa River, Minajerwi River (Minajerwi, Kopi, Aimua) Mawati River, Otokwa River, Mamoa River (Mamoa, Seruka) Atuka River (Atuka, Wapuka) [The Atuka River meanders north to its junction with the Kamora River, very close to the village of Mioko.]

The dark colouration in 'blackwater' streams is due to the presence of organic substances. This black or tea colouration (these rivers are called kali kopi in Indonesian, or coffee stream – kali is Indonesian for river) is quite common in the area, due to extensive heath forest. These streams start in the





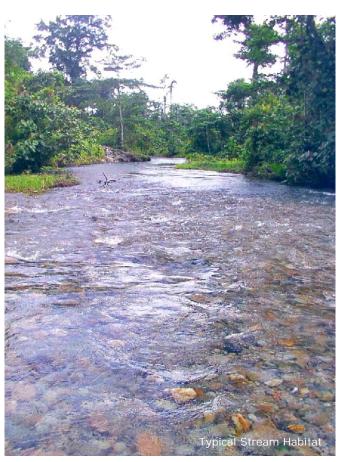
heath and are shorter and narrower than the rivers which begin in the mountains and have enough volume from rainfall to dissolve the colouration. The blackwater streams all drain into the rivers.

#### Remarks

In 2004, Iain Wilson and Charles Nishihira collected live specimens of *Pseudomugil pellucidus*, *P. ivanstoffi* and *Melanotaenia goldiei* from the Iwaka River (Deky Creek), which is a tributary of the Kamora River. *M. goldiei*, *M. s. rubrostriata*, *P. novaeguineae*, *P. pellucidus* and *P. ivantsoffi* were collected in small streams of the Kopi River, which is a tributary of the Minajerwi River.

A different colour variety of Pseudomugil pellucidus was collected from the Kamora River catchment. Males had spotted red and black on the dorsal fin. The anal fin colouration was also different with lots of yellow. They were collected from two streams only metres apart and actually intersect further downstream. A yellowcoloured form of Melanotaenia splendida rubrostriata was also found. It may have been an environmental issue as their stream was red with iron seepage from the mine site. ~ Iain Wilson

Live specimens have also been collected Heiko Bleher. Although a very attractive species if kept under suitable conditions, they are rarely seen in the retail aquarium trade.







#### Pseudomugil majusculus

Ivantsoff and Allen, 1984 Cape Blue Eye

#### **Species Summary**

*Pseudomugil majusculus* have a moderately compressed and elongated body that usually doesn't exceed 5 cm. The body colour is mainly pale yellow with fine, dark scale outlines. There is a horizontal row of about 10 vertically elongated, white spots along the middle of the side. The lower edge of the breast is yellow and there are white margins on the anal and second dorsal fins. Mature males have a slightly larger first dorsal fin than females (sometimes with a short filamentous extension). *P. majusculus* is similar in appearance to *P. signifer*.

*Pseudomugil majusculus* can be distinguished from *P. inconspicuus*, *P. novaeguineae* and *P. paludicola* by a low predorsal scale count and the more anterior position of the origin of the first dorsal fin; from *P. gertrudae*, by the coloration of the latter and the anal fin ray count; from *P. tenellus*, by the gill raker, anal fin ray and transverse scale row counts; and from *P. signifer*, by the gill raker count and the gill raker length.

#### **Distribution & Habitat**

*Pseudomugil majusculus* were initially collected in 1979 by Bruce Collette, an American ichthyologist, from brackish water on the northern coast of New Guinea, near Cape Ward Hunt, approximately 190 kilometres northeast of Port Moresby. They have also been collected from a freshwater stream on Tagula (Sudest) Island, in the Louisiade Archipelago. Tagula Island is about 280 kilometres southeast of Papua New Guinea. It is the largest island of the archipelago with an area of 800 km<sup>2</sup>. They are probably euryhaline and inhabit a wide range of natural habitats including mangrove swamps, marine estuaries and freshwater streams.

#### Remarks

Live specimens were collected in June 1993 from Tagula Island, but they are currently rare (or non-existent) in the aquarium hobby. It was named "*majusculus*" (Latin), meaning somewhat larger or greater, thus implying that this species grows to a larger size than other species of *Pseudomugil*.





**Pseudomugil mellis** Allen & Ivantsoff, 1982 Honey Blue Eye

#### **Species Summary**

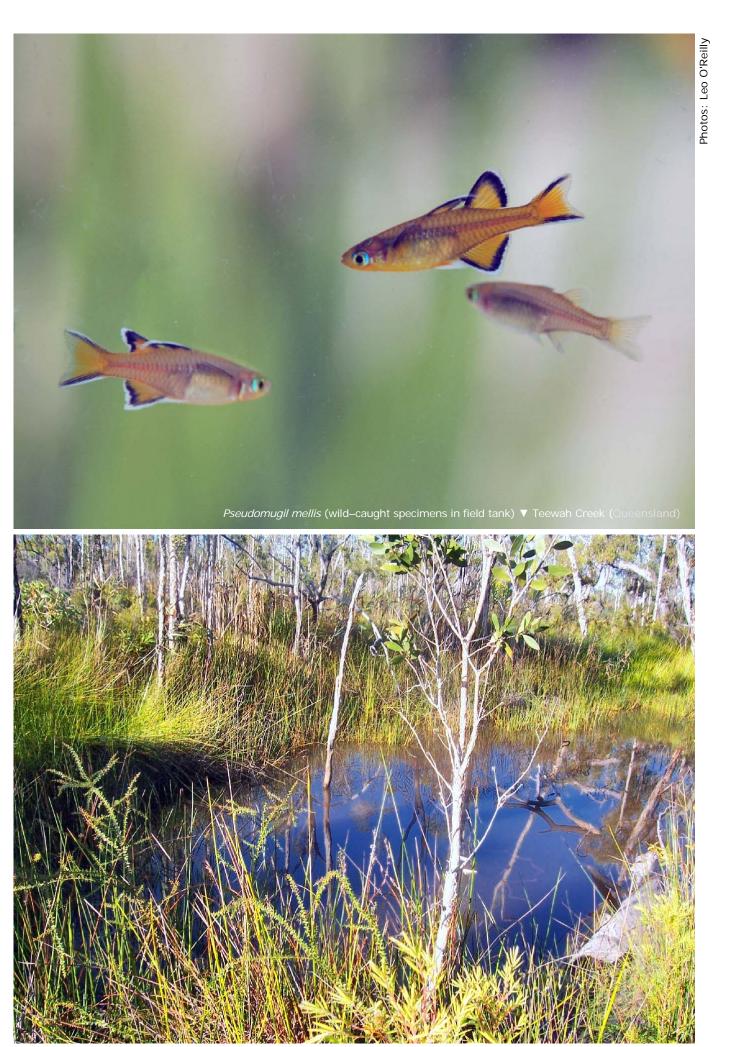
*Pseudomugil mellis* is a small freshwater species endemic to Australia. They have a moderately compressed and elongated body; usually not exceeding 40 mm, but are more commonly found at lengths between 25 and 30 mm. Males are honeycoloured with the first two rays of the dorsal and anal fins black with creamy-brown centres and outer white margins. The body scales are lightly edged with black forming an attractive latticework pattern. They have two dorsal fins, separated by a small gap, the first much smaller than the second. Males are easily distinguished from females by their brighter colours and longer and more elongated dorsal fins. Female and juveniles have a plain light-amber coloured body with small unmarked translucent fins. *P. mellis* was formally described by Gerald Allen and Walter Ivantsoff in 1982.

#### **Distribution & Habitat**

*Pseudomugil mellis* have a patchy and restricted distribution in southern Queensland, extending from about 65 km north of Brisbane to Maryborough, including Fraser Island. They have also been collected at the northern end of Dismal Swamp, south of Shoalwater Bay. Although their range has been severely reduced, they are currently known from about 19 locations on both the mainland and Fraser Island. It is still relatively abundant in the Noosa River catchment and Fraser Island localities. Lacustrine populations occur in seven lakes (six on Fraser Island and one at Cooloola). *P. mellis* is known to co-occur with *P. signifer* in Lake Wabby and Bool Creek on Fraser Island, and Schnapper Creek and Big Tuan Creek on the mainland. Big Tuan Creek is a small coastal creek about 5 km south from the mouth of the Mary River. Most populations are totally isolated from one another.

The former distribution of P. mellis may have extended from Woodgate in central Queensland extending down to the Myall Lakes, in the northern coast region of New South Wales, including the offshore sand islands. This range has been fragmented by residential development, forestry plantations and agriculture. Most existing locations have been similarly affected by changes within their catchments. While a number of suitable habitat streams still exist in the region between Brisbane and Noosa, it is likely that most will show substantial differences in water chemistry in comparison to similar undisturbed streams. Its abundance within this area has been drastically reduced and it now only occurs as a number of isolated populations where there is suitable remaining habitat. Its continued existence is being threatened by continuing urban development and the spread of the introduced mosquitofish (Gambusia holbrooki).







*Pseudomugil mellis* are typically found in slightly acidic and tannin-stained water in coastal heathland (wallum) swamps and streams. Wallum is a restricted region and, thus, any species of fauna confined to it are restricted in their distribution. However, they can also be found in clear water habitats. They inhabit freshwater dune lakes, creeks, swampy areas and wetlands. These waterbodies are characterised by low pH (4.4 to 6.8), and very low dissolved mineral salts.

The first time I collected this species from the wild I recorded a water hardness of 10 mg/L CaCO<sub>3</sub>, *p*H 5.8, and conductivity 170  $\mu$ S/cm. They can tolerate wide fluctuations in stream temperature, from 11°C in winter to 38°C in summer. They usually occur where there is little or no flow over sandy or muddy substrates with abundant emergent and submerged aquatic vegetation. The presence of aquatic vegetation appears to be essential for this species for shelter, foraging, spawning and the growth of larvae and fry. They may occur with *Rhadinocentrus ornatus*, *Nannoperca oxleyana*, *Melanotaenia duboulayi* and other small bodied native fishes.

#### Breeding

The reproductive biology of *Pseudomugil mellis* has been well documented from studies of wild populations and from specimens maintained in captivity. The following summarises much of that information.

In their natural habitat females ready to release eggs have been found from September to January. Spent fish (examination of the ovaries indicates that only a small percentage of eggs are sufficiently developed to the stage at which they are able to be fertilised) have been collected from November to April. The number of spent males and females was found to be highest in January. This indicates that *P. mellis* begin to spawn in the wild as early as September/ October and that most members of the population have ceased to spawn by January-February. Spawning in the wild occurs at temperatures in the range of 26–28°C. Females spawn at around 17–22 mm in size with the larger females producing more eggs than smaller females.

The small maturation size and the use of batch spawning over successive days were considered to be reasons for the successful recruitment of this species in the wild. Recruitment is further maximised by several mechanisms that decrease predation: territorial defence of the spawning site by the male, direct pairing with a short courtship, and larvae that swim at the surface and seek cover in the meniscus of floating objects when predators attack.

In captivity males exhibit territorial behaviour guarding the spawning site from intruding conspecifics. Prespawning behaviour is initiated adjacent to the spawning site by the male raising his fins and swimming in a zigzag pattern to block the





female's retreat. Spawning usually occurs in late morning and early afternoon with the female entering the spawning site first, followed by the male. After spawning, the male will continue to display, defending his territory and thus the fertilised eggs. Spawning usually commences at around 10–12 months of age when water temperatures exceed 20°C and the fish are about 20 mm in size. Sexual dimorphism is evident, with adult males having larger and more colourful dorsal and anal fins. Males develop black and white fin marking with a golden bronze sheen to the body when displaying.

Females spawn 1-15 eggs each day for about 7-9 days, with 1-4 eggs at a time being released amongst aquatic vegetation or spawning mops. A total of 42-125 eggs can be released over that period. Eggs are demersal, adhesive and attached to aquatic plants or spawning mop. After spawning females rest for 4-9 days, during which time they formed schools. Water hardened eggs range from 1.29-1.64 mm in diameter, probably depending on the size of the female and have adhesive tendrils or filaments to attach them to the spawning site. Eggs hatch 12-14 days (288-336 hours) after fertilisation at a constant water temperature of 24°C. At slightly elevated water temperatures of 25–27°C eggs hatched between 5 and 8 days (112–190 hours). Larvae are around 3.60-5.00 mm SL at hatching and begin feeding at the surface after absorption of the yolk and oil droplets (3-80 hours after hatching). Juveniles feed mid-water or from benthic surfaces.

#### Remarks

*Pseudomugil mellis* currently has a conservation status listing as vulnerable. You can do your part in helping to preserve this species and their natural habitat by breeding and maintaining captive populations. Collecting wild specimens for the hobby can have a direct impact on their population numbers to the extent that they could become locally extinct. It is becoming increasingly difficult to find them at the popular spots in Tin Can Bay. We know that their populations undergo large fluctuations associated with seasonal recruitment and if they are over-collected during a period of low population numbers, they may never recover and we could well see them disappear from that location. Habitat destruction or alteration in some areas has also favoured the spread of the aquatic terrorist, *Gambusia holbrooki*.

The secret in maintaining long-term captive populations of *Pseudomugil mellis* is to constantly breed them. There is a reduction in the frequency and intensity of spawning activity in fish over one year of age. If you fail to notice the change they become too old and then you just lose them. Try to obtain young specimens and breed them early and regularly and you will always have some around to enjoy. Because each female only lays a few eggs each day, it can take a while before you have significant numbers. If possible, start with 6-12 juveniles rather than adults pairs.





## Pseudomugil novaeguineae

Weber, 1907 New Guinea Blue Eye

#### **Species Summary**

*Pseudomugil novaeguineae* have a yellowish semi-translucent body with a thin dark mid-lateral line on the sides. The body scales are lightly edged with black forming an attractive latticework pattern. Common maximum size for this species is about 4–5 cm SL. Males have elongated anterior first, second dorsal and anal fin rays. Adult males have a red first dorsal spine and the outer margin of the first dorsal fin is also red. The second dorsal and anal fins, as well as the lower lobe of the caudal fin have white, yellow or red margins. The colour can change rapidly back and forth. The adult male can also be recognised on the blackish anterior edge of his dorsal fin and the black streaks that adorns both the upper and the lower edge of the caudal fin.

Specimens collected from the Fly River in New Guinea have been reported as having a transparent body with glistening bluish or violet colour on the head and abdomen. The eye has a faint gold ring around the pupil with the iris silvery or faintly blue. The second dorsal fin of males is either clear, as in females or carmine red. Due to the wide geographical range of this species the colours and markings on the body and fins can be variable. Red coloured eggs are laid by *Pseudomugil novaeguineae*.

#### **Distribution & Habitat**

This species has patchy distribution in central southern New Guinea between the Fly River, Papua New Guinea and Etna Bay, West Papua. They have also been collected from the Aru (Aroe) Islands in the Arafura Sea. Scientific specimens of this species were first collected from the Lorentz (Noord) River by Hendrikus Albertus Lorentz during the Dutch Expedition to New Guinea in 1907. Lorentz participated in three expeditions to Dutch New Guinea. The first expedition was in 1903, led by A. Wichmann. Lorentz led expeditions in 1907 and 1909-1910.

*Pseudomugil novaeguineae* inhabit small clear rainforest streams; well shaded but with occasional open patches exposed to sunlight. A temperature of  $24^{\circ}$  Celsius and *p*H 7.8 were recorded at one collection site in a tributary of the Ok Smak River, about 35 kilometres north of Kiunga. *Kiunga ballochi* sometimes occurs in the same streams.

#### Remarks

Around 1976 and 1989 live specimens were collected and taken back to Europe. In 2004, Iain Wilson and Charles Nishihira collected live specimens of *Pseudomugil pellucidus*, *P. ivanstoffi* and *Melanotaenia goldiei* from the Iwaka River (Deky Creek), which is a tributary of the Kamora River. *M. goldiei*, *M. s. rubrostriata*, *P. novaeguineae*, *P. pellucidus* and *P. ivantsoffi* were collected in small streams of the Kopi River, which is a tributary of the Minajerwi River in West Papua. There have been a number of other collections, but *P. novaeguineae* is still not widely available in the aquarium hobby.







#### Pseudomugil paludicola

Allen and Moore, 1981 Swamp Blue Eye

#### Species Summary

*Pseudomugil paludicola* are a small species growing to a maximum size of around 4–5 cm. They have a translucent body, with silvery head and abdomen. The pelvic fins are yellowish. The edge of the second dorsal fin in males is yellowish. In females, the caudal fin base is yellowish and the anterior portion of the anal fin is pale yellow. Males are easily distinguished from females by their brighter colours and longer and more elongated dorsal fin. Females are smaller and have smaller fins but lack the colours of the males.

There are probably a number of geographically isolated populations that have their own distinctive colouration. A variety collected from the Sorong area in West Papua have a semi-transparent body and when viewed under overhead lighting the dorsal area of the male's body has a beautiful blue coloration much like *Pseudomugil cyanodorsalis* but not as intense. The pelvic, pectoral and anal fins show a slight hint of yellow. A yellow coloured form has been reported from the upper Kikori River in Papua New Guinea.

#### **Distribution & Habitat**

*Pseudomugil paludicola* were first collected in 1973 by R. Moore in a mangrove creek near Bulla at the mouth of the Morehead River, Papua New Guinea. They are also reported as being very common in the swamplands of the lower Pahoturi River. Other collections have been in the Sorong region in West Papua and the Kikori and Binaturi river systems in Papua New Guinea. However, they are probably widely distributed in coastal streams over much of southern New Guinea.

*Pseudomugil paludicola* is a stream dwelling species found in clear coastal rainforest streams and swamps, often associated with thick aquatic vegetation. A temperature of  $26.8^{\circ}$ C and *p*H 7.6 were recorded from one collection site. They are probably euryhaline and tolerate a wide range of environmental conditions.

#### Remarks

This species was named "*paludicola*" (Latin for "swamp dweller") with reference to its habitat. In 1979 Gerald Allen returned to Australia with live specimens but unfortunately they departed this life before any were bred and distributed in the hobby. Another collection was made in the Sorong area and small populations were established in the Europe and North America. Eggs were imported into Australia in 1996 whereupon they were bred, but again, failed to become established in the hobby.





#### **Pseudomugil paskai** Allen and Ivantsoff, 1986

Paska's Blue Eye

#### **Species Summary**

*Pseudomugil paskai* is a small slender-bodied species, usually not exceeding 35 mm in length. Two dorsal fins, very close together, the first much smaller than the second. Males have a semi-translucent body colour that is bluish ventrally and yellowish above the mid-lateral line, with narrow dark scale outlines. The fins are generally translucent with white or yellow margins and scattered oval black spots. Lobes of the caudal fin have either white, yellow or reddish tips. Pelvic fins are yellowish with elongated anterior rays. Females do not have spots on their fins and do not show the colours of the males.

#### **Distribution & Habitat**

*Pseudomugil paskai* were first discovered by David Balloch and Gerald Allen in 1983. They are very similar to *Pseudomugil gertrudae*, but differ in colouration and fin shape. Known only from a few locations between Etna Bay and the Fly River system in New Guinea. However, they are probably widely distributed in coastal streams over much of southern New Guinea. They are a stream dwelling species occupying slow-flowing muddy or teacoloured rainforest streams.

Aquatic vegetation is generally abundant. The pH and temperature ranges recorded at the collection site were 6.0–6.5 and 25–26° Celsius.

#### Remarks

Live specimens were collected in 1983 by Gerald Allen and returned to Australia, but failed to become established in the hobby. However, further live collections were made and small populations were established in the hobby. In 1996 eggs were imported into Australia from Europe, but again, they failed to become established. Their current status in the hobby is unknown.







## **Pseudomugil pellucidus**

Allen, Ivantsoff, Shepherd and Renyaan, 1998 Transparent Blue Eye

#### **Species Summary**

*Pseudomugil pellucidus* was described on the basis of 30 specimens collected from tributaries of the Iwaka and Kopi rivers in the vicinity of Tembagapura, West Papua. The species is very closely related to *Pseudomugil novaeguineae* but is distinct from the latter on the basis of greater number and length of first dorsal fin spines, number of anal rays, shape of vomer, basibranchials, pectoral girdle, urohyal, and anal pterygiophores. *Pseudomugil pellucidus* is also different in colouration, and rather transparent. It was named "*pellucidus*" (Latin), meaning clear or transparent.

*Pseudomugil pellucidus* is a small slender-bodied species, usually not exceeding 3–4 cm in length. The head is silver-reddish blending into a silver-reddish opercle and peritoneum. The rest of the body is quite transparent. The swim bladder is transparent and obvious. The upper edge of midlateral band is neon-red, while the rest of band is solid black, extending to the hypural joint and then fanning out onto the caudal as thin black stripes on each of the mid-caudal rays. The first dorsal fin is jet-black, with a small flash of orange on the first dorsal spine.

The second dorsal fin is dusky with rays suffused with melanophores to form black stripes; posteriorly edged with brilliant orange. Thin black and orange bands extend along entire length of the anal fin. The upper half of caudal has small flashes of orange. The edges of the body scales are outlined heavily in black above the midlateral band and lightly below. The iris has an orange hue. Individuals possess a pupil-sized white spot on top of the head which is readily visible when observed from the stream bank. Reddish coloured eggs are laid by *Pseudomugil pellucidus*.

In 2004 a new colour variety of *Pseudomugil pellucidus* was collected from Kali Meyon. Males have spotted red and black on the dorsal fin. The anal fin colouration was also different with lots of yellow. Two streams were collected that are only about 15 metres apart and intersect about 150 meters downstream and each stream contained a different colour variation (*Iain Wilson pers. comm.*)

#### **Distribution & Habitat**

*Pseudomugil pellucidus* have been collected from tributaries of the Iwaka and Kopi Rivers in the Timika-Tembagapura region of West Papua. They are generally found in small shallow streams usually slow flowing through dense rainforest, always in clear water which may be deeply stained with tannin. Aquatic vegetation was sparse or absent at the collection sites.





The bottom substrate is variable from sand, to gravel, pebble and rocks. Water temperature 24–28°C, pH 6.7–7.8. *Pseudomugil pellucidus* swims close to the surface, in contrast with its sympatric congener, *Pseudomugil ivantsoffi* which is found in mid-water or near the bottom. Other fishes collected from these habitats include *P. ivantsoffi*, *P. novaeguineae*, *M. goldiei*, *M. splendida rubrostriata*, and *M. ogilbyi*.

These 'blackwater' steams are dark coloured due to the presence of organic substances which drain heath forests. This black or teastained colouration (these rivers are called kali kopi in Indonesian, or coffee stream – kali is Indonesian for river) is quite common in the area, due to extensive heath forest. These streams start in the heath and are shorter and narrower than the rivers which begin in the mountains and have enough volume from rainfall to dissolve the colouration. The blackwater streams all drain into the rivers.

#### Remarks

Live specimens were collected for the aquarium hobby in 1999 by Heiko Bleher. In 2004, Iain Wilson and Charles Nishihira collected live specimens of this species from Kali Iwaka (Deky Creek) and Kali Kopi, along with specimens of *Pseudomugil novaeguineae*, *Pseudomugil ivantsoffi*, *Melanotaenia goldiei* and *Melanotaenia splendida rubrostriata*.

Although a very attractive species if kept under suitable conditions, *Pseudomugil pellucidus* are rarely seen in the retail aquarium trade and are mainly kept by a few aquarists who are principally interested in rainbowfishes.









#### **Pseudomugil reticulatus** Allen and Ivantsoff, 1986 Vogelkop Blue Eye

#### **Species Summary**

*Pseudomugil reticulatus* is a small species, slender and laterally compressed, growing to a length of around 3–4 cm SL. They have a translucent greenish-brown body; abdomen and swim bladder region silvery. The ventral part of the breast is yellow. The upper half of first dorsal fin and anterior half of second dorsal and edge of anal fin is coloured brick-red. The dorsal and ventral contour posterior to second dorsal and anal fins are edged with a similar red. The upper and lower third of caudal fin also red. Head with red tinge, ventral fins pinkish-red. The eyes are intensely blue. The edges of some scales on abdomen are edged with black. The females are similar in colouration. They superficially look very similar to the Redfin Blue Eye (*Scaturiginichthys vermeilipinnis*). Eggs are orange-red in colour.

#### **Distribution & Habitat**

Currently known only from the Ajamaru Lakes region in Vogelkop Peninsula, Irian Jaya. Previous records of this species from elsewhere in New Guinea are in error. They were collected about two kilometres east of Ajamaru Lake in the centre of the Vogelkop Peninsula. The lakes are located at the headwaters of the Ajamaru River which drains into the Kais



River, eventually flowing into the Ceram Sea to the south. Habitat variable, relatively clear shallow water, with abundant vegetation. The lakes and streams are alkaline with *p*H always slightly above neutral (7.1–7.6). Water temperate about 24–28° C. Co-occurs with *Melanotaenia boesemani*.

#### Remarks

Live specimen were collected by Heiko Bleher in 1998. However, they are rarely seen in the retail aquarium trade and are mainly kept by a few aquarists who are principally interested in rainbowfishes.





## **Pseudomugil signifer**

Kner, 1865 Pacific Blue Eye

Atherina signata Gunther, 1867 Atherinosoma jamesoni Macleay, 1884 Atherinosoma signata Ogilby, 1886 Pseudomugil signata Ogilby, 1896 Pseudomugil signatus Jordan & Hubbs, 1919 Pseudomugil signatus affinis Whitley, 1935 Pseudomugil affinis Munro, 1958

*Pseudomugil signifer* is a small, colourful blue-eye species inhabiting freshwater streams and estuaries in coastal drainages along much of the east coast of Australia and offshore islands. They are the most common and abundant freshwater/estuarine species along the east coast of Australia.

They were first discovered near Sydney, New South Wales in the 1860's and were the first blue-eye species to be scientifically described. A few years' later specimens were collected from northern Queensland. Various name changes followed, and in the late nineteenth and early twentieth centuries, they were separated into two species, the northern *Pseudomugil signatus* and the southern *Pseudomugil signifer*. Also, it was suggested that a population resident on the offshore Low Isles and some northern mainland populations were distinctive and were afforded sub-specific status as *Pseudomugil signatus affinis*. The various populations exhibit remarkable morphological variation throughout their range, which is evident from their confused taxonomic history. In 1979, researchers using electrophoretic analyses and a large sample of specimens taken from 14 localities along the east coast determined that all populations were scientifically indistinguishable from one another. Although they did recognise that there were some slight differences. This, however, is inconsistent with recent genetic research that indicated there are extensive differences among the various populations.

The research provided a good reason for suggesting that *Pseudomugil signifer* may represent at least two distinct species. Support for this contention is not only consistent with some of the previous taxonomic designations, but is also in keeping with recent behavioural studies. Breeding experiments showed that fish from opposite ends of the species range would not interbreed. In Queensland, there are two major geographical populations, those north and south of the Herbert River, with an intermediate form in the Townsville area.

It has long been the belief of native fish hobbyists in Australia that the various forms of *Pseudomugil signifer* are different, particularly the northern and southern populations. Also that the variety found in the Townsville region is different. I maintained and bred populations from Harvey and Lacey Creeks in north Queensland, the Ross River variety from Townsville, and a number of different populations from southeast Queensland over a period of 20 years. However, you only have to maintain these species for a very short time before you realise that they not only look different, but that they behave differently as well. Therefore, they should be maintained and bred within their own localised groups and it is wise not to interbreed the various geographical varieties.

Pseudomugil signifer is one of the most readily identifiable and ubiquitous members of the Australian fish fauna. They have a moderately compressed and elongated semi-transparent body that can vary in colour from pale olive, yellow to bluish, with fine, dark coloured scale outlines on the upper body. They have two dorsal fins, very close together, the first much smaller than the second. As the common name suggests, the iris is blue. The operculum and belly region are silvery. There is often a midlateral row of 10-12 vertically-elongated white or reflective spots along the side of the body. The males are larger and more colourful than females. The males display spectacular fin embellishments that are rapidly raised and lowered during courtship and agonistic encounters with other males. Due to the wide geographical range of this species the colours and markings on the body and fins can be variable. There is also substantial inter-population variation in male body size and fin length. Male specimens from northern populations can reach 90 mm and females 65 mm, although they can be considerably smaller over much of their range. Southern populations rarely exceed 40 mm.

Male specimens from north Queensland populations (especially specimens from Harvey and Laceys Creeks) have extremely long extended filaments on both the dorsal and anal fins. These filaments are usually shed during capture and if not, once placed in the confines of an aquarium, are nipped off by the other fish. From my experience, these fin extensions never regrow or appear in captive populations. They have a silvery to vellowish body, with the elongated parts of fins blackish, also the margins of the second dorsal and anal fins. Edges of upper scales dark, and often a dark stripe along the side. The young are yellowish-brown with dark spots on the dorsal and caudal fins. A black band along the middle of sides with a similar but shorter dark band above and below it on the caudal peduncle. About six rows of dark spots along the scale rows, fading out posteriorly. Spawning males display a coppery-gold body colouration, particularly along the lower jaw region.

The first thing that is evident about the northern variety at *Pseudomugil signifer* is the size of these fish in comparison to the southern forms. The specimens I received from Laceys Creek in 1980 were about 60 mm in size and a number of the males had long extended filaments on both the dorsal and anal fins, extending past the tail. Another obvious difference is that northern males are also highly territorial and aggressive and will often kill sub-dominant males. Outside of breeding, aggression can be suppressed by maintaining them in reasonably sized aquaria in company with a small group of rainbowfishes.

The Ross River variety is a large, deep-bodied species with huge fins that are wide and long, but less intense in colouration. They usually have less body colour, but have a row of brilliant blue or purple reflective scales along the posterior section of the lateral line, often merging to form a continuous band, that is flashed on and off like a neon sign as they display to passing females. They are generally found in brackish reaches of the Ross River and surrounding streams. A similar form extends south, down at least as far as Eurimbula Creek just north of the Town of 1770.

Southern populations have a body colouration of translucent to olive-greenish above, canary yellow below; the caudal peduncle is tinged with red. There is sometimes a broad dark band from pectoral to tail. The first and second dorsal spines are long and white though blackish at the base, while the rest of the fin is translucent. The second dorsal has the front and exterior margins black while the rest of the fin is yellow or orange. The anal fin is similar to the second dorsal. The caudal fin can be clear to orange or yellow with the outer rays, tips of lobes, and sometimes the central rays blackish. The pectoral fins are clear to opaque, with upper rays black. The ventral fins are usually bright yellow or orange. As the name indicates, the eyes are a beautiful blue. Females are less colourful; have smaller rounded clear dorsal and anal fins with a dark line on the anterior edge of the second dorsal fin. Specimens collected from freshwater habitats generally have deep orange coloured dorsal and anal fins whereas specimens from saltwater or brackish water habitats have yellow fins. Fish from acidic, tannin strained streams in south-eastern Queensland also show deep orange on the dorsal and anal fins. However, colour is extremely variable and will depend upon the mood of the fish, water conditions and diet.

#### **Distribution & Habitat**

Pseudomugil signifer is the most widely distributed blue-eye in Australia. They have extensive distribution from Merimbula Lake just north of Eden on the southern coast of New South Wales to Cape York Peninsula including islands in the Torres Strait. They have also been found in the Embley and Mission rivers near Weipa on the west coast of Cape York Peninsula. They are abundant in freshwater habitats, and inhabit rainforest streams, riverine habitats and freshwater swamps, but do not usually penetrate far inland. They can tolerate brackish to fully marine conditions, being found in tidal mangrove creeks, estuaries and saltmarshes, and on several offshore islands, including Moreton Bay in south-east Queensland. They are frequently found in the waters of canal housing estate developments on the Gold Coast in south-east Queensland. Diadromous migration is not an essential requirement of the species. Rather, the species is characteristic of the estuarinefreshwater interface. They inhabit waters with a temperature range of 15-28° Celsius, and pH 5.5-8.3.

#### Keeping & Caring

Pseudomugil signifer has been maintained in the aquarium hobby for many years. David G. Stead published a report on Pseudomugil signifer in his book "Fishes of Australia ~ A Popular Systematic Guide to the Study of the Wealth within Our Waters" in 1905: "The Blue-eye is sprightly, vivacious and an active swimmer; being, in addition, very tenacious of life, and, therefore, embracing, in its little self, all the qualities which go to make up a desirable aquarium-fish. Its common name is derived from the blue-colour of the irides of both sexes. In general shape it is more Mullet-like than any other





Bernd Jung

species of the Atherinidæ. The male is far more handsome than the female; the second-dorsal, anal and caudal fins being greatly elongated; each being beautifully barred with yellow and black. The beautiful colours are particularly noticeable during the spawning season, which is the summer-time. The body is usually of a silvery tint on the sides, the back being somewhat greenish. If in very dirty waters, the body-colour is often a yellowish-brown, and only slightly silvery. But little is known in regard to the habits, and, nothing so far, in regard to the life-history, of this entertaining little Atherinid."

On May 31, 1911, David G. Stead exhibited examples of Pseudomugil signifer to a meeting of the Linnean Society of New South Wales. "These were part of a number obtained, during April, from Wamberal Lagoon, at a spot where the water was "sweet" or brackish. These were brought away in that water, and, on April 19th, one was placed in an aquarium of sea-water (of about three years' standing), and the others were put into a freshwater aquarium. All had done well up to the present; the one in salt water, equally with those in fresh. This is an interesting experiment, inasmuch as it demonstrates the power of this little species to withstand sudden changes in its surrounding element. The coastal lagoons are very rich in this species, and these lagoons become practically fresh, and very salty alternately; it is, therefore, greatly to the advantage of this (and other species of aquatic life present) if they can adjust themselves to the varying conditions."

In 1915, Albert Gale wrote in his book Aquarian Nature *Studies - "One of the most beautiful indigenous aquaria fish we* have in New South Wales, both in colour and markings, and at the same time the most shapely, is the little blue-eye (Pseudomugil signifer). The adults never exceed 2 inches in length, and its depth is symmetrical to its length. Among imported aquaria fish specimens, the Paradise fish and the Fighting fish are its only rivals. In spring time, the breeding season, the sexes are very readily distinguished; the male, as is always the case in the fish-world, attires himself in his wedding garb, his fins become more developed and expansive. His whole body assumes a maize tint, his fins the colour of old gold edged with black, having a very narrow margin mottled with gold and white. The eyes of the male form a very striking contrast with the general markings of the body, being azure or sky blue and bright and lively; the whole contour giving the fish the appearance of aquatic butterflies rather than that of fish. The males are far more resplendent in the breeding season than are the females. Their natural food is rotifers, and the various varieties of animalculae. They are very fond of mosauito larvae. In confinement they do well on coffee biscuit, occasionally varied with a little flesh food or gentles." [Gentles are the larvae of blow-flies - the first stage after leaving the egg.]

*Pseudomugil signifer* were introduced to the international aquarium hobby as early as 1932. During 1933 the Shedd Aquarium in Chicago, dispatched an expedition to Australia, which, in addition to the larger fishes that were the object of the trip, obtained a number of smaller specimens. Notably in this collection were *Hypseleotris galii*, *Hypseleotris compressus* and *Pseudomugil signifer*.

*Pseudomugil signifer* are an attractive species that are easy to maintain and breed in captivity and are hardy aquarium fishes despite their small size. Ideally, they should share their

aquarium with similar sized tankmates and be kept in small groups. Regardless of the water conditions of their natural habitat they will survive in most dechlorinated tap waters available to the home hobbyist. Even those collected from brackish and saltwater habitats will survive and breed in freshwater. For general aquarium maintenance water conditions can be as follows: pH 6.5–7.5, hardness <200 ppm and a temperature range of 20–28°C. Prolonged exposure to temperatures above 30°C may cause some casualties.

The diet of *Pseudomugil signifer* in their natural habitat consists primarily of small insect larvae, aquatic crustaceans, worms, zooplankton and phytoplankton. In captivity, they can be fed live or frozen foods such as daphnia, copepods, mosquito larvae or brine shrimp. Whiteworms and other small worms are also an excellent food. Flake food or small bite-sized pellets can also be fed with success however for best results some supplementary feeding with live or frozen food is required. Obviously, no single food will meet their needs at all life stages, and the best way to ensure that they are getting a well-balanced diet is to feed them as wide a variety of food as possible.

*Pseudomugil signifer* are a relatively short lived species in the wild and females may only spawn once, usually at around one year of age, rarely living to spawn a second season. Males often live around two years. In captivity, life expectancy can increase to four years or more if appropriate aquarium management procedures are employed.

#### Breeding

Not a lot is known about the breeding biology of Pseudomugil signifer in their natural environment. Most information is mainly based on aquarium observations. In their natural environment spawning usually occurs from October to January for southern populations, while the breeding season for northern populations can be year round. They are an egg-scatterer, generally spawning amongst aquatic plants and grasses. Males are territorial and engage in spectacular fin-flashing displays during contests with rivals over the acquisition and defence of spawning sites. In the wild, males maintain and guard these sites while females move between them, inspecting the males along the way. Females may swim along in small shoals or also swim past alone. Males do not venture far from their territories and rely on the females to swim past. Males will actively swim over and try and entice the females with conspicuous courtship displays (body tilted in a head-down position, raising and lowering his fins). If successful, the female will follow the male to his territory and spawn. In the wild they tend to spawn amongst stems and roots of marginal aquatic vegetation close to the water's edge. In laboratory research, males were found to prefer courting larger females presumably because female fecundity increases with body size.

Group spawning with multiple males and females is probably the preferred method for breeding. Several spawning mops should be provided for egg placement. It is advisable to provide a spawning mop for each male in the group. In addition, this method should provide you with more eggs. Spawning pairs or trios is possible but you may end up with damaged or dead females due to the male's aggression. Group spawning spreads this aggression.





In captivity pre-spawning behaviour is initiated adjacent to the chosen spawning site by the male actively pursuing a female with raised fins and swimming in a zigzag pattern to block the female's retreat. When receptive, the female will enter the spawning site first, closely followed by the male. Disinterested or non-gravid females move away and swim to the surface or hide amongst aquatic plants. Females do not always choose the dominant male/s and will usually achieve higher spawning success when they mate with their preferred choice. In the wild, a female has the choice of either spawning or fleeing. In captivity, the flight of the female is reduced or confined to the size of the breeding aquarium. Therefore, the size of the breeding aquarium is vitally important and should be appropriate for the variety being bred.

Males exhibit territorial behaviour towards conspecifics such as lateral fin-flaring displays and pursuits. They do not actively care for the eggs other than through defence of the spawning site. A male might cannibalise his own eggs, or defend his spawning site badly against other egg predators. During spawning the males colour intensifies with the fins becoming brighter and the body turning from silvery to a golden bronze colour. The females body darkens with the scales edged in black. Females usually shed 2 to 3 eggs (1.1–1.8 mm) each spawning with up to 18 eggs being laid per day. Eggs have adhesive filaments that attach to plants or spawning mops.

Spawning usually occurs in late morning or early afternoon. After spawning, the male will continue to display, defending his territory and thus the fertilised eggs. The eggs can be difficult to find in large mops and if the eggs are being "picked" from the mops, thorough checking is required if all the eggs are to be found. Eggs were usually found singularly or in groups of 2 or 3.

I found that *Pseudomugil signifer* seem to prefer a dimly lit aquarium for breeding. The number of eggs being laid was not as high as I expected. Perhaps some were being eaten, although this was never observed. Also free hatching larvae were never observed in the breeding aquarium. If your floating mops are not producing eggs, try mops without floats, and if you still can't find any eggs you might find that the fish are spawning in the gravel. You might be surprised how many larvae actually hatch out from the gravel.

Eggs were collected daily and placed in a 4 litre plastic container with the addition of methylene blue as a fungicide. Upon hatching the fry were carefully transferred to a 135 litre raising tub. I found that the larvae nearly always emerge from the eggs during the night or early morning. Water conditions of the fry raising tub were maintained at around the same chemistry of the breeding tank. Weekly water changes of 20% were provided.

Eggs will take around 12–17 days to hatch at a temperature range of 22–28° Celsius. Although *Pseudomugil signifer* are only small their newly hatched larvae are rather large. The hatching size of the larvae is around 4–5 mm. At around 15–20 mm the males began to show some fin colouration but the fins were still the same shape as the females. By 20–25 mm the elongated anal and dorsal fins are developed and the sexes should readily be distinguished. Rapid development of larvae continues with maturity being reached within 6 months with males at about 30 mm and 25 mm for females. Males show slender elongated dorsal and anal fins with yellow, orange and red markings, while the females and juveniles have short rounded clear dorsal and anal fins with the only colouration being a dark line on the anterior edge of the second dorsal fin.



#### **Pseudomugil tenellus** Taylor, 1964

Delicate Blue Eye

#### **Species Summary**

Pseudomugil tenellus is a small fish growing to a length of around 4-5 cm. Adult males generally have a translucent golden-brown body colour above the mid-lateral line and yellowish-brown with a silver sheen below. The mid-lateral line consists of a series of discontinuous silvery, reflective scales that become larger in older fish. The body scales are edged in black and form an attractive latticework pattern. They have two dorsal fins, very close together, the first much smaller than the second. The fins have a background colour of golden burnt-orange, with the outer margins light yellow; often edged with white. The second dorsal and anal fins have a semi-circular pattern of several small white spots. The caudal fin has a black margin that is fringed with white. The pectoral fins are fringed along the anterior edge with orange. As the common name suggests, the iris is blue. The operculum and belly region are silvery. However, colour can be variable and will depend upon the mood of the fish, water conditions and diet. Females and juveniles have a similar body colour but not as intense, and have much smaller uncoloured rounded fins with no markings. Females generally have a deeper body than the males whilst the adult males have larger dorsal, anal and pelvic fins. The differences in colour of the body and especially the larger size of the males' fins make the sexes of P. tenellus easily distinguishable.

#### **Distribution & Habitat**

*P. tenellus* was first collected from the East Alligator River near Oenpelli, in the Northern Territory during the American-Australian Scientific Expedition of 1948. They were reportedly abundant in large billabongs and creeks below escarpment waterfalls in the Oenpelli area. However, they were not scientifically described until 1964. They have patchy distribution throughout the northern areas of the Northern Territory, around the Gulf of Carpentaria to Cape York Peninsula in Queensland. In New Guinea they have been found in the Bensbach River and the Aru Islands; although I suspect that their distribution in southern New Guinea will be much wider.

In Australia, *P. tenellus* has been recorded from catchments of the Alligator, Blyth, Daly, Finniss, Howard, Liverpool and Mary river systems in the Northern Territory, where they are commonly found in riverine floodplain billabongs. They have also been collected from Leanyer Swamp, a tidal swamp north-east of Darwin; Rapid Creek, Benjamin Lagoon and a number of other minor streams in the Darwin region. In Queensland they have been collected in the Coleman, Edward, Jardine, Lockhart and Watson river systems; Jacky Jacky and Scrubby Creek (near Coen).

*P. tenellus* are usually found inhabiting coastal brackish or fresh waters. They are most common in the lower riverine floodplain swamps and in slow-flowing streams, generally in areas with





dense aquatic vegetation. They are usually found in the greatest numbers during the mid-wet season. Juveniles have been collected in all seasons with a peak in the late-wet to early-dry season. Juveniles are mainly found in floodplain billabongs. Larger juveniles can be found in mainchannel waterbodies. Adults are found in essentially the same habitats as the juveniles as well as in the upper reaches of freshwater streams. Both adults and juveniles have been collected in estuary and tidal salt marshes; presumably they can spend their entire lives in these brackish habitats. Water conditions recorded in their natural habitats are: Temperature 27–38°C; *p*H 5.0–7.1 and Conductivity 6–120  $\mu$ S/cm. This indicates that this species has a preference for warmer waters.

#### Keeping & Caring

*P. tenellus* are a relatively short lived species in the wild and most die in their first year, even if conditions are ideal. Mortality becomes more predictable for most fish after the first two years of life. Natural factors affecting them include disease, parasites, predation and competition for food and space. Females may only spawn once, usually at around one year of age, rarely living to spawn a second season. Males often live around two years. In captivity, life expectancy can increase up to four years if appropriate aquarium conditions are provided. The key for successfully maintaining *P. tenellus* is excellent water conditions. This can be easily provided with regular partial water changes.

P. tenellus have a generalised diet in their natural habitat feeding opportunistically from the lower and mid-water areas of the waterbodies. The main items are algae, microcrustaceans and aquatic insects. The identifiable algae were green filamentous and blue-green algae and dinoflagellates. The microcrustaceans were mainly cladocerans, ostracods and copepods. Chironomid larvae were the main aquatic insects eaten. Other food items found in the stomachs were terrestrial insects and miscellaneous organic matter. One study found that the diet in the late-dry season was mainly based on detritus (with associated unidentified organic material) and small quantities of chironomid larvae and pupae, and algae; no micro-crustaceans were eaten. In the early-wet microcrustaceans appeared in the diet and detritus decreased in importance; aquatic insects also appeared in the diet during this season. In the mid-wet season P. tenellus ate mainly microcrustaceans (particularly cladocerans) with smaller amounts of terrestrial and aquatic insects. By the late-wet-earlydry season algae were the main component of the diet.

An ideal diet for *P. tenellus* in captivity could include foods such as live and frozen brine shrimp, daphnia, mosquito larvae, bloodworms and microworms. Spirulina-based foods flake, and bite-sized pellets designed for ornamental fishes are also acceptable. Obviously, no single food will meet their needs at all life stages, and the best way to ensure that they are getting a well-balanced diet is to feed them as wide a variety of food as possible.



#### Breeding

Very little is known about the breeding biology of this species in their natural environment. Most information is mainly based on aquarium observations. In their natural environment, *P. tenellus* are most likely aseasonal spawners, breeding continuously at intervals throughout the year. However, a peak in reproductive activity usually occurs during the early-wet season from October to December. The spawning season will vary from region to region, but will usually coincide with the conditions that offer the greatest amount of protection for the eggs, and food and shelter for the newly hatched young. Spawning in captivity usually begins when water temperature are maintained above 24°C.

*P. tenellus* are an egg-scatterer, generally spawning amongst aquatic plants and grasses. Ovaries examined in one study contained 33–45 eggs with a mean diameter of 1.0 mm. However, the number of eggs shed by a single female is directly related to the size of the female. The total number of eggs released will increase with the maturity and size of the fish.

Breeding this species under suitable aquarium conditions is generally uncomplicated. Group spawning with multiple males and females is probably the preferred method for breeding. This will allow the females to choose their own mate and in doing so; a variety of genetic factors will be passed on to the next generation. Spawning pairs is possible but group spawning should provide you with more eggs.

Males commonly remained near a suitable site for spawning and exhibit territorial behaviour towards other males such as lateral fin-flaring displays and pursuits. The males swim parallel to each other in a circular pattern; frequently changing direction. This display is a ritualised test of strength between the males and usually comes to an end when one of the males retreats. The males do not actively care for the eggs other than through defence of the spawning site. They may eat their own eggs, or fail to defend the spawning site against other egg predators. They may also cannibalise their young and whilst larvae may survive in a heavily planted aquarium it is a better to transfer the mops or spawning medium to a hatching container or aquarium, at least until you have established a sizeable population. Spawning mops are my preferred spawning method as better survival rates can be expected if the eggs are removed. Several spawning mops should be provided to offer the males a choice of spawning sites and females a choice of hiding places. The mops can be attached to a block of styrene foam and floated in the water or alternatively, just drop the bundle of loose thread into the aquarium.

In captivity, males displayed during the daylight hours with the peak of activity occurring in the late morning and early afternoon. Pre-spawning behaviour begins in an open area of water adjacent to a spawning mop or floating plants. The male approaches the female from the side as she either attempts to flee or accepts the male as a suitable spawning partner. Nonreceptive females will move away and swim to the surface remaining motionless to avoid detection, often in the corners of the aquarium; begin schooling with the other fish or hide amongst the aquatic plants or spawning mops. Direct spawning activity involves the male raising his dorsal and anal fins while actively pursuing the female. Once the female accepts the male she will move into the spawning medium. The male follows beside and parallel or slightly behind the female. During the egg release the females' body quivers as she releases 1 to 30 eggs. The male will remain beside and parallel to the female, his fins erect as he fertilises the eggs. During this procedure the colour intensifies in both sexes. Males will often spawn with other females more than once a day; however females appeared to only spawn once a day. Spawning may continue over several days followed by a period of 1-2 weeks inactivity. A water change or slight change in temperature will sometimes induce spawning after a period of inactivity.

Eggs have adhesive filaments that attach to aquatic plants or amongst the strands of the spawning mops, and sometimes even in the substrate. Spawned eggs are relatively large; negatively buoyant in freshwater and average  $1.2 \pm 0.5$  mm in diameter, and are usually clear to light amber in colour.

Eggs are best left attached to the spawning mops to minimise handling stress and removed from the spawning tank and placed in another aquarium for incubation and larval rearing. Maintain a constant temperature  $\pm$  1°C and gently aeration. Hatching will begin after an incubation period of around four to six days at a temperature range of 25–30°C. Temperature is one of the major factors that influences the embryonic period of blue-eyes.

Although *P. tenellus* are only small their newly hatched larvae are rather large at around 4 mm. Upon hatching the larvae swim at the surface of the water, generally within the upper 1-cm water layer. The mouth is well developed and functional, and they begin feeding within hours of hatching. They can be fed finely powdered dry foods, newly-hatched brine shrimp, copepods, phytoplankton and microworm. *P. tenellus* grow fairly rapidly and reach maturity in about three months; at a size of around 18–24 mm. They can attain a length of 20–30 mm in around 4–5 months, and by the end of 12 months they should be around 40–50 mm. It has been reported that *P. tenellus* can attain a total length of around 19–28 mm in 144 days in captivity with a water temperature of 26°C. At a size of 18 mm both sexes developed breeding colouration. The smallest male spawned successfully when 20 mm; the smallest female spawned when 18.5 mm.





#### **Rhadinocentrus ornatus** Regan, 1914 Ornate Rainbowfish

#### **Species Summary**

*Rhadinocentrus ornatus* is a sub-tropical species that were first collected from Moreton Island, a sand dune island off the south Queensland coast in Moreton Bay. The type description of *R. ornatus* was from six specimens collected from a pond on Moreton Island by the signal station operator at Cowan Cowan. They were described by Charles Tate Regan at the British Museum of Natural History in 1914. They are the only species currently recognised within the genus. In earlier times they were commonly known as the 'Moreton Island Sunfish' and were very popular with early native fish enthusiasts.

*R. ornatus* is a small, slender and relatively elongated species, with two dorsal fins that are very close together; the first much smaller than the second. They exhibit considerable colour variation over their geographical range. Generally the body is semi-transparent with two rows of black scales in the midlateral region. The iridescent scales immediately below the dorsal fin and above the lateral line can be either red or metallic pale blue. The dorsal, anal and caudal fins are generally blue (sometimes red) with black edges. In 1995, ANGFA (Qld) members reported collecting an unusual golden yellow coloured form of *R. ornatus* in the Key Hole Lakes system on

Stradbroke Island. Another coloured form on Stradbroke Island showed a distinctive black striped pattern on the sides of the body giving the fish an overall dark colouration of the body. The actual location was not documented.

Body colour can be variable throughout their range with different colours, shades, or patterns. Most populations have their scales outlined with black that form an attractive latticework pattern over the body. However, some populations only show the scale pattern on the dorsal half of the body while the lower half can be either scattered with rounded or oblong markings, or completely clear. The body can be blue overall or a translucent light brown or olive colour with just a hint of either blue or red. Some males are completely red on the posterior third of the body. The dorsal fin can have a red outer border with black rays or black outer border with blue rays. The caudal fin is usually blue or reddish with a central dark sector. The anal fin usually has a black margin. The blue form is usually light brown on top of the body and fades ventrally to various shades of blue that increase towards the tail. Fish from dark tannin-stained waters can often be very dark. Generally, the difference between the blue and red forms is just that the blue colour is replaced by red. All have scattered iridescent neon-blue spangles of varying densities that run along the back and nape. The different coloured forms can often be found in the same habitat. A third form does not have the black lateral lines or scale pattern and only has iridescent blue scales along





the nape. This form is usually smaller than the other varieties. Females are not quite so highly coloured. Males, apart from their brighter colours, can be distinguished from females by having more elongated rays in the second dorsal and anal fins. Males also display a red nuptial stripe running from the upper lip and along the nape to the second dorsal fin during spawning activities.

A phylogenetic study (Page *et al.* 2004) found that *R. ornatus* exists as four separate populations. These populations correspond to:

- 1. Byfield (Water Park Creek) south to Tin Can Bay and Fraser Island.
- 2. Searys Creek (Rainbow Beach) population.
- 3. Noosa River south to Brunswick River in NSW, including Bribie, Moreton and Stradbroke Islands.
- 4. Northern New South Wales south of the Brunswick River.

*R. ornatus* can grow to a maximum size of around 7-8 cm total length, but are usually more common at around 5-6 cm. In captivity they have been reported to grow somewhat larger at around 10 cm. However, that is more the exception rather than the rule. They tend to develop humped backs and faded colours as they get older and larger.

#### **Distribution & Habitat**

A 1924 survey of Moreton Island, noted that *R. ornatus* were found in swamps and unnamed streams south of the lighthouse. The first report of *R. ornatus* being found on the mainland that I can find was in an Aquarium & Terrarium Society of Queensland excursion report in 1927. At that time they were known only from Moreton Island. They were found in Belmont (Bulimba Creek), an outer southside suburb of Brisbane. Later that same year they were reportedly found in the Pimpama River catchment (Pimpama Island) on the Gold Coast in southeast Queensland. Members of the Aquarium & Terrarium Society were also the first to discover *R. ornatus* on Stradbroke Island. They were found in "a little freshwater creek a few miles north of Myora". A description of the creek from the secretary, Amandus Rudel, in 1930 follows:

"The water in this particular creek is very soft and of a slightly brownish colour. Except for a slimy sort of algae, the creek has no under-water plants of any description. No snails or other small insects were to be found and how *Rhadinocentrus* gets a living is rather a puzzle. The creek is rather narrow but very deep in places and winds its way through a very flat piece of country on the Island. It's really more of a bog and one never knows when the ground gives way. Ferns and sphagnum moss grows all over the flat. Tracks of Kangaroos and Wallabies lead to the creek from all directions, and take it all around it's a very interesting piece of nature."

A description of the fish was given as follows: "Back dark red with a row of blue shining scales. Body yellowish with two rows of black scales and black spots distributed all over body. Dorsal fin red with black edges and the same colour on the anal fin. Tail whitish edged black." Today we know that *R. ornatus* have patchy distribution along the narrow coastal drainages of southern Queensland and northern New South Wales. Its range is nearly continuous amongst the unconnected coastal river catchments from the Nambucca River in the south to the Mary River in the north. North of the Mary River there is a break in the distribution of over 350 km to a disjunct northern population in the Byfield region (Water Park Creek) near Yeppoon. Their distribution includes lake and stream habitats on Bribie, Fraser, Moreton and Stradbroke Islands, a group of islands off the southern Queensland coast. Many of these lakes have never been connected to the ocean. The fish that are present have presumably been introduced as eggs on the feet of birds.

Although not endangered, *R. ornatus* has a restricted distribution. Their distribution has likely contracted as a consequence of urban and rural development and the attendant altered hydrology and water quality. Habitat alteration and urban development are still having negative impacts in several areas. Extensive sampling in rivers and streams of the southeast Queensland mainland over the last few years has yielded relatively few individuals. *R. ornatus* was a rare find during a survey of the Nerang River in 2007. Individuals were found both above and below the Heinz Dam. Nevertheless, they can still be found in reasonably numbers in the Sunshine Coast region north of Brisbane.

The introduced mosquitofish (*Gambusia holbrooki*), which are tolerant of a wide range of environmental conditions may displace native species such as *R. ornatus. Gambusia holbrooki* have been reported to prey on their eggs and larvae. *R. ornatus* have also been observed to switch from areas of open water to refuge areas in the presence of Gambusia. Furthermore, experiments under captive conditions have shown Gambusia to exhibit aggressive behaviour towards *R. ornatus*.

R. ornatus congregate in small schools and occupy small, slowly moving creeks and marshy swamps, usually over a sandy substrate. The water temperature range has been reported from 12-32°C. However, they are known to survive water temperatures as low as 8°C. They are most abundant in unpolluted streams, lakes or swampy areas amongst emergent vegetation and reeds of the 'wallum', which is low, sandy, coastal heathland of southeast Queensland and northern New South Wales. These waterbodies are usually very soft and acidic (pH: 3.9 to 6.8), either clear or more often stained brown or black by prolonged contact with high accumulations of leaf litter and other terrestrial material. The dominance of humic acids among this organic material and the relatively low pH are not conducive to bacterial degradation, so particulate and dissolved humic compounds are metabolised very slowly. The dark colour of the water severely limits penetration of light, which, together with low concentrations of inorganic ions, restricts photosynthetic activity in aquatic plants. Limited photosynthesis and slow bacterial degradation results in low zooplankton and phytoplankton development.

*R. ornatus* have also been found in clear rainforest streams with dense overhanging vegetation and leaf litter. In these streams they are usually found around marginal vegetation, submerged logs, or branches. They may be found on their







own or with other small-bodied native fishes. Many habitats occupied by *Rhadinocentrus ornatus* also contained individuals of *Nannoperca oxleyana* and *Pseudomugil mellis*.

A dietary analyses of their natural foods showed that the main components of their diet are terrestrial insects (29-34%), Aquatic dipterans (16-24%), Copepods (8-11%), Cladocerans (0-6%), Ostracods (5-9%), Decapods (0-2%), algae or pollen, mostly pollen (6-32%), seeds (0-1%), aquatic acarinids (0-1%), and miscellaneous organic matter (0-7%).

#### **Keeping & Caring**

Very little is known about the natural life history and ecology of *R. ornatus* in the wild. Most information is mainly based on aquarium observations. In their natural environment spawning usually occurs during the warmer period of the year (Spring-Summer) when water temperatures are around 24–32°C. Eggs and larvae have however, been found in the upper Orara River in northern New South Wales during Spring at temperatures of 16–17°C. Eggs adhere to fine-leaved plants or among the roots of floating vegetation.

*R. ornatus* have been a popular aquarium fish with Australian native fish enthusiasts for many decades. They are a fish that requires a little more attention than most other rainbowfishes. They can be maintained and bred in water conditions that are

suitable for most aquarium species. However, they will survive best if maintained at a temperature range of 20–28°C; pH 6.0– 7.0; and water hardness from 5–100 ppm. They will display their best colouration when maintained in a well-planted aquarium of their own, and in a group of around 10 to 20 individuals, particularly if maintained under aquarium conditions that will show their iridescent reflective colouration.

Breeding *R. ornatus* is similar to most rainbowfishes in captivity. Females spawn small numbers (3-5) of eggs each day. The total number may range from around 20 to 80 eggs over several days. Hatching occurs in about 6–10 days at temperatures between 23–28° Celsius. Hatched larvae are well developed and competent swimmers. Upon hatching the larvae swim at the surface of the water, generally within the upper 1-cm water layer. The mouth is well developed and functional, and they begin feeding within hours of becoming free-swimming. They can be fed commercially available fry foods, newly-hatched brine shrimp, copepods, phytoplankton and microworm.

*R. ornatus* characteristically display a considerable range of growth rates, depending on conditions such as food, space, numbers, competition and water temperature. Growth rates of 20 mm in 3 weeks and 30–40 mm in 10 weeks have been reported in captivity with maturity at about 9–12 months. Life expectancy in captivity is around 3–4 years.







# Scaturiginichthys vermeilipinnis

Ivantsoff, Unmack, Saeed and Crowley, 1991 Redfin Blue Eye

#### **Species Summary**

*Scaturiginichthys vermeilipinnis* were originally collected from a number of artesian springs located on Edgbaston Station, a sheep and cattle property located 35 km north-east of Aramac in central-western Queensland in 1990, and were scientifically described in 1991. The scientific name is a reference to the unique habitat (scaturginis is Latin for spring; ichthys, pertaining to a fish) and the red colouration on the margins of the dorsal and anal fins (vermeil - old French red or vermilion; pinnis, Latin for fins).

*S. vermeilipinnis* is Australia's smallest freshwater fish reaching a maximum total length of around 28 mm. They have a translucent silvery to golden body that becomes darker dorsally and around the head region with a plainly visible swim bladder. Iridescent scales are visible above the anterior midlateral line. Opercles iridescent; eyes silvery-blue with a dark vertical stripe through the orbit. The males' unpaired and pelvic fins are edged with red, hence their common name of Redfin Blue Eye. Fins are clear to faintly yellowish in juveniles and females. Juveniles have the posterior half of the body

golden-yellow. Males are generally larger than females with larger fins. External morphology separating *S. vermeilipinnis* from other blue-eyes include a narrow rounded caudal fin, lower position of the pectoral fin and frequent absence of ventral fins which may be an adaptation to a very shallow habitat.

*S. vermeilipinnis* is Australia's most endangered freshwater fish and share their habitat with another endangered species *Chlamydogobius squamigenus*, the Edgbaston Goby.

Although population numbers in individual springs have varied since their discovery, specific population trends are not well known and they have disappeared completely from a number of springs. It is estimated that their numbers may range from a few hundred to a few thousand individuals. Their continued existence is being threatened by the introduced mosquitofish Gambusia holbrooki and habitat destruction caused by harvesting water from the Great Artesian Basin; trampling and grazing by stock and feral animals, and modification of springs to provide for stock watering. The Great Artesian Basin Bore Rehabilitation Program may have some long-term benefits in terms of increased water flow to the springs. Bores are been capped and drainage canals are being replaced with pipes to reverse declining groundwater pressures and water levels; this has led to the restoration of some spring wetlands.



The fragile nature of the springs at Edgbaston implies that extinction of these truly unique features of the arid Australia landscape and the associated aquatic fauna is very real. A recent survey has found that sub-populations of this species are not healthy and potential threats are increasing.

*S. vermeilipinnis* was originally recorded as naturally occurring in eight separate springs. Since its discovery in 1990, five populations have been lost and subsequent colonisation has occurred in two springs. In 1994 five naturally occurring populations were known, plus a translocated population (from one of the above springs). A 'Species Recovery Plan' was prepared for the Australian Nature Conservation Agency (now Environment Australia) in 1995, although it was generally not implemented. When the springs were visited in 1998 the five springs still contained existing populations. At the most recent survey in 2005, redfins were present in five relatively small shallow springs. They appear to have become extinct from three of the larger deeper springs. The cause of their demise has not been clearly established.

#### **Distribution & Habitat**

S. vermeilipinnis are endemic to the Edgbaston Springs complex near Aramac, which is located 67 km north of Barcaldine (about 930 km west-northwest of Brisbane). Aramac is one of those tiny little settlements in western Queensland which has outlived its original purpose and now stands forlornly in the middle of nowhere supporting the surrounding pastoral properties and sustaining the few people (approx. 300) who continue to live in this inhospitable, hot and dry environment. The area was first explored by Europeans and settled in the 1850s. The town was named after Robert Ramsay Mackenzie who, at the time, held land leases totalling 1536 square miles. Mackenzie, really nothing more than a land speculator, was Queensland's first treasurer and future premier. He was of limited talent and left no great impression on the public life of the newly formed colony. William Landsborough explored the area in 1859 and called a nearby watercourse Aramac Creek. In a letter he explained "The Aramac, as many wrong reasons for the name have been given, I may say here I named, in honour of the late Sir R. R. Mackenzie, 'Ar-Ar-Mac', who was so well known in Queensland, and who had acted in a very friendly way to me".

Edgbaston Springs are located in the upper reaches of Pelican Creek within the Thomson River system in the Lake Eyre drainage about 31 km north-east of Aramac. Pelican Creek is ephemeral although some waterholes may persist between rainfall events. Pelican Creek (35 km) merges with Aramac Creek. Aramac Creek flows through Boundary Waterhole and Middle Waterhole on its way to joining the Thomson River. The following creeks flow into the Aramac Creek: Curlew Creek, New Year Creek, Emu Hills Creek, Gum Creek, Sandy Creek, Pelican Creek, Politic Creek, Ibis Creek, Corinda Creek, Middle Creek, Willoughby Creek, Four Mile Creek, Langharne Creek, Mountain Creek, Tommlins Creek, Tuaburra Creek, Rodney Creek, Scarrbury Creek, Neil Creek and Gambling Creek. Edgbaston Springs are a complex of artesian springs scattered across an alluvial plain and supporting an unusual habitat type, which is distinct from the surrounding arid region. The actually wetland area is small; the total surface area of known habitat varies seasonally between approximately 6 to 8 km<sup>2</sup>. At least 44 springs have been identified at Edgbaston Springs, but only about 30 have permanent water, some of which have become extinct. They are derived from faults allowing water to flow from thin confining beds of the Great Artesian Basin aquifer. They are permanent artesian springs, with some evaporation and associated reduction in extent during the summer months. Most of the springs are very small, shallow, and marshy. Some springs lower in the catchment are occasionally connected by floodwater.

Water depth varies throughout the springs with depths between 3 and 7 cm. Some springs have associated pools which are usually less than 20 cm deep but may be up to 50 cm. Temperatures recorded in the region show an average high of 29–30°C, average low of 14–16°C. Temperature extremes have been recorded of -3°C and 51°C. Annual rainfall average is 117–161 mm, with a recorded high extreme of 543 mm and a low of 30 mm. Water chemistry of the springs have been reported as follows: Conductivity 560–3270 mS/cm; TDS 478–2597 mg/L; pH 7.1–9.1; Alkalinity: 235–1380 mg/L.

The water of the springs that contain *S. vermeilipinnis* is generally clear with a pH 7.8 to 8.0 (average 7.93) and high alkalinity. Water temperatures are extremely variable from spring to spring and within each spring. In May 1990 the temperatures varied from 7 to 28° Celsius. During May 1991 at 7.00 am water temperatures of 7 to 20°C were recorded in different parts of the springs. While at other times of the year minimum and maximum water temperatures of 3 and 38.5°C have been recorded. *S. vermeilipinnis* are usually located in areas of the springs where the temperature is above 16°C.

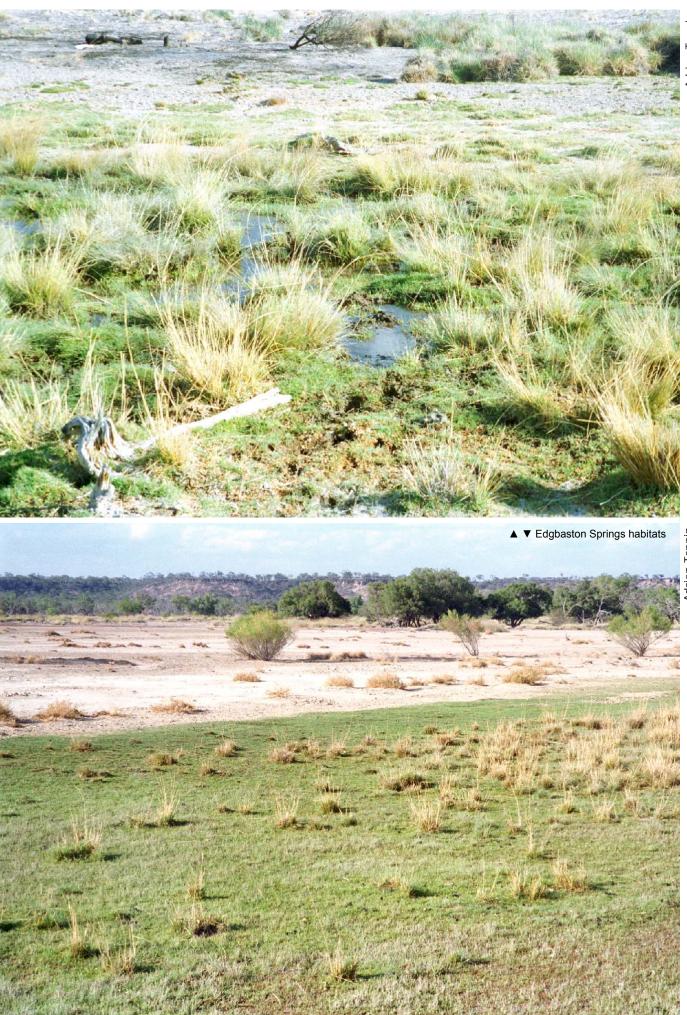
*S. vermeilipinnis* may be found throughout all areas of a spring. Adults generally occur in the deeper areas of the springs, while newly hatched fry and juveniles are usually found in the shallower areas. When approached they form large schools, which is probably a predator avoidance response. If undisturbed the fish disperse into smaller groups, and begin feeding and displaying.

Edgbaston Springs contains a diverse endemic fauna (fishes, invertebrates) which rivals that of Dalhousie Springs. Increasing evidence of extraordinary endemism in some groups (especially molluscs and crustaceans) shows that many taxa are confined to single springs or groups of springs. Plant communities include *Eryngium fontanum*, *Sporobolus pamelae*, *Fimbristylis dichotoma*, *Utricularia*, and *Eriocaulon carsonii*. The spring's distinctive fauna suggests they have been isolated for a very long time.

*Scaturiginichthys vermeilipinnis* may have had a much wider distribution in other artesian springs throughout the Lake Eyre region at one time.







#### Biology

Not a lot is known about the biology of S. vermeilipinnis in their natural habitat. Most information has mainly been based on aquarium observations. They probably possess both eurythermal and euryhaline characteristics, which are acquired by many desert fishes in response to a changing environment. Males defend variable territories against other males, usually around emergent vegetation and will display to any female that enters their territory. During spawning the colouration of the male's fins intensifies to a bright red colour, the overall body colouration becomes much darker and the fish develops a dark stripe that runs vertically through the eye. The female undergoes only minor changes with her body becoming slightly darker. Eggs are released either onto the globular algae on the substrate or vegetation. Territorial males, eggs and juveniles have been found throughout the year but are more common in warmer months.

The natural diet of *Scaturiginichthys vermeilipinnis* is unknown. Individuals have been observed taking a mouthful of substrate, expelling matter from the mouth and then picking particles from the expelled cloud. They have also been observed picking particles directly from the substrate, from the surface of submerged vegetation and from the water column.

#### **Keeping & Caring**

I first obtained this species in April 1994, and although I encountered some difficulties, I managed to maintain a captive population of around 40–60 individuals. However, as I progressed through the numerous generations, the overall population was declining. The major problems encountered were reduced egg numbers and a large percentage of 'soft' eggs, resulting in a rather small number of young fish. Under normal aquarium conditions water-hardened eggs are firm and can be rolled between the fingers without any problems. The "soft" eggs however, would burst under slight pressure.

Forty years of experience as an aquarist and breeder has taught me that "natural" conditions are not necessarily the most favourable for maintaining captive specimens. *S. vermeilipinnis* have probably experienced extreme changes of both water temperatures and chemistry during their evolution. This adaptability to different water conditions is presumably why they have survived in this uncharitable environment. Therefore, I experimented with various water conditions to see what conditions were most suitable for *S. vermeilipinnis*, and managed to breed and raised them under the following water chemistry ranges: Temperature 20–31° C, *p*H 6.8–9.0, Hardness 100–160 ppm, Alkalinity 20–200 ppm, and 415–975  $\mu$ S Conductivity.

I was not happy with the results however, and continued to investigate other possible causes. I thought perhaps that inbreeding or something similar was causing the problem. Fortunately, I managed to obtain additional wild-caught specimens in October 1997. These were added to the population, but still no improvement was forthcoming. During the period I maintained this species I had for the most part provided them with bottom substrate mops. I had thought that coming from such shallow water in their natural habitat they would be bottom spawners particularly when I was maintaining them in water around 30–35 cm deep. This belief was further enforced by the numerous observations of males displaying to each other and driving the females into depressions in the gravel or algae mat that covered much of the gravel substrate. More than one male would be involved in this behaviour and sometimes as many as three were observed with one female. Although no eggs were actually spawning.

I used reasonably large bulky substrate mops and on a number of occasions had observed a couple of dead specimens among the strands of the mop. After testing the water quality, I really didn't give it much thought, and had convince myself that it must just be old age, as this species doesn't have a very long life span. However, something must have clicked in my mind and I thought perhaps that they were being entangled in the mops and subsequently died.

I decided to change the bottom substrate mops for smaller floating ones. The results were a surprise – not only did egg numbers increased, but also there was a major reduction in soft eggs. Although, this may have related to the existing water chemistry, which was as follows: temperature  $25-30^{\circ}$  C, *p*H 8.5, hardness 160 mg/L and alkalinity 150 mg/L. Almost all the eggs collected from the floating mops were laid in the upper extremities of the mop, just below the water surface, and often in small clusters of 2–4 eggs. In addition to the above results, I did not find any dead specimens in any of the floating mops.

I had also suspected that soft egg production may have been caused by a lack of environmental calcium. However, upon testing the calcium level it was in the range of 25–50 ppm, which is consistent with Brisbane tap water. I then decided to try an increased alkalinity level and used a product called "Aquasonic KH Generator" to increase the alkalinity to around 250 ppm. I found that using the kH generator greatly increased the number of eggs and the general health of the fish.

Over the six-year period that I maintained and bred *S. vermeilipinnis* they were mostly kept in small (130-L) aquariums, although later they were transferred to a 175-L aquarium. The fish were fed twice daily at 07.00 and 16.00 hours. Their diet consisted mainly of newly-hatched brineshrimp, microworm, frozen bloodworms, homemade fish food and a commercial fine powdered larvae diet. They were also fed the occasional sprinkle of powdered spirulina.

The spawning mops were checked twice a day, at 07.00 hours and 16.00 hours. Any eggs were picked from the mops by hand and placed into a hatching container. Eggs collected were visibly different in size; probably due to the size difference in the females (Fry were also clearly different in hatching size).



During a period of 21 days, egg collection and hatching rates were recorded. Eggs were hand-picked from the mops morning and night (07.00 and 16.00 hours). The collected eggs were placed in a 4-L plastic hatching container with 2-L water and 12 drops of methylene blue as a fungicide. The container was then floated in a 135-L raising tub. As the eggs hatched, the fry were carefully transferred to the raising tub. Water in the raising tub was maintained at a temperature of 30°C, *p*H 7.6 to 7.8, TDS 275 mg/L, hardness 120 mg/L and alkalinity 50 mg/L.

The total number of eggs collected during that period was 178, with the maximum number of eggs collected in one day being 23. Fry survival from the 178 eggs was 89. The survival rate was a lot lower than I would have anticipated, but it was evident during the egg collection process that some eggs had not developed. Some eggs burst during collection, giving me the impression that they had not water-hardened. This was before using the kH Generator. Later spawnings gave a much improved hatching rate than reported above.

In captivity, *S. vermeilipinnis* usually spawn when the fish are around 12–15 mm in length. Spawning has been observed at water temperatures between  $20-32^{\circ}$ C. The spherical, opaque eggs are about 1.0–1.5 mm in size and have filaments that attach to vegetation or the substrate. The eggs take between 10–14 days to hatch, depending on water temperature (at 28°C, eggs hatch in eight to ten days). The larvae hatch at between 4–5 mm in length and begin feeding within 24 hours.

Fry grow rather quickly and may reach 15 mm in six to ten weeks. The colour of the fry is unusual, with the front third of the body blue and the rest golden-brown, but this changes to adult colours as they grow. They normally swim near the surface of the water until about 10–12 mm in length when they are found at lower levels in the aquarium.

Spawning activity is similar to other blue-eyes with males displaying to passing females. Males develop a smoky golden-brown body and more intense red colouration in the fins. Sometimes the males can be quite aggressive and will actively pursue the female. The spawning display involves the male swimming around the female with outspread fins. If the female is receptive the pair will come together and swim side by side. Eggs are then released amongst the spawning mops or over the substrate.

From my experience it was obvious that *S. vermeilipinnis* spawn either late evening or at daybreak (or both). Also, most of the eggs hatched during daylight hours. This is different from most other blue-eyes that I have bred as they usually hatch during the night. I believe the night-time hatching probably affords a certain amount of protection for newly hatched larvae. Due to their isolation, perhaps such protection is not required by *S. vermeilipinnis* as they do not appear to have any natural predators.

#### Remarks

A draft 'Recovery Plan' for *S. vermeilipinnis* was being prepared back in 2005-06 for the Department of the Environment and Heritage, Canberra and the Queensland Parks and Wildlife Service, Brisbane. What happened with that 'plan' is not known to me. However, a paper was published in 2007 (Fairfax *et al.*), but no recovery plans were outlined. It did mention that "*The failure of attempts to maintain captive breeding populations suggests that an intensive and dedicated effort would be required with due consideration given to water quality, population structure, microhabitat and diet*".

There was an earlier suggestion that establishing a population in an artificial wetland at Edgbaston Station may have greater likelihood of success than off-site aquaria. Groundwater is available from a low-volume permanently flowing house bore. Using this supply, an outdoor tank or artificial wetland that would exclude gambusia, feral pigs and other threats could be designed and established to maintain a reserve population of *S. vermeilipinnis*. This population could be used, if necessary, to restock springs and could also provide stock for ex-situ experiments if field studies prove unsuccessful.

The conservation of their natural habitat alone, of course, would be useless if developments in the region seriously depleted groundwater stocks and flows. To this end I would also like to see them become established in the aquarium hobby. However, I doubt if anyone will ever be given the opportunity again? I also don't have much faith in the idea of Government agencies being able to secure their future. Australia does not have a good record in regard to the protection of the nation's biodiversity. Governments and their policies change - when funding runs out - that's the end of the story. That's what happened with the first recovery plan. No more funding - no more work, and everyone involved just went on to do other things. That is also why I put a little extra time and effort into maintaining my captive population. However, it needs a number of dedicated people for total success.

Before aquarium hobbyists can become involved in species maintenance programs we have to get 'runs on the board' and show these Government Agencies that we can manage captive stocks. However, I'm afraid that we failed miserably with *S. vermeilipinnis*. Several aquarists in Australia and internationally had captive populations of *S. vermeilipinnis* at some time. However, most captive populations were lost due mainly to inadequate aquarium keeping practices or inexcusable neglect.

Within Australia at least 32 attempts involving 26 aquarists are known. Of these, the outcome of eight is unknown but suspected not to have lasted beyond F1. Sixteen resulted in no breeding, two bred but not beyond F1, three went to at least F1, two to at least F2 and one went to F4~F6. Only two breeders are known to have had more than 100 individuals at any one time. In 2000, at the time of deciding to retire from fishkeeping, I had approximately 100~150 adults and fry of various sizes. Today, there are no *S. vermeilipinnis* left in captivity anywhere in the world.





I believe that the real secret of keeping them in captivity is that they need your personal attention at all times. You need at least 2 or 3 dedicated breeding aquariums and continuous collection of eggs. You also need at least 4 raising tanks or tubs as the older ones will eat the smaller fry so you need to have raising tanks that can hold fry in various stages of growth. They need live foods as they are not all that fussed on prepared foods. They also require good water conditions with regular water changes. I do believe that the alkalinity level has some effects, but just what, I really don't know?

The tank size I would use now if I ever kept them again would be at least a 120 cm square tank but only about 30 cm deep. In this I would only keep maybe 3–4 males and 6–8 females? I would set up at least 3 of these tanks. In these tanks I would use 4–6 small floating mops that reach down to the gravel. Raising tubs of about 135 litres should be adequate with maybe 4–6 of these.

I believe that the survival of *S. vermeilipinnis* is going to depend not only on those who care enough to preserve their natural environment but also to committed aquarists who are willing and capable of maintaining viable populations in captivity. Collectively, it may be possible to prevent its extinction.

In July 2008, Edgbaston Station was purchased by Bush Heritage Australia. Bush Heritage Australia is a not-for-profit organisation that protects Australia's unique animals and plants and their habitats. It owns and manages thirty-one conservation reserves throughout Australia. The purchase of Edgbaston Station was assisted by a significant contribution from the Australian Government's 'Maintaining Australia's Biodiversity Hotspots' program. Such support was in recognition of how critical these properties are to the protection of threatened species and systems of high conservation value in Australia.

Work has now commenced on restoring the unique terrestrial and aquatic environments present at Edgbaston Station. Of particular concern is the future of the two critically endangered fish species, *S. vermeilipinnis* and *C. squamigenus*. Both species have suffered extensive range reductions since their discovery in 1990, and this is generally thought to be due to invasion of the springs by *Gambusia holbrooki*. Gambusia control using physical and chemical methods has now commenced at Edgbaston, as have relocation events seeking to expand the distributional range of these species. In concert with breeding programs scheduled to begin in early 2010, the on-ground measures at Edgbaston Station aim to preserve the dwindling populations of these unique fish species that are restricted to one of Australia's most isolated spring complexes.



## **Rainbowfishes** Disease Prevention & Control

The information provided herein is intended as a guide to the most common disease problems of rainbowfishes maintained in aquaria or ponds. Basic treatment is also included; however, this information should not be substituted for consultation with an experienced fish health professional.

Veterinary medicine is an ever-changing field. Standard safety precautions must be followed, but as new research and clinical experience broaden our knowledge, changes in treatment and drug therapy may become necessary or appropriate. Readers are advised to check the most current product information provided by the manufacturer of each drug to be administered to verify the recommended dose, the method and duration of administration, and contraindications. It is the responsibility of the reader, relying on their own experience and knowledge, to determine dosages and the best treatment for their aquarium inhabitants.

While all responsible efforts have been taken to ensure the accuracy of information contained in the following section, neither the publisher nor the authors assumes any responsibility or liability for any lose or injury and/or damage which may result from an inaccuracy or omission, or from the use of information contained in this publication.

John Gratzek presented an overview of the diseases of ornamental fishes in 1980. He reported that most (60%) aquarists who discontinue keeping fishes do so because the fishes die. He identified common problems with keeping ornamental fishes and stated that the treatment of diseased fishes is the most common problem. For new hobbyists this can be a major problem compounding the fact that they have often not researched the necessary information to be successful in their new hobby. Initial purchases of fish are often done for aesthetic rather than practical reasons and regularly, newly bought fish will often die resulting in the hobbyist losing interest in ornamental fish altogether. Those that get past this initial stage often find an appetite for becoming better informed and often acquire greater knowledge than some "experts".

The increasing popularity of rainbowfishes has resulted in a significant increase in the number of commercial operators breeding, rearing and distributing rainbowfishes. This increases the potential for dissemination and exacerbation of infectious diseases. The increased interest in keeping rainbowfishes has also increased awareness of and experience with pathogens that affect their health, growth, and survival. Rainbowfishes maintained in aquariums and ponds are entirely dependent on our management of their environment to maintain their health. Health or disease is a complex balance between pathogens, the fish or other organisms and the aquarium environment.

A wide range of pathogens (bacteria, parasites, etc), environmental factors (water quality, etc), and even aquarium keeping methods, can cause disease problems in aquaria. Often these factors are linked in disease outbreaks. Mismanaged aquariums allow opportunistic organisms to cause disease. For example, a decline in water quality associated with poor aquarium keeping practices may lead to an increase in the incidence of bacterial infections. Another important source of pathogens is new fish obtained from other hobbyists and suppliers, both these sources need to be appropriately screened to ensure that introduction of pathogens to your aquarium is minimised.

Despite what you may read elsewhere, reproducing the intricacies of the natural aquatic ecosystem of rainbowfishes in an aquarium is not only a very difficult task, but almost impossible to accomplish with overall success. It is not as simple as having the water at a particular temperature or pH. Water chemistry (hardness, nitrates, alkalinity), suspended particles, levels of oxygen/CO<sub>2</sub>, parasites, prey and predators, symbionts, wet and dry seasons, light (intensity, cycle, spectrum), droughts, floods, water flow, substrate, vegetation, woody debris, sound, space, shelter, etc., etc., are all variables that are either difficult to replicate or are difficult to control in a captive environment. In consequence, something is likely to go wrong-at the very least occasionallymaking the captive aquatic animals become chronically or acutely ill. Opportunistic bacteria and parasites can cause dermal and systemic infections. Intestinal nematodes can cause chronic wasting (anorexia) and considerably damage to the intestinal lining. Additionally, water moulds and fungi may also present disease problems in poorly managed aquariums.

Many diseases can be eradicated or kept to minimal levels by careful management and a good knowledge of the basic biology and natural history of the species (both fish and pathogen) concerned. Therefore, disease prevention and control are a significant part of the rainbowfish hobby. For this reason, it is extremely important that you read and understand a practical book on this subject. Several excellent choices are available and they represent one of the best investments in time and money for the serious rainbowfish keeper.

Diseases causing organisms fall into two categories:

- **Obligate Pathogens**: always cause disease when able to invade the host's bodies. They may however, only cause disease in certain circumstances.
- Facultative Pathogens: don't usually cause disease but can when their population explodes due to environmental conditions or for instance because a fish's immune system is depressed, the water quality is diminished or the fish skin has been damaged. Most fish pathogens fit this description and are very opportunistic rarely causing disease in healthy well-managed aquariums.

Fish disease usually occurs as a result of adverse interactions between the fish and their environment. The natural environment of rainbowfishes is vastly different from that of an aquarium. Most aquarium environments are completely devoid of structure. They are generally featureless, monotonous enclosures with little opportunity for the inhabitants to display any natural behaviour. They bear no resemblance whatsoever to the fish's natural environment and densities can be up to 100 times greater than those in nature. Reductions in density alone will produce healthier fish. When fish that are already over-crowded in the aquarium are further stressed; for example, by inappropriate aquarium conditions, nutritionally inadequate feeds, or the nature of captivity itself, their natural immune system may be weakened and the ability of the fish to protect itself against infectious diseases is reduced.

A variety of parasites and pathogens can and do infect rainbowfishes. Most are naturally present in low numbers and normally do not cause problems. However, disease is rarely a simple association between a pathogen and a host fish. Usually other circumstances must be present for active disease to develop in a population. These circumstances are generally grouped under the umbrella term "stress". There is a wealth of scientific research suggesting that captive fish populations suffer from stress. There are publications with in-depth chapters on disorders associated with general 'stress factors' in fish. Stress is the response of the fish to a 'stressor' (external or internal) and is a normal feature of life, serving important adaptive functions. The stress response consists of a combination of four general biological responses: behavioural, autonomic, neuroendocrine and immunological. The nature of this biological response varies between individuals and is influenced by factors such as previous exposure, genetics, age and physiological state.



The concept of biological stress was first introduced by Hans Selye who described it as the non-specific responses of the body to any demand. The idea for stress arose from the concept of homeostasis: a relatively constant steady state maintained within certain tolerable limits and the concept of a milieu intérieur or internal environment of the body in which cells are nourished and maintained in a state of equilibrium. Since its original introduction, the concept of stress has evolved and our ideas about stress continue to change. More recently stress has been described as the physiological cascade of events that occurs when an organism is attempting to resist death or reestablish homeostatic normality in the face of a perceived threat.

Regardless of the combination of biological responses, the result is an alteration in the fish's normal biological function as it attempts to adapt to or cope with the stressor, behaviourally and/or physiologically. In most cases, this altered biological function has a minimal effect on the fish's wellbeing; the stressor is either brief or it is eliminated, so biological function soon returns to normal. However, if the stress is not alleviated or if the stressor is large enough, the fish is forced into a prepathological state that makes it vulnerable to disease, abnormal behaviour, reduced growth, reproductive failure, immunosuppression or some other type of undesirable shift in biological function. Although a stress response is necessary to enable an organism to adapt and overcome the threat, under conditions of chronic stress, the adaptive value of the stress response is typically lost, as indicated by reduced health and fitness.

Identified stressors in fish are numerous and include sudden and extreme changes in the physical environment (e.g., temperature and water chemistry), animal interactions (e.g., predator avoidance, being kept with the wrong species or individuals, aggressive interaction, parasitism, competition for space, and spawning), aquarium practices (e.g., netting, handling and transport) and water quality (e.g., high ammonia, organic pollutants, etc.). Light (excessive or rapid changes in intensity), noise and other disturbances can also stress fish and should be minimised. Rainbowfishes will often dash frantically about the aquarium injuring themselves, in response to normally harmless stimuli such as turning on the fishtank light in a dark room.

Although temperature can be considered a stressor, it is considered separately as the effects observed differ from other stressors. Temperature regulates their metabolism and is a major environmental factor in determining growth rate, metabolism, and nutritional efficiency. In fact, temperature will influence all biological and chemical processes in an aquarium. The temperature tolerance of fish can be further defined as stenothermic or eurythermic. Stenothermic species are those that thrive in a very narrow temperature window. Eurythermic fish are those that can tolerate a fairly broad temperature range. In addition to its direct effect on animal health and immunocompetence, many infectious diseases occur in very specific temperature ranges. The growth of parasite populations is also at least in part attributable to how quickly they can complete their lifecycle, which is closely linked to water temperature.

Fishkeeping practises directed at limiting stress are likely to be the most effective in preventing disease outbreaks. Simple measures like increasing filter performance, providing dark backgrounds, natural substrate material, aquatic plants, submerged items such as rocks or driftwood, and overhead cover (floating plants) will improve the aquarium environment. If you want your rainbowfishes to survive and grow, then it is imperative that their aquarium is maintained under conditions conducive to good health. Well-nourished rainbowfishes maintained in highly favourable environmental conditions will be resistant to most pathogens.

#### **Diagnosing Disease Problems**

Monitoring your fish and aquarium is the most important part of early identification and management of disease problems. Since the fishes cannot simply tell us that they are feeling sick or uncomfortable we must educate ourselves as to their normal and abnormal behaviour so that we can have a functioning 'early warning system' before the majority of the population becomes affected. In captivity, rainbowfishes frequently exhibit behaviour which may be described as abnormal because they are not known to be a feature of the natural/wild behaviour of the species, or because they appear inappropriate in time or frequency of performance. Under suitable aquarium conditions, healthy rainbowfishes display "normal" behaviour. As aquarium hobbyists, we should become familiar with the normal behaviour of our fish.

Changes in behaviour may vary considerably, from stress due to their housing conditions-or the nature of captivity itself-but some of them are symptoms of specific diseases. These symptoms may include reduced or cessation of feeding, lethargy, floating near the water surface and/or swimming in an oblique position with the head directed toward the surface. Increased or laboured respiration (as indicated by "gasping", flaring of opercula or rapid opercula movement) and breathing at the surface. Staying relatively still in one place in the aquarium, often accompanied by shimmering or shaking. Flashing-every now and then an individual will swim towards the bottom and then suddenly move forward, turning on one side and appearing to rub one flank on the substrate or some other object. Fish will often injure themselves when they do their "flashing" against sharp rocks or other objects, and will often lose scales in the process. Generally this is a sign of ectoparasite infestation. Usually the fish will flash repetitively as opposed to just once. Poor water quality can also be the cause of flashing behaviour.

Physical chances may also become apparent, such as abnormal growths, lesions, loss of some scales and cloudiness of eyes. Fins may become tattered or eroded. The gills may become pale; clogged with mucus and the filaments may appear swollen or fused together. Change of body colour that does not match expected changes due to maturity, courtship or reproduction. This may involve a fish becoming dark in colour or lost of colour, with decreasing intensity over the entire body.



Most of these behaviours are difficult to assess based on a quick glimpse of the individual fish in the aquarium, and it is only the accustomed eye of the experienced aquarist that can detect them. The clinical signs of fish disease that can be directly observed by the eye are generally quite indicative of the disease group. However, fish diseases often have clinical signs that can be easily mistaken for a number of diseases. Visible examination of the skin and gill tissue may be sufficient to detect the presence of common parasitic infections (some fungi, protozoans and metazoans). However, identification of many pathogens, especially bacteria, requires the skill of a specialist using specialised analytical techniques.

If one is certain that a problem exists the next step is to determine if the problem is related to environmental or biological factors. Any time you observe a problem with your fish; always check the temperature, pH, ammonia and other water parameters. Far more rainbowfishes die from inappropriate aquarium conditions than disease. Water quality problems may be alleviated by dilution or changing with fresh water. Quite often, a simple water change will result in the recovery of the fish. However, treatment of infected fish with a therapeutic drug or chemical may ultimately be necessary. If treatment is indicated, it will be most successful if it is implemented early in the course of the disease while the fish are still in good condition.

#### Responding to Disease

Regardless of how diligent one is, if you keep rainbowfishes long enough you will inevitably encounter a disease problem. When a disease problem develops, quick and effective response is essential. Many disease problems of rainbowfishes begin as external infections. If uncontrolled, the infections may become systemic, resulting in death of the fish. Correct use of chemical treatments can effectively control many bacterial, parasitic and fungal agents before systemic infections become established. In general, an experienced fish health professional should carry out treatment for all but the most common disease problems that your fish experience. However, even veterinarians with laboratory diagnostic experience cannot make an accurate diagnosis of some problems without microscopic examination of the fish or cultivation for bacteria.

Without professional assistance, any diagnosis is purely an educated guess based on yours or someone else's experience. This usually means that any medications or procedures used to attempt to cure the disease are chosen on the basis of the type of behaviour and physical appearance of the fish, as opposite to decisions made on the basis of diagnostic tests. In most cases a 'shotgun' approach using some medication or combination of medications will be used. The downside of this idea is that in most situations the majority of the chemicals given to the sick fish are useless in terms of efficacy, since they are not targeted at the desired pathogens.

Experienced aquarium hobbyists can and do make accurate, presumptive diagnosis's based on examination and assessment of the clinical signs, and then apply affirmative control measures. Nevertheless, if abnormal behaviour persists and/or

mortalities occur, then you should seek professional assistance. However, the cost of treatment may exceed the value of the fish in the aquarium or pond. An aquarist with a large-scale breeding set-up stocked with valuable, rare or endangered fish, for example, would probably be wise to spend the money on proper diagnose. On the other hand, if the loss only involved common species, then spending a lot of money for a fish health professional may not be economically sensible. Therefore, economics and other factors will determine the appropriateness of treatment.

Following any disease outbreak, infected fish should be immediately removed from the main aquarium and isolated in a quarantine or treatment tank. Infected fish can shed pathogens, even when they show no signs of diseases, but especially when they are either morbid or dead. Pathogens tend to leave dying and newly dead fish as they are no longer any use as hosts! However, it is acknowledged that this is not always possible. If treatment is undertaken in the main aquarium, a large water change (50–75%) together with thorough gravel cleaning in order to remove any waste (faeces, uneaten food, detritus, etc.) from the substrate should be undertaken. This includes the removal of algae from tank walls and particulate matter from the filter (change filter wool or wash sponge etc.).

Most aquarium medications are affected by variations in water chemistry such as pH, carbonates, chlorides and dissolved or particulate matter. In water that is high in dissolved organic wastes the chemicals will oxidise the wastes rather than attacking the disease causing agents. This has the effect of lowering the effectiveness of the chemical dosage. Failing to ameliorate water conditions while treating sick fish with medication will usually either prevent the medication from being effective or will cause the disease to recur after the treatment is completed. Most chemicals used to treat fish diseases can be toxic to nitrifying bacteria at therapeutic levels for fish. Antibiotics are generally toxic and have a severe detrimental effect on nitrifying bacteria. Therefore, it is advisable to take biofilters off-line during treatment and to do a large (75%) waterchange prior to re-establishing the filtration system.

#### **Chemical Treatment**

It is important to keep in mind that all fish medications are toxic to fish and treatment should not be undertaken without a thorough understanding of the potential problems that can occur. Fortunately, it usually takes a higher concentration of the drug to harm the fish than it does to harm the pathogens. Nevertheless, subtoxic doses for the fish are still stressing, and repeated doses can build up to toxic levels. The toxicity as well as successful use of chemicals frequently depends on dosage, abiotic parameters (e.g., temperature, hardness, organic load or pH, the species, and the developmental stages of fish.

When using any treatment for the first time, a bioassay (a test to determine safe concentration) should be conducted on a few fish before large numbers of fish are exposed. Over treatment with chemicals can cause serious damage to fish. Fish species can react differently to various concentrations of the chemical;



therefore, fish undergoing treatment must be monitored closely for adverse reactions. If the fish react negatively to treatment, the chemical should be flushed immediately from the system, or the fish should be moved to fresh water. However, fish that do not improve as expected should be rechecked and retreated if necessary.

Before applying chemicals double-check the chemical you are using and the concentration to be delivered. Proper calculation of concentration is contingent upon accurate determination of volume of water to be treated. If you are uncertain as to the volume of water to be treated then take the time to measure the size of the aquarium so that volume can be accurately determined.

A popular means of medicating fish is to place a chemical into the water with the fish. However, the addition of chemicals to water containing fish must be done carefully so that the entire chemical is mixed uniformly throughout the water column. If the chemical is not thoroughly mixed, areas of high concentration of chemical may be formed which can damage or kill the fish. To ensure uniform application, chemicals can be pre-dissolved in water prior to application. Aeration can be used to help distribute chemical throughout the water column. If treating in an aquarium with undergravel filtration remove the airstones or powerheads from the uplift stacks and place them in the aquarium proper to prevent the medication from passing through the gravel. Remove any activated carbon from filters but maintain flow for maximum water movement.

Any time a water treatment is utilised, attention must be paid to the concentration of the chemical applied and the duration of exposure to that chemical. There are three basic water treatments: dips (less then 1 minute), short-term (about 1 hour), and prolonged treatment (indefinite). The difference between these treatments is the concentration of the chemical applied and the period the fish are in contact with the chemical. If too little chemical is added the treatment will be ineffective; if too much is added or if the fish are left in contact with the chemical too long, they may become stressed or die.

Antibiotics are drugs that are usually taken internally to control bacterial infections. Therefore, medicated feed or injection, are preferred for treating systemic (internal) bacterial infections. Dose rates are based on fish weight and are expressed as weight of chemical per weight of fish per day for a specified number of days. Improper doses may result in ineffective treatment or mortalities. Water treatment with antibiotics should only be considered when treating primarily external bacterial infections of the skin and gills of fish. However, the effectiveness of antibiotic therapy for aquarium fishes has been inconsistent and, as a consequence, mortalities continue to occur.

Antibiotic efficiency has been declining for various reasons, not least the development of bacterial resistance. Antibiotic susceptibility testing on fish isolates is rarely performed, and resistance appears to be highly dependent on the infecting species and strain. Resistance to commonly used antibiotics is an emerging problem in the ornamental fish industry. When faced with a tank of sick or dying fish, usually the person is more concerned with how to treat the problem rather than with resolving what is the cause of the problem. A common mistake of many hobbyists is misdiagnosing disease problems and treating their sick fish with the wrong medication or chemical. When the chemical doesn't work, they will try another, then another. Selecting the wrong treatment because of misdiagnosis may be more detrimental to the fish than no treatment at all. Successful aquarists learn by experience.

#### **Obtaining Medications**

As anyone who has visited an aquarium store knows, there is no shortage of commercial medications available for treating fish disease. However, medications sold and used in the aquarium hobby vary in quality and effectiveness. In fact, some fish medications simply do not work. In 1974, Trust and Chipman tested eight products marketed for the treatment of bacterial diseases in aquarium fishes. The products contained erythromycin, neomycin, nitrofuran, penicillin, sodium sulfathiazole, sodium sulfamerazine, sodium sulfamethazine, streptomycin or tetracycline. When used at the concentration recommended by the manufacturer, the products failed to inhibit the growth of bacterial species known to be potential pathogens of aquarium fishes. Furthermore, one of the more effective antibacterial formulations was toxic to the fish. The results also showed that markedly higher levels of the formulations also failed to significantly decrease the numbers of viable bacteria in the aquarium water.

Medications for the aquarium can be found as dry ingredients (crystals or powders), or in liquid form (solutions). Sometimes, the chemical comes in pre-packaged amounts. One should follow the manufacturer's instructions for treatment, as different manufacturers use different chemicals and concentrations of the active ingredients. It is advisable to use only those fish medications that list the active ingredients on the label so that you know what you are using.

Many popular fish medications sold in pet stores will simply not work when used as directed. The delivered dose of active chemical is often below that recommended in the scientific literature - many don't even state the active ingredients. However, it is NOT a safe practice simply to increase the dose rate, and in fact, such measures can have disastrous results. In addition to this, many aquarium chemicals can cause problems with established biological filtration. It is worth noting that control of disease in aquariums may further be complicated by the presence of filtration (biological and chemical), sand, gravel, plants, and organic matter, since these will reduce the efficiency of the chemical compound by direct inactivation. Therefore, you need to be well informed on the subject of aquarium fish medications and should always seek out the most reliable aquarium specialists when attempting to treat a problem unknown to you.

Medications have an expiration date beyond which they may lose their effectiveness. Light, moisture and elevated temperatures are factors that may dramatically accelerate this rate of degradation. Unfortunately, many remedies available in



pet stores do not carry any expiration date, nor are kept under acceptable storage conditions. Warm temperatures, moist environment, or stored on shelving under bright lights are factors that will affect commercial preparations sold in the hobby. Other times it is the hobbyist who decides to use old products! These situations should be avoided.

Unless you have a very large number of aquaria, do not buy medications "just in case". Few treatable conditions require an immediate use of medications, and in most cases it is better to start with water changes or just careful observation; then buy a fresh package of the exact medication that you need. Here are some useful tips to consider on the subject of purchasing and keeping medications:

- Buy medications from a shop that has a high volume of sales and stores them under acceptable conditions.
- Stay away from dusty or moisture-stained packages.
- Sealed containers are preferable to boxes or bottles that can be easily opened and contaminated.
- Formulations with an expiration date and a clear label explaining contents and concentrations are much more reliable than those with unknown or poorly described content.
- Store medications in a cool and dry place.
- If you opened an airtight container, it is unlikely that after a few months the content will still be viable, and you should dispose of it in a safe place.
- Keep you aquarium medications out of the reach of other pets or children.

#### **Disease Prevention**

Successful fish health management begins with prevention of disease rather than treatment. Fish health management can be as challenging and complex as the actual control of existing diseases. Prevention is accomplished through good water quality management, nutrition, and cleanliness. Without this foundation, it is impossible to prevent outbreaks of opportunistic diseases. Even the use of sterilisation technology; for example, ultraviolet sterilisers and ozonisation, does not eliminate all potential pathogens from the aquarium environment and will not prevent the spread of pathogens within the system. Key elements of disease prevention include the reliable detection of disease carriers, knowledge of how pathogens are transmitted, and development of effective methods to limit the entry of pathogens or carriers into uncontaminated aquariums, and the capability to provide environmental conditions conducive to good health.

Direct contact between fish is, except at breeding and during territorial disputes, rare in nature. In captivity, dense stocking in aquarium systems or transport bags and catching fish in nets may mean direct contact is an important route of disease transmission. When ornamental fish are sold by retail stores to the public, the fish and a volume of their aquarium water is generally transferred to a plastic bag. Although this allows the fish to be transferred to the purchaser's aquarium, it also represents one means by which the aquarium fish could be exposed to pathogenic organisms. The aquarium water supplied with ornamental fish purchased at retail outlets may contain significant numbers of a wide variety of pathogens.

Avoidance of exposure to disease is the primary method of prevention and the first consideration in preventing fish disease is effective quarantine. Proper and appropriate quarantine is a vital component of any successful fish health management program. There are several beneficial reasons for using quarantine before placing rainbowfishes into their final aquarium. First of all, newly arrived fishes can be isolated so their overall health condition can be properly and thoroughly evaluated and examined. Secondly, prophylactic measures can be taken and already diseased fishes can be isolated to prevent further spread of the problem. A quarantine tank also provides an easier way to treat diseased fishes rather than treating them in the main aquarium.

The term quarantine is defined as "isolation imposed on persons or animals that have arrived from elsewhere or been exposed to, and might spread, infectious or contagious disease." It is derived from the Italian quarantina, which means forty days. Quarantine (that is a forty-day period of isolation) can be applied to any animal, but was originally applied to humans and warm blooded animals. The expectation was that forty days (or other period of time stipulated by law) would be longer than the incubation period of serious diseases like small pox or rabies. Thus any infected animal would become identifiably unwell in that period.

Fish are not warm blooded, and their diseases do not have incubation periods that are similar in all conditions. Fish adopt the temperature of the water that surrounds them. If their environment is temperature stable, then the incubation period of a disease may be predictable. However, most fish are subject to quite wide fluctuations in temperature, and there are wide differences in incubation periods for diseases across this temperature range.

The length of quarantine should reflect the length of time required for disease entities common to the species to be detected, either via diagnostic procedures or clinical manifestations. Ideally, new fishes should be held in quarantine for at least 30 days. A period of at least 30 days should be adequate for most parasite problems to become apparent, as well as those caused by most bacteria. The same applies to plants, rocks and driftwood, which may also carry fish pathogens. Nevertheless, it must be recognised that certain species or disease problems may require more time.

Furthermore, to avoid or improve the possible consequences of environmental and physiological stress, an acclimatisation period may be useful. An acclimatisation period will let the fishes adjust to the new environmental conditions. Acclimatisation of new fishes should ideally begin before they arrive. This may involve acclimatisation to the temperature of the water, the light intensity, the *p*H, the chemical condition of the water and their new environment. It is important to know as much about the quality (i.e., temperature, *p*H, hardness, etc.) of the water from which the fishes are coming.



This way the environment in the quarantine tank can be adjusted to match the old environment. If need be, once the fishes have settled in, these parameters can be changed in small steps. Acclimation can be considered complete when the measurable parameters are the same (or at least similar) as those in the main aquarium. Disturbing the fishes as little as possible and keeping the lighting low will help reduce stress.

A quarantine system should be very simple so that fish are readily accessible for observation and handling, water can be easily changed, and treatments readily administered. A quarantine tank should be bare, with just a few plastic plants if the fish requires cover to prevent or reduce stress and a pre-cycled sponge filter. Quarantine tanks can be intrinsically more unstable than an established tank, and the importance of adequate water changes should not be underestimated, unless contraindicated by the treatment therapy being used. The walls and bottom of the tank should be kept as clean as possible. Even apparently minor slime coating (biofilm) on the glass can hide massive amount of microorganisms which are capable of causing health problems. A reliable and adequately powered heater with easy-to-adjust temperature settings should also be used. Ideally, rainbowfishes should be quarantined at a temperature of 22–25° Celsius.

A suitable quarantine tank should be available at all times. Such a tank doesn't need to be any larger than 50 litres and can be set up and maintained just like any other aquarium. This has to be done in such a way, as to prohibit physical contact, to avoid splashing and water contamination, or aerosolisation. Aeration and splashing creates small water droplets than can become suspended in the air as an aerosol. Aerosols can contain small pathogens such as bacteria and viruses. Particularly in humid environments aerosols can be long lived and thus act as a transmission agent for diseases between holding systems. Ichthyophthirius multifiliis and Aeromonas salmonicida have been shown to be transported by aerosols. Aerosol droplets persist longer in damp or humid conditions. Aeromonas bacteria have been known to spread at least seven metres in aerosols. That was the size of the room in which the experiment took place and may not represent the distance that could be travel in ideal conditions.

The quarantine tank should remain empty to receive new arrivals for the purpose of quarantine, and to take in any diseased fishes from the main aquarium(s) should the need arise. As a precaution against transmission of diseases, nets, syphoning equipment, buckets, and any other equipment used in the quarantine tank should not be utilised for any other tank.

Benefits of Quarantine:

- Evaluation of the health condition of the new fish.
- Diseases in stage of incubation may become manifest days or weeks after an apparently healthy fish is acquired.
- A quarantine tank allows a more effective observation of the fish than a display tank.
- Reduction of disease transmission risk to pre-existing fish.
- Although pathogens may be transmitted to other tanks by contaminated equipment (nets, etc.) or even by air-borne

particles, most bacteria and parasites remain contained to the quarantine tank until proper treatment eliminates them.

- More gradual acclimatisation of the new fish.
- A display tank is often a highly competitive environment where new fish are at disadvantage.
- Administration of medication or chemicals is convenient.
- Quarantine tanks are often smaller than the display tank, and fewer chemicals are needed (if dissolved in water).
- Less organic material that may inactivate the active medication.

Nevertheless, many aquarium hobbyists are not convinced of these benefits and show no interest in using or developing quarantine protocols. It should, however, be understood that while quarantine procedures greatly reduce the problems associated with the acquisition of new fish, there is no guarantee that any problems will be eliminated. Some diseases may have such a prolonged incubation period that it takes months before symptoms appear. In other cases, a new fish may simply be an asymptomatic carrier of an infectious disease. This means that the carrier does not show signs of the infection although a potential pathogen is present, and the quarantine is completed without any problems. However, other fish later infected by the same pathogen in the main aquarium may start to manifest symptoms.

#### General Maintenance

Cleanliness is one of the cornerstones of fish health management. Accumulation of organic material, often associated with inappropriate feeding and stocking rates, creates an environment where opportunistic bacteria, fungi, and parasites can flourish. To minimise this, water exchange should be adequate for the stocking densities and feeding rates. Be sure to precondition the water before adding it to your aquarium. Particulate matter (faeces, uneaten food, dead plant material etc.) should be removed on a regular basis. This includes removal of debris by syphoning, manual removal of algae from tank walls, and regular cleaning and removal of particulate matter from the filter. Any sick or dead fish should be removed promptly, as they are an important means of transmitting infectious disease to other fish in the system as well as adding to the organic load if they are left to decay.

Faeces can provide a survival capsule for some fish pathogens. Faeces is made of what remains of a fish's food after digestion, mucus, cells lining the gut that are shed continually, dissolved chemicals such as ammonia and harmless (and even helpful) bacteria naturally found in the gut and pathogens. Lipids (fats) can create a waterproof coating on the outside of the faecal material, thus the contents, including any pathogens present, are at least for a time insulated in a cocoon of material that is conducive to their survival. In recirculation aquarium systems little dilution or dispersion can occur. Thus not only is infection



passed to "clean" fish, but the infected fish are subject to reinfection. While a fish may be able to deal with a small dose of a pathogen, if subjected to continual re-infection a disease may be caused. This is especially so as in an untreated recirculation or under gravel system, the amount of the disease causing organism in the water rises and often the water quality falls, both of which make the fish more susceptible to infection and disease.

It is probably wise to regularly empty and disinfect breeding and raising aquariums to try to ensure that pathogens cannot build up in a system. You may choose to do this at the end of a breeding session to ensure any pathogens are not lying in wait to infect new fish placed in the aquarium. When disinfecting a aquarium system, remember to clean all parts including filter and water return lines. It is important that syphon hoses, nets, brushes, and other equipment used to clean tanks should be treated with a sterilising solution when used in different tanks. An easy way to achieve this is to have a plastic container into which equipment can be dipped or placed between uses. Equipment should then be removed, and thoroughly rinsed with fresh water, before being stored dry. Chemicals used should be minimally toxic to fish yet be effective at removal of infectious particles and other organic debris likely to accumulate on equipment.

Solutions of chlorine are effective for disinfecting equipment by submersion at concentrations of 200 mg/L for 30 to 60 minutes. Concentrations of 10 mg/L for 24 hours are effective for disinfections of tanks. However, repeated use and extended exposure of the silicon sealant to high chlorine concentrations will destroy or render the adhesive bond ineffective on glass aquariums with disastrous results. Also, certain materials may deteriorate after repeated exposure to chlorine. Chlorine will dissolve sponge filters and cause mesh nets to rot. Rubber and synthetic or natural fibres may degrade rapidly, but most plastics are unaffected.

If using chlorine for disinfecting equipment or tanks which are not in use, but which are in the vicinity of others housing live fish, the granular form (Calcium hypochlorite) should be used. Granular chlorine does not volatilise as readily as liquid chlorine (Sodium hypochlorite). In a poorly ventilated fishroom, fumes from liquid chlorine can cause fish kills in adjacent tanks. Chlorine fumes can also be harmful to the respiratory system of aquarists. Always wear eye protection and rubber gloves when handling large quantities of chlorine. Chlorine residue can be neutralised by using 7.4 mg/L Sodium thiosulfate for each 1 mg/L chlorine present in solution (7.5 grams of sodium thiosulfate will neutralise the chlorine present in 5 litres of a solution of 200 mg/L).

Commercial sanitisers such as benzalkonium chloride, iodophores, or quaternary ammonium compounds are often used. Sanitisers are compounds effective against all types of infectious agents including bacteria, fungi, viruses and protozoa and differ greatly in their physical, chemical and biocidal properties, mode of action, trade names, composition and availability. All should be used according to label instructions. Iodophores are effective against a broad range of bacteria and fungi and their spores. Although iodophores can stain plastic components. Quaternary ammonium products are cationic surfactants with strong bactericidal but poor sporicidal properties. They are also questionable fungicides and their germicidal activity is suppressed in the presence of organic matter. Chlorine compounds are traditionally the most popular disinfectants because of their rapid killing ability against many microorganisms. Although its activity is deceased by even small amounts of organic matter.

Regular control and monitoring of water quality is imperative and will greatly reduce the likelihood of a disease occurrence. Critical water quality parameters include temperature (particularly sudden and dramatic shifts), dissolved oxygen, pH, alkalinity, hardness, nitrogenous wastes, and toxic substances. Water quality should be monitored frequently and corrective measures initiated if conditions become unfavourable. Aim to keep your ammonia and nitrite at zero levels, and nitrate down to a minimum. Nitrate levels over 20 mg/L (ppm) can cause problems with excessive algae growth and can lead to fish health problems in the longer-term.

Optimum water conditions must be maintained at all times. Sub-optimum conditions, while not immediately lethal, may stress the fish, resulting in delayed mortality. Therefore, it is important to become familiar with water testing and have the necessary test kits available.

The use of a good quality food will provide the fish with all the nutrients that they need to remain healthy and to grow. Poor nutritional health can greatly enhance the progression and severity, and reactivation of disease. Rainbowfishes fed a nutritionally complete diet are better able to cope with stress and to resist disease. However, you should note that even good quality food will deteriorate if improperly stored or kept too long. Storage time for most commercial fish foods will vary depending upon environmental conditions; however, as a rule of thumb, 90 days is normally the maximum safe storage time for fish feed. Fish foods should be stored in a cool and dry place (refrigerator), and used within 30 days of opening. Never feed mouldy, discoloured or clumped feed. Moulds on feed may produce aflatoxins, which can kill fish.

Diets consisting of material derived from the wild, such as fish fillets or wild caught foods such as daphnia, mosquito larvae, tubifex, etc., are often perceived as having great nutritional benefits as they are believed to contain many macro nutrients essential to the good health of the animal. However, there is an inherent risk in using "wild diets" as they may introduce pathogens to the aquarium fish. These may take the form of parasites that use an intermediate host to enter their final host, or simply be concentrated by the feeding of the prey item to form an infectious particle which when ingested can establish the infection.

Potentially the highest risk here is the use of wild fish in diets which can, under appropriate conditions, be a very high risk factor allowing large amount of pathogen to enter the diet if the source material was infected. Zooplankton samples collected from the wild can carry diseases or parasites. Artemia cysts can carry bacteria such as vibrio and live collected daphnia, mosquito larvae and oligochaetes have been found to be contaminated with mycobacteria.



#### **Biofiltration**

It is not the aim of this section to explain how filters work to improve water quality. However, it is worth reiterating the point that the stress caused by poor water quality does create the conditions in which pathogens are likely to cause disease. Fish kept in good quality water are better able to resist pathogen invasion because it is more likely that their immune system is in better condition. That said, biofilters can act as a means of reducing pathogen loads but care is required as they can also harbour pathogens.

A well established biofilter can reduce the population of pathogens in an aquarium or pond system. The reasons for this are very complex and poorly understood; however, one of the most important consideration is the interaction between the bacteria and protozoa that colonise a biofilter. The normal flora of a biofilter consists of a well established population of bacteria and a varied population of protozoa either living in, on or in close association with the filter media. These organisms are very well adapted to their environments and:

- Produce enzymes that digest other bacteria and viruses from the water column
- Feed on bacteria or viruses directly (in the case of protozoa)
- Produce aggressins that mop up micro-nutrients from the surrounding environment, for example chemicals known as siderophores, take up iron directly from the environment. Aggressins are released to starve competing organisms.
- Produce natural antibiotics to prevent the new microbe becoming established.

They also have two other minor roles acting as a:

- Reservoir for bacteriophages (a type of virus) that can kill pathogenic strains of bacteria
- Mechanical filter trapping bacteria, viruses and parasites either in the media or on the biofilm.

The biofilm is a very hostile environment to new bacteria, each bacterial species that has already colonised the filter is competing for nutrients with each other; less aggressive species starve to death and, in turn their organic components are recycled. In addition to this the protozoa are consuming microbes continuously and again their waste and any dead protozoa will be recycled. All of these defence mechanisms used by the established bacteria can make it a slow process for a new species of microbe to become established in a filter. An obvious example of this is the long period of time it takes for a biological filter to mature from the ineffective filters colonised initially by bacteria such as *Pseudomonas spp.* to a bacterial flora dominated by Nitromonas, Acetobacter and Nitrosomas species. In an established system a mature biological filter can significantly reduce the level of circulating pathogens.

As always there has to be a word of warning with this approach to reducing pathogen load. The pathogen may become established in the filter and ultimately act as a reservoir of pathogen continually shedding pathogen into the environment. This can happen when the load of pathogenic bacteria is so high that it out competes the established bacteria in the filter. The pathogen will have its own suite of aggressins that it uses to survive in the filter biofilm. If present in sufficient numbers it can out-compete established bacteria populations. Should this happen then there is little option but to sterilise the biofilter with hypochlorite or something similar to remove the pathogen before restocking occurs. Some may argue that this should be done after any serious disease outbreak as part of the control regime.

### Handling Specific Problems

There are two broad categories of disease that affect fish, infectious and non-infectious. Infectious diseases are caused by pathogenic organisms present in the environment or carried by other fish and are broadly categorised as parasitic, bacterial or fungal. They are contagious, and some type of treatment will be necessary to control the disease outbreak. In contrast, environmental problems, nutritional deficiencies, or genetic anomalies cause non-infectious diseases, they are not contagious and cannot be cured by medications.

It is impracticable to cover all the diseases that rainbowfishes are capable of catching in an aquarium. There are many good books readily available on fish diseases and their treatment. However, I will cover some of the common diseases that you, or rather your rainbowfishes, are likely to encounter. Fish disease outbreaks are often complexed, involving both infectious and non-infectious processes. Therefore, appropriate therapy often involves medication and changes in husbandry practises. Often assistance from a qualified veterinarian or aquarium specialist will be required to help you treat disease outbreaks and develop a management program.

Compared to mammalian diseases relatively little (and in most cases very little) is known about individual fish diseases. The biology of most fish diseases (how they can spread, how infective they are etc.), especially those of rainbowfishes, is little understood, and thus assessments of and solutions to the problems they can cause is, at this point of time, based on incomplete information.

Fish health has been a relatively small discipline of veterinary attention in the past because of many factors, the most important of which is the perceived value of aquarium fish. Other reasons include the failure of pharmaceutical companies to become involved in ornamental fish health, or a failure of the standard of health care to keep pace with improvements seen in the care of other companion animals. This though is not the fault of veterinarians. As more people invest in expensive species, such as koi and various reef species, the demand to provide a higher level of care for these animals is increasing. This trend is also evident in the commercial food and bait fish industry, where aquaculture producers are expecting improved standards of care for populations of fish that are worth millions of dollars. With increasing numbers of aquarium and aquaculture operations, veterinarians will be expected to have the abilities and knowledge to diagnose and treat aquatic species and provide a standard of care commensurate with other commonly treated animal species.

#### **Parasitic Infections**

Most of the commonly encountered fish parasites are protozoans and can cause disease in their own right. In a



confined setting such as a home aquarium or pond, parasites can impair growth and reproduction and cause substantial morbidity and mortality. Parasites of the gills can cause irritation, leading to hyperplasia and increased mucus production, which may result in decreased respiration and ionexchanging capabilities. On the skin, parasites can cause defects that predispose the affected fish to osmotic imbalances and serve as a portal of entry for viruses, fungi, and bacteria. In the intestine, parasites compete for nutrients and cause ulcerations, inflammation, and emaciation. Several parasites actually physically carry pathogens. They may ingest infected materials and carry them to their next host.

Parasitic infections can be among the easiest to identify, and are usually the easiest to control. Protozoans are singlecelled organisms, many of which are free-living in the aquatic environment. Their reproduction cycle is temperature dependent, which affects treatment. Typically, no intermediate host is required for the parasite to reproduce (direct life cycle). Consequently, they can build up to very high numbers when fish are crowded causing weight loss, debilitation, and mortality. Most protozoans do not seem to bother the host fish until numbers become excessive. Uncontrollable or recurrent infestations with protozoans are indicative of a fishkeeping problem. Many of the parasites proliferate in organic debris accumulated in the bottom of the aquarium or pond. They are easily transmitted from tank to tank by nets, hoses, or on the fishkeepers' wet hands.

The major ectoparasitic pathogens and the diseases affecting rainbowfishes in captivity are Ichthyophthirius multifiliis (Ichthyophthiriasis), Piscinoodinium pillulare (Piscinoodiniasis) and Trichodina sp. (Trichodiniasis). These diseases probably account for almost 80% of all parasitic infections reported. Because of the lack of information on protozoan parasites of rainbowfishes, most cases are not identified or more often, are simply misdiagnosed. There is almost nothing known about freshwater parasites that affect rainbowfishes in their natural environment. Langdon et al. (1985) reported mortality of Melanotaenia tatei due to the ciliate protozoan Chilodonella hexasticha in the Finke River, central Australia. Rainbowfishes in captivity are also subjected to common fish parasites from ornamental species imported into Australia. However, there is such poor data on Australian fish parasites that there are doubts about what is endemic and what is translocated or introduced.

Ciliates such as *Tetrahymena*. *Ichthyophthirius* and Chilodonella can cause gill and skin lesions and may give rise to more serious disease if they invade internal organs. Infections often appear as small, white patches on the skin, especially around the eye. Because these organisms can survive off the host, the environment must be cleaned and disinfected in addition to treating the fish. Fish can develop severe osmotic imbalance due to parasitic damage to the skin and gills. Dinoflagellates such as Piscinoodinium are commonly found on the gills but may also affect the skin, fins, and gastrointestinal tract. In severe infections, the skin may become velvety gold in appearance (velvet disease). Mortality is attributed to severe osmotic imbalance. Trichodina species are usually indicators of poor water quality or overcrowding. They can survive off the host for 1 or 2 days and can be transported

with plants and other aquarium objects. Although they are not problematic in low numbers, heavy infestations can cause epithelial damage resulting in anorexia, loss of body condition, and low-level mortality.

Several larval trematodes infecting fish causing what is commonly known as "black spot" because of the characteristic, small (about 1–2 mm in diameter) dark brown or black spots which develop in the muscle and on the body, fins, gills and eyes of infected fish. They are easily visible to the naked eye. When the parasite infects the fish it forms a cyst (metacercaria) within the host tissue. The cyst then becomes surrounded by pigment cells, giving it the characteristic dark colour. "Black spot" infection is often found infecting wild-caught rainbowfishes but occur in several species of freshwater fish. Galaxiids and *Retropinna semoni* appear to be particularly susceptible to infection.

There are several species of trematodes which have larval stages which cause black spots; these species have yet to be identified. These trematodes usually will not harm the fish and will not progress unless the fish is consumed by an appropriate primary host animal. The adult trematode is generally found infecting fish-eating birds. There is no practical treatment or control of these parasite available at this time. If the metacercaria are not too numerous, they can be removed safely with a clean scalpel.

The larvae (glochidia) of freshwater mussels are parasitic on fish. They are released into the water by adult mussels and, when a fish passes close enough to disturb them, the glochidia attach themselves to the skin or gills of the fish by means of their barbed valves. Irritated host tissue then grows and forms a cyst over each glochidium. Development from glochidium to small mussel takes about 10 weeks, at which time the mussel bores through the cyst, leaves its host and settles to the substrate. The presence of a glochidia infestation is indicated by numerous white or greyish "bladders" on the gills, skin and fins of the fish. Fish may be severely stress by the attachment of large numbers of glochidia, particularly when the infestation affects the gills and may greatly impair respiration. Glochidia are able to affect most native species but are not known to affect introduced species.

Leeches are occasionally seen on wild or pond-raised rainbowfishes. Leeches resemble trematodes but are much larger and have anterior and posterior suckers. They have a direct life cycle with immature and mature worms being parasitic on host's blood. Pathogenesis varies with number and size of worms and duration of feeding. Heavily infested fish often have chronic anaemia. Fish may develop secondary bacterial and fungal infections at the attachment site. Dips in 3% (30g/L) saltwater are effective in controlling leeches. Ponds with heavy leech infestation require drainage, treatment with chlorinated lime, followed by several weeks of drying. This will destroy the adults and their cocoons containing eggs.

Monogenean flatworms such as *Dactylogyrus* and *Gyrodactylus* are also fairly common. These parasites may be found on the gills and skin, but *Dactylogyrus* are generally found on gill tissue. The life cycle is direct; the eggs of dactylogyrids hatch into free-swimming larvae that



locate a fish and begin maturation. Gyrodactylids, on the other hand, bear live young that spread to other fish through direct contact. Large numbers of these organisms can be lethal to fish, as both the attachment of the parasite by hooks and/or suckers and its feeding activity cause physical damage to the skin and gills.

Digenean trematodes have indirect and often complex life cycles involving two or more intermediate hosts. They are generally found in the gastrointestinal tract or musculature of the host. As they cannot complete their life cycle without intermediate hosts, these parasites are often an incidental finding in aquarium fish. However, they may cause severe internal damage in large numbers. Intermediate hosts (e.g., mollusks) should be removed from the environment to prevent fish-to-fish transmission. Final hosts (e.g., birds) should be deterred from outdoor ponds.

*Lernaea* (anchor worms), *Ergasilus*, fish lice, and isopods appear as tiny pill bugs or crab-like creatures. In low numbers, they may cause local inflammation and ulceration that can lead to secondary infections. Most of these parasites are rarely a problem in aquaria but can be common on wild-caught or pond raised rainbowfishes. Some of these organisms can be seen with the naked eye and removed manually.

Other parasites that cause clinical disease in rainbowfishes can be encysted in various tissues or reside in the gastrointestinal tract or other organs (e.g., gallbladder). These parasites include various species of cestodes, nematodes, acanthocephalans, and coccidia. They include harmless free-living types, such as roundworms, tapeworms, and trematodes. However, others have larval stages that live in lymph ducts and blood vessels, and they are difficult to treat without dangerous side effects. Most worms do not pose a serious health risk to rainbowfishes because they often have complicated life cycles in which the fish may serve as only one of possibly several intermediate hosts. Rainbowfishes with internal worms may appear completely healthy, exhibiting no symptoms of infestation.

Although specific therapeutic options vary for each organism, chemical treatment is of temporary value if water quality and management are poor. Aquarium conditions should be improved first. Many parasitic infections of fish are treated with medicated baths (e.g., formalin. praziguantel, metronidazole, etc.) or altered salinity. Copper sulphate is very effective for treatment of some parasitic infections. Lufenuron baths have been used to control crustacean parasites. Intestinal parasites are readily removed with various drug treatments. Gastrointestinal parasites may be treated by incorporating medication in a gel food form, but only if the animal is eating. Gel food can be homemade and is useful for administering common antiparasitic drugs (e.g., fenbendazole, metronidazole, praziquantel, pyrantel pamoate). These drugs can also be given via gastric irrigation using an appropriate-sized rubber catheter. Ivermectin should be avoided because it can cause neurologic signs and death in fish at therapeutic doses.

### **Bacterial Infections**

Bacterial disease is the most common infectious problem for aquarium fishes. Most cases require scientific identification of the bacterial types involved and selection of a specific antibacterial agent under guidance of a veterinarian. The most common bacterial infections in aquariums are caused by organisms such as *Aeromonas*, *Pseudomonas*, *Mycobacterium* and *Flavobacterium*. They can cause diverse pathological conditions that include both acute systemic and/or chronic diseases. The effects on the fish are as varied as the signs.

External signs of bacterial infection are variable and include shallow reddened ulcers with irregular edges; loss of tail and finnage; missing or raised scales; haemorrhagic areas on the body, in the fins, and on the mouth; protruding eyes (exophthalmia); dropsy; and a protruding and inflamed vent. Dropsy is a distention of the abdomen, giving the fish a "pot belly" appearance. This is a strong indicator of disease problems which may include swelling of internal organs (liver, spleen or kidney), the build up of body fluids, parasitic problems, or other unknown causes. At this stage, the infection has usually become systemic. External lesions expose the body surface to secondary invaders as well as serve as sites for the loss of salts and body fluids. Fish stop feeding and abnormal swimming may become pronounced. When diagnosed early, treatments available from an aquarium fish specialist may help.

### **Fungal Infections**

Fungi are a group of organisms that require living or dead matter for growth and reproduction. In most cases, fungi serve a valuable ecological function by processing dead organic debris. Fungal problems appear as cotton-like tufts on the body or fins of fish. Fungal infections are rare in a well-kept aquarium and are very seldom primary causes of disease. In most instances, fungus infections are secondary or tertiary infections. Unless the primary problem is solved, even an effectively treated fungal infection is likely to return. Most fungal infections of rainbowfishes and their eggs are probably associated with the fungi genera Saprolegnia and Achlya, although other groups are undoubtedly also involved. Achlya is commonly found on wild-collected rainbowfishes, which have had skin/scale damage during the collecting process. Epizootic Ulcerative Syndrome or 'red spot' disease has been identified in rainbowfishes from a number of river systems in the Northern Territory. This condition is frequently fatal to juvenile fish.

### Picornavirus in rainbowfish

Turquoise rainbowfishes (*Melanotaenia lacustris*) that exhibited whirling symptoms and obvious impairment of the central nervous system were submitted by a producer for diagnostic evaluation. All were negative for bacterial and parasitic pathogens, Electron microscopy revealed numerous picornavirus particles in the brain. Attempts to grow the virus in cell culture were unsuccessful. No similar cases have been reported since. Picornaviruses have been





observed as incidental findings in various freshwater fishes. The significance of their presence is unknown. A similar virus has been found in crayfish in Western Australia with associated mortalities, but the conditions under which disease outbreaks occur are not understood.

### **Obtaining Information**

Through the availability of books, magazines, the internet and aquarium clubs, the hobbyist has a wide range of information at their disposal. Problems arise though due to incorrect information being handed out or correct information being ignored, as hobbyists will often seek advice on the same problem from several sources, and this advice may well vary.

This problem occurs mainly on the internet with its wealth of information - and misinformation; anyone can publish anything on the internet. Be very careful what you accept as reliable information. Not everything in print on the Internet is factual or correct. It could be the latest brilliant piece of science, effective practical tips or garbage; you have to decide. For a variety of reasons information may be misguided, misinterpreted or misrepresented. Information may become dated or what was thought to be fact is proved incorrect. Of course it may be that there are two or more entirely reasonable interpretations of the "facts" and you must choose which to use. Today there is an increasing amount of information available from a variety of sources. Sometimes you need to do your own research and trials. The following comments might be useful in evaluating information sources:

Scientific Journals: these may or may not be "peer reviewed". Information in "peer reviewed" journals has greater creditability because the information has been read and edited by several other scientists before being published.

Textbooks: these may also be peer reviewed. They may also just reflect the author's own ideas or their interpretation of others' ideas and findings.

Magazines: care should be taken when evaluating information from this source. There may for instance be links between the article and the products sold by the author. This does not necessarily mean there is anything wrong just that a cautious approach may be best.

Internet: more and more information is available electronically. You can now access encyclopaedias, dictionaries and other traditional reference books online, as well as many journals, magazines and newspapers. There are also databases which not only provide citations to thousands of articles in scientific and other journals, but often have links to the full texts of those articles.

One final note on searching for information, whether in print or online: unless you are looking for a simple answer and find it right away, always check more than one source. That way you not only get more information but also ensure greater accuracy.

# White Spot Disease

Ichthyophthiriasis or white spot disease is regarded as one of the most pathogenic diseases of freshwater aquarium fishes. It is commonly known as "Ich" and has been a common problem to aquarists for many years. Most species of freshwater fish are susceptible to infection by this virulent parasite, although some may be more so than others. Infections have been reported in both cold and warm water species. The mature parasite is very large, ranging in size from 0.5 mm up to 1 mm in diameter, and becomes easily visible owing to the opacity of the cytoplasm in the fish's skin and the formation of a somatic cyst around the parasite. Immature forms of *Ichthyophthirius multifiliis* are smaller and more translucent in appearance.

Severe damage of the skin epithelium occurs due to the break of the parasites through the host skin during infection and their release. This damage might lead to concession of osmoregulatory process and ion regulation and might serve as a portal of entry for secondary bacterial or fungal invaders, leading eventually to death of the fish. If the disease is left untreated, outbreaks can result in 100% mortality. Even very young fry (larvae) can be infected causing substantial mortalities.

In aquarium systems, ichthyophthiriasis outbreaks are more common due to the confinement of fish under "unnatural" condition and the exponential increase in parasite numbers. Fish may maintain low, subclinical infection, while encysted tomonts may persist in the aquarium. Transition from nonclinical enzootic to epizootic clinical infection is often stressrelated, prompted by adverse aquarium conditions such as overcrowding, improper feeding and poor water quality.

### Life History & Biology

Ichthyophthiriasis is caused by the sub-epidermal ciliate protozoan *Ichthyophthirius multifiliis*. The life cycle of *Ichthyophthirius multifiliis* is a direct one and requires no intermediate host. *Ichthyophthirius multifiliis* have both fishassociated and free-swimming stages. The life stages of the parasite include a parasitic trophont, a reproductive tomont, and an infective theront. Theronts can penetrate the epithelium of susceptible fish within minutes thus completing the life cycle. Visible spots on the fish (the clinical sign for which the disease is named) are individual parasites known as trophonts. However, a single white spot does not necessarily represent a single trophont, since aggregations of trophonts can occur in one large white spot as a result of multiple entries at single site.

Invasion of the infective theront gives rise to the trophont (parasitic feeding stage) that grows inside the host epithelium. By maturity, which is reached in 2 days at 25–28°C (3–4 days at 21–24°C), the parasite evacuates the host tissue and settles within 2–6 hours on a substrate (gravel, plants, aquarium glass, etc.) to form a cyst-encapsulated tomont (reproductive stage). Parasites evicted from the tissue before the scheduled time for their spontaneous departure, fail to develop into tomonts and eventually die. Within the cyst, tomonts undergo successive binary fissions with a resulting yield of 250–2000 theronts



(infective, free swimming stage), which after release will seek a suitable host. The division of tomonts into theronts, in temperatures of 25–28°C, is completed within 15–20 hours. After being released, the free-swimming theronts can infect a new host or re-infect the same host, thus compromising its health status.

Invasion of theronts (20–50 µm long) into the host integument is facilitated by the excretion of a sticky substance. In the absence of a suitable host, theronts will lose their infective potential within 24 hours at 24–28°C. Higher temperatures hasten trophont maturation and tomont division, but at lower temperatures, slower development allows the growth of larger trophonts (0.8–1.0 mm in 5–10°C vs. 0.5–0.7 mm in 20–24°C), yielding tomonts with higher numbers of theronts. In lower temperatures the survival of the theronts is prolonged, thus, allowing more time to locate a host. Low temperatures do not interrupt propagation; a full cycle is completed at 20°C in 3~5 days, at 15°C in 7–14 days and at 10°C in 21–35 days. Data on the effect of other environmental parameters is less conclusive, although it has been suggested that dissolved oxygen levels below 1 mg/L affect parasite reproduction.

Some researchers have reported that *Ichthyophthirius multifiliis* can multiply directly by dividing underneath the fishes' top skin layer, bypassing the usual three-stage life cycle. When this occurs, one can see multiple cells of similar size lined up or in clumps underneath the thin layer of host cells. The disease is not treatable when it becomes established to this degree and reproduces in this manner, because it does not need to leave the host where it would ordinarily be vulnerable to treatment.

### Diagnosis

Identification of the parasite is necessary to conclude that the fish has an ichthyophthiriasis infection. Look closely at the clear parts of the fins for cloudiness and/or white pinhead size spots. Rainbowfishes infected with this parasite typically develop small white spots on the body, fins and gills. If the infection is restricted to the gills, no white spots will be seen. The gills will appear swollen and be covered with thick mucus. In some cases, the small white spots coalesce together forming larger white spots. Hemorrhagic patches appear on the bases of the fins, body and mouth. Fin rot sometimes appears on some affected fish. The presence of white spots along with associated haemorrhages and behavioural changes is considered the ultimate indication for the presence of an infection with ichthyophthiriasis.

Early warning signs include loss of appetite and listlessness. The fish will scratch or flash on the sides of the aquarium or other objects. As the disease progresses the fish may congregate near the filter outlet or appear to gasp for air. The fish may produce a thick mucous over the skin in an attempt to protect itself. Occasionally the mucus will come off removing many of the *Ichthyophthirius multifiliis* cells and leave the skin dry. When this occurs, the outer defence against infectious invaders is removed and essential salts and body fluids are lost. Within six days, if left untreated, the fish will stopped feeding, appear lethargic and swim

near the water surface. Death is likely after about 10 days. Skin scrapping under a microscope will revealed the presence of different developmental stages of *Ichthyophthirius multifiliis*. The most predominant stage will be the mature trophont with the C-shaped macronucleus.

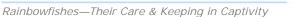
### Treatment

The first step in the successful treatment of ichthyophthiriasis disease in aquarium fishes is early recognition and proper diagnosis. The organism can only survive if live fish are present for completion of its life cycle. If only one parasite is seen, the entire system should be treated immediately. *Ichthyophthirius multifiliis* is an obligate parasite and capable of causing massive mortality within a short time, and in severe cases, control may be impossible. Because both trophont and tomont stages are resistant to practically all externally applied antiparasitic chemicals, a single treatment is not sufficient to treat the parasite. Infection can be effectively controlled only by destruction or elimination of the free dividing tomonts or the tomites (theronts) they release. Repeating the selected treatment every other day at water temperatures from 20–25°C will disrupt the life cycle and control the outbreak.

Daily cleaning of the aquarium is also beneficial, as the encysted forms are physically removed from the environment. The use of one of the gravel-cleaning vacuum devices is recommended. These are usually available at most aquarium stores. The design of the gravel vacuum ensures that even very small fry are not drawn in or damaged by it, whilst retaining sufficient suction and mechanical action to remove the cysts and other debris. An additional benefit is that the vacuum is also very effective at removing uneaten feed and faeces, resulting in improved water quality and other environmental benefits.

There are two main drugs which have been found to be highly effective for treating Ichthyophthirius multifiliis in freshwater systems: malachite green and formalin. Formalin is a generic term, which describes a solution of 37-50% formaldehyde gas dissolved in water. These chemicals can be used separately, but are particularly effective when used together, because they exert a synergistic effect; that is, together they give a greater effect than the sum of their separate individual capabilities. The combination of both chemicals is less toxic than either drug used separately. Malachite green and formalin are toxic poisons. Malachite green acts as a respiratory poison, damaging the cell's ability to produce energy to drive vital metabolic processes. Formalin is a powerful disinfectant used to kill microorganisms or as a preservative for biological specimens. It works by reacting with cell proteins and nucleic acids - altering both structure and function. Formalin is more toxic in soft, acidic water and also removes oxygen from water. Each 5 mg of formalin removes 1 mg of dissolved oxygen from the water.

The malachite green/formalin (37%) combination is used at a concentration of 0.15 ppm and 25 ppm respectively for 24 hours. They exert a mild anti-bacterial effect but in most circumstances will not destroy biological filtration bacteria, although they may have a slight effect for a short period.





As you might expect both of these chemicals are affected by variations in water chemistry. Both can be deactivated by high dissolved and particulate organics such as fish waste, detritus and algae. In water that is high in dissolved organic wastes the chemicals will oxidise the organic wastes rather than attacking the parasites. This has the effect of lowering the effectiveness of the chemical dosage. It is a good idea to do a 50-75% waterchange and a thorough cleaning of the aquarium to reduce the level of organic wastes before starting the treatment. This includes the removal of particulate matter (faeces, uneaten food, detritus, etc.); the removal of algae from tank walls and the removal of particulate matter from the filter (change filter wool or wash sponge etc.).

Treat the fish every other day for a total of three to five treatments. Change 50–75% of the water in between the chemical treatments. In simple terms, treat on day 1, waterchange on day 2, treat again on day 3, waterchange on day 4 etc., etc. If the fish is heavily parasitised, you may not see any remission of the disease until after the third treatment. Treatment effectiveness should be evaluated after the third treatment schedule. If you use the correct method and dose of malachite/ formalin and the fish do not show some signs of improvement within 3 treatments, you may have misdiagnosed the problem.

The chemicals must be at full strength when the theronts are free swimming in the water. This is why it is important to observe and be aware of the cycle of appearance and disappearance of the spots on the fishes. The treatments (including the water changes) should still be done when the spots have disappeared or decreased in number.

It is often suggested that the temperature of the aquarium water be raised. This is because the free-swimming, infective stage theronts are heat sensitive and raising the temperature several degrees above the normal aquarium temperature tends to kill them. In addition, increased temperatures enhance the fishes' immune responses. The treatment consists in increasing the temperature of water with infected fish to 30-32°C and maintaining it there for ten days. After treatment, the water is allowed to gradually cool to the original temperature. However, given that temperature stress might have been a contributing factor in the start of the disease it is not logical to change the temperature again. Also, increasing the temperature, as many aquarists do, might actually increases the stress on the diseased fishes. As the water temperature increases the dissolved oxygen content is reduced proportionately. For instance, increasing the temperature from 20 to 30°C decreases the dissolved oxygen level by more than 17%. Additionally, the metabolic rate of the fishes also increases as the temperature goes up. This causes the fishes to consume more oxygen at a time when the increased temperature causes a decrease in the oxygen concentration.

### **Treatment Summary**

(1) Don't increase the water temperature!

(2) Remove carbon and other chemical filtration media; thoroughly clean or replace the mechanical filtration media. Most aquarium medications are readily adsorbed by activated carbon. Use fresh carbon to remove the medication after treatment is completed.

(3) Do as large a water change as possible before starting the treatment.

(4) Add malachite green/formalin formulation at the recommended dosage.

(5) Re-treat as indicated (above) by the cycle of the disappearance and reappearance of cysts (spots) on the fishes. Re-treat at least every 24 hours. Do large water changes prior to each re-treatment.

(6) Continue treatments and water changing for at least 24 hours after the disappearance of all signs of the disease.

### Remarks

Several alternative medications for treatment of ichthyophthiriasis have been developed, but there has been some debate as to how effective they are compared to the widely used malachite green and formalin combination. For many years, malachite green has been used (often in combination with formalin) to treat fish infected by *Ichthyophthirius multifiliis*. In recent years, however, there have been strong moves against malachite green application, especially with respect to its use in food fish. This is because the chemical is believed to have potential teratogenic, mutagenic or carcinogenic attributes. However, a recent study (2005) has shown that most of the new treatments are not as effective as the malachite green and formalin combination. The study reported that with the exception of one treatment, all were less reliable than the malachite green and formalin products they are supposed to replace.

The study tested the performance of formalin, potassium permanganate (KMnO<sub>4</sub>), chloramine-T, hydrogen peroxide and two new chemicals called Per Aqua<sup>®</sup> and Desirox<sup>®</sup>. The latter two products, which are mixtures of acetic acid, peracetic acid and hydrogen peroxide, were also tested on their own, and in conjunction with formalin. The results showed that all of the chemicals were able to successfully lower the parasite burden so that the mortality rate dropped within a month of the fish picking up an infection. However, with the exception of one product, none were as good as the malachite green - formalin mixture. Large differences in parasite burden and mortality occurred among the replicates in all except the Desirox-formalin tests, which means that they were not as reliable as the malachite green - formalin combination. It was also evident that the chemicals and their concentrations must be planned carefully to suit the conditions of each situation.

In Germany, as an alternative substance, peracetic acid (peroxyacetic acid) was tested to treat the free invasive stage (theront) of the *Ichthyophthirius multifiliis* parasite. Peracetic acid concentrations of 0.3 ppm were able to kill all theronts in 120 min in the laboratory tests. As a result of these investigations it was recommend an interval-application of 0.3 to 0.5 ppm peracetic acid for 30 to 150 min. This application should be prolonged for two life cycles of the parasite. Biotic parameters as e.g., fish species, and age as well as abiotic parameters as e.g. temperature, pH and organic load of the water could possibly influence the efficiency of the peracetic acid application and should therefore be taken into account when picking the dosage and length of the peracetic acid exposure.



Peracetic acid is an ideal antimicrobial agent due to its high oxidising potential. It is broadly effective against microorganisms and is not deactivated by catalase and peroxidase, the enzymes which break down hydrogen peroxide. It also breaks down to safe and environmentally friendly residues (acetic acid and hydrogen peroxide). It can be used over a wide temperature range (0–40°C), wide *p*H range (3.0–7.5), in hard water conditions, and is not affected by protein residues.

### Other Treatments

#### Sodium chloride (NaCl)

A continuous well-aerated salt bath of 2–5g/L until disease controlled (may be for up to 20 days) has been reported as effective in controlling ichthyophthiriasis. One study found that fish maintained at 4g/L salt for 23 days showed a gradual reduction of white spots and survival was 100%. There appear to be significant differences among species and possibly families in the tolerance of larval and fry stages to salt.

The efficacy of salt in controlling and preventing ichthyophthiriasis in the silver perch (*Bidyanus bidyanus*) was evaluated in aquaria. Concentrations of 2 or 3g/L salt controlled infestations of *Ichthyophthirius multifiliis*, and fish were free of both theronts and trophonts by day 8 at temperatures of 17.3–21.3°C and by day 6 at 19.2–23.5°C. Fish treated with 1g/L salt remained infested and all fish in a control treatment (0 g/L salt) died.

#### Copper sulphate (CuSO<sub>4</sub>)

Copper sulphate at concentrations of 0.1–0.2 mg/L have been reported to control ichthyophthiriasis, but higher concentrations of 0.25–1.0 mg/L were toxic to some fish species, and 0.05 mg/L was ineffective. However, copper sulphate is for specialist use only as it is highly toxic and requires removal from the aquarium after treatment. It is inadvisable to use this compound where other treatments are available.

Sometimes three to four daily transfers of fish to clean tanks will effectively reduce infection, while enabling the fish to develop tolerance to reinfections. Maintain temperature at around 24–28°C. Management techniques such as alleviate stressing conditions by improving water conditions, reducing stocking densities, etc., can also be helpful.

### Research

Increasing reports are continually being published indicating that different fish species and populations have significance difference in their resistance to *Ichthyophthirius multifiliis*. These differences in susceptibility were attributed primarily to environmental factors and/or genetic make up of the host (Gleeson *et al.*, 2000). Wild-caught rainbowfish originating from three isolated populations were infected with a quantified dosage of *Ichthyophthirius multifiliis* in a controlled environment. *Melanotaenia eachamensis* from Dirran Creek were much more susceptible to ichthyophthiriasis than were *M. splendida* from the Lake Tinaroo or Bluewater

Creek populations. When the highly susceptible Dirran Creek rainbowfish were crossed with rainbowfish from a fourth population, M. eachamensis (Lake Eacham), they produced hybrids with significantly higher resistance than pure-bred Dirran Creek, but not higher than pure-bred Lake Eacham fish. Hence, intraspecific hybridization increased resistance to Ichthyophthirius multifiliis infection in M. eachamensis. Hosts from all three populations were much less susceptible to infection on their second exposure to the parasite. However, the Bluewater Creek population was better able to acquire immunity to Ichthyophthirius multifiliis than either the Dirran Creek or Lake Tinaroo populations. It was tentatively suggested that there may be a link between the heterozygosity of populations of rainbowfish and their initial ability to resist infection by Ichthyophthirius multifiliis.

Although infection with *Ichthyophthirius multifiliis* is often lethal, a number of studies have indicated that sublethal infections in the host are able to induce an acquired resistance against re-infection. Under laboratory conditions fish have been routinely immunised by exposure to controlled numbers of parasites. Serum and mucus antibodies from immune fish have immobilised free-swimming theronts in vitro, suggesting several potential antibody-mediated mechanisms of protection.

Spontaneous recovery from infection and resistance to reinfection of recovered fish indicates that fish are capable of developing defence mechanisms against *Ichthyophthirius multifiliis*. Spontaneous recovery has been observed in both natural infections in natural habitats and in captivity. The potential for spontaneous recovery varied with fish species. Infection in scaled fish regressed faster than in smooth skinned fish. After recovery, fish were resistant to reinfection or retained a merely subclinical chronic infection. The infective stages were also shown to be unable to penetrate the skin of resistant fish or reacts in such a way which forces the parasite to leave the body without completing its life cycle in response to the immune reaction.

The likelihood of maternal immunity could be raised as another assumption for the mild infection in some fish species. It is well documented that maternal antibodies passed from mothers to their offspring directly via eggs or indirectly in mouth-brooding fish via mucus of the buccal cavity.

Another line of reports assumed the presence of more than one strain of *Ichthyophthirius multifiliis* which differ in their pathogenicity to different fish species. This assumption was based primarily on the wide distribution of the parasite, subtle variation in cell morphology and serotypic variations among isolates based on immobilisation antigens. In 2006 a team of parasitologists from the Department of Infectious Diseases at the University of Georgia's College of Veterinary Medicine studied two forms of *Ichthyophthirius multifiliis* and found that one of them killed all of the fish infected while the other only killed half.



# **Velvet Disease**

Piscinoodiniasis or velvet disease is quite common in aquarium fishes. Outbreaks tend to be of an explosive nature, and large numbers of fish can succumb to the disease within the course of a few days. The parasite appears to be non-specific and indiscriminately infects various fish species invading skin, fins and gills. Occasionally, the occurrence of trophonts is reported from the oesophageal epithelium and intestine, and even in the subcutis. It has historically been responsible for a large number of fish mortalities. However, aquarium hobbyists rarely have infected fish closely examined to confirm that the infection is consistent with Piscinoodiniasis.

Piscinoodiniasis is often confused with ichthyophthiriasis (white spot disease). It is caused by the dinoflagellate *Piscinoodinium pillulare*, formerly known as *Oodinium pillularis*. Dinoflagellates are (unicellular) protists that exhibit a great diversity of form; this group includes gill and skin parasites that can cause serious disease of freshwater and marine fish e.g., *Amyloodinium* (saltwater), *Crepidoodinium* (estuarine and marine) and *Piscinoodinium* (freshwater). All three genera have been traditionally classified as belonging to the family Oodiniaceae. The relatedness of species belonging to these three genera is primarily based upon a similar mode of attachment to the host, i.e., attachment disc with holdfasts. Their life cycle is also similar, with each having 3 stages: a parasitic trophont; a reproductive tomont; and a free-swimming infective dinospore.

The first dinoflagellate parasite recorded on a fish host was *Amyloodinium (Oodinium) ocellatum* in 1931, which was found on a marine fish. The first dinoflagellate found on a freshwater fish was *Oodinium limneticum*, which was identified from aquarium fish in North America (Jacobs, 1946). Schäperclaus (1951) described a second species, *Oodinium pillularis*, from aquarium fish in Europe. In 1981, the genus *Crepidoodinium* was created for *Oodinium cyprinodontum* and *Piscinoodinium* was created for *Oodinium pillularis* and *Oodinium limneticum*. However, it is unclear as to whether *Oodinium limneticum* is a valid species.

*Piscinoodinium pillulare* is a sedentary parasite that attaches to the skin, fins, and gills of fish. Infestations of the skin are usually less serious than those affecting the gills. The rhizoids of the trophont actively penetrate into the epithelium of the host fish, which responds by means of a pronounced hyperplasia, often sufficiently intense as to entrap the parasites themselves. Areas of haemorrhage and focal necrosis appear which are frequently invaded in a secondary manner by bacteria and fungi. In the gills, the response ranges from desquamation and separation of the epithelial layer to a manifest hyperplasia affecting the entire length of the gill filament. Degenerative changes and necrosis of the gills are frequently observed in severely affected fish.

*Piscinoodinium pillulare* is a dangerous ectoparasite of aquarium fishes and is most pathogenic to young fish. Although young fish may die quickly from piscinoodiniasis, older fish may live for 2–3 months. The pyriform, sack-like

trophont is up to 160  $\mu$ m long and has an amber or yellowishgreen colour, visible on heavily infected fish.

The life cycle of *Piscinoodinium pillulare* is comprised of a parasitic feeding stage (trophont) which attaches to integumentary epithelial cells, and an encysted dividing stage (tomont) which is detached from the host. Mature trophonts drop of the host's surface, sinks to the bottom and becomes a tomont. Tomonts undergo successive binary fission and divides successively into 64 or 128 small cells. They divide again to produce 128 or 256 cells which differentiate into free-swimming infective dinospores. The division of tomonts into dinospores, in temperatures of 23–25°C, is completed within 4–6 days. At 15–17°C, the process of division is lengthened to 11 days. After being released, the free-swimming dinospores can infect a new host or re-infect the same host, thus compromising its health status.

There is some evidence that suggest fish recovering from the epizootic infestation through a gradual decrease in infection develop immunity against re-infections, and specific antibodies have been demonstrated in the blood serum of infected fish.

### Diagnosis

The diagnosis of cases of piscinoodiniasis is based on the clinical signs observed, and detection of the typical trophonts in wet mounts from the gills or the skin. Trophonts, when reaching the final stage of growth, are visible to the naked eye (80–100  $\mu$ m diameter) as white spots (similar to that seen in ichthyophthiriasis). On the body surface, an increase in mucous production, scale loss, suffusion, ecchymosis, petechiae, and small ulcers may be observed. The gills may also present an increase of mucous production besides epithelial hyperplasia, suffusion, petechiae, congestion, oedema, and brownish areas. Visible signs begin as a light golden dusting in oblique light and then progress to more severe infestation intensity associated with dense white dusting of the skin. This surface sheen is easiest to observe by placing the fish in the dark and shining a beam of light through the water parallel to the surface of the skin.

Infected fish generally swim near the surface of the water or gather near the filter outlets, and the fins may be folded. Fish with skin infections may exhibit 'flashing' behaviour. Fish with heavy gill infections typically exhibit rapid respiration (spreading opercula) as large numbers of parasites compromise gill function. Other clinical signs included dyspnea, lethargy, cachexia, localised secondary infections, and erratic swimming with loss of equilibrium. Mortalities progressively increase.

### Prevention

Successful fish health management begins with prevention of disease rather than treatment. Regular control and monitoring of water quality is imperative and will greatly reduce the likelihood of a disease occurrence. Without this foundation, it is impossible to prevent outbreaks of opportunistic diseases. *Piscinoodinium pillulare* tend to be opportunistic pathogens of aquarium fish and outbreaks can be treated and also controlled by improving hygiene, water quality and reducing stocking



densities. It's possible that the infective dinospore stages can be transmitted in aerosol droplets, particularly where tanks are situated in close proximity. Therefore, proper disease preventative management should be observed. All syphon hoses, nets, brushes, and other equipment that has been in contact with the infected fish should be disinfected.

#### Treatment

Parasites numbers on individual fish can be rapidly reduced by osmotic shock (e.g., altering either temperature or salinity beyond that tolerated by the parasite) if the fish can tolerate such treatment. It must be remembered that salt tolerance by fish may vary with the species and age. Baths in sodium chloride at 35 g/L for three to ten minutes are effective in dislodging the trophonts, but care should be exercised to avoid reinfestation when the fish are returned to their aquarium.

The life cycle of this parasite can be completed in 10–14 days at 22–25°C, but lower temperatures can slow the life cycle. Also, the cyst stage is highly resistant to chemical treatment. Therefore, several applications of a treatment may be necessary to eliminate the parasite. The free-swimming dinospores are usually the stage that is most susceptible to a variety of interventions including chemotherapy.

Chloroquine phosphate (15 mg/L prolonged bath) has been reported to be effective. Other treatments include malachite green (at 0.05 to 0.1 mg/L), formalin (at 15 to 25 mg/L) or a mixture of both malachite green/formalin (37%) combination used at concentrations of 0.15 ppm and 25 ppm respectively. The malachite green/formalin mixture has been shown to be more effective and less toxic than either drug used separately. Formalin baths are somewhat effective against trophonts but required rather prolonged immersion and may not completely kill the tomonts, which may resume development once the formalin has dissipated. Quinine hydrochloride (at 30 mg/L), or a combination of quinine hydrochloride and malachite green (at manufacturer's recommendation) has also been used with some success.

Under laboratory conditions (Schmahl et al. 2006), the ionophoric polyether salinomycin given in the fish diet was shown to be effective against the skin-inhabiting trophozoites of Piscinoodinium pillulare. Experimentally infested swordtails (Xiphophorus hellerii) were fed once a day ad libitum food pellets containing either 60 or 90 ppm salinomycin for a 16 or 19 days period. The efficacy of the treatment was monitored by counting the numbers of trophozoites of each fish at day 0, 3, 6, 9, 12, 16 and 19, respectively. As revealed by transmission electron microscope investigations, the damages in the trophozoites caused by the treatment consisted in malformation of the trophozoites, aggregation of droplets within the cytoplasm, and the formation of electron dense bodies along the limiting membrane. Following a prolonged treatment period, ruptures in the trophozoites limiting membranes were seen and the rhizocysts were no more detectable. Under the experimental conditions described, fishes showed no signs for adverse effects.

Copper sulphate (CuSO<sub>4</sub>) will kill both dormant (destroying the chloroplasts) and the free swimming stage as well. However, copper sulphate treatment can be unpredictable under certain aquarium conditions and is extremely dangerous to some species of fish, plants, shrimp and snails. Copper sulphate may be used as a bath for up to 10 days duration, at a concentration of 0.15 ppm of copper ion.

Copper sulphate should never be used without testing the alkalinity of the water, carefully measuring the volume of the aquarium or pond to be treated, and weighing the amount of chemical to be applied. The concentration of copper sulphate to apply is usually calculated by determining the total alkalinity of the water and dividing that number by 100. For example, if the total alkalinity of the aquarium is 100 mg/L, then  $100 \div 100 = 1$  mg/L copper sulphate. Do not use copper sulphate if the total alkalinity is less than 50 mg/L. If you are unsure how to measure the alkalinity of your water, or have never used copper sulphate, then do not use it. Copper can also bind to the substrate in the aquarium and leach back into the water for a long time. In addition, copper levels must be monitored frequently if good results are to be expected, and this may not be practical for the average hobbyist. Therefore Copper sulphate is best used by aquarium specialists.

When using a commercially formulated copper cure, always follow the label instructions for dosage rates. Chelated copper will stay in solution longer than copper sulphate and appears to be safer to fish. You can create your own chelated copper by using two parts citric acid to one part copper sulphate, by weight. Combine both in distilled water and dissolve them together. It is important to remember that you will be treating with the copper sulphate and not the citric acids, so when weighing the formula, use only the weight of your copper sulphate in calculating dosages.

Most fish are extremely sensitive to copper. Concentrations of copper as low as 42  $\mu$ g Cu/L were found to be acutely toxic to *Denariusa bandata*. *Melanotaenia inornata* and *Ambassis* species have been found to be sensitive to copper; half of the individuals tested died at copper concentrations between 120 and 200  $\mu$ g Cu/L. The freshwater shrimp genus, *Caridina*, is extremely sensitive to copper, dying at levels of only 2  $\mu$ g Cu/L. *Macrobrachium* species were found sensitive to copper with half the individuals dying at 160  $\mu$ g Cu/L. Snails are also known to be very sensitive to copper.

It is important to keep in mind that all fish medications are toxic to fish. Fortunately, it usually takes a higher concentration of the drug to harm the fish than it does to harm the pathogens. Nevertheless, subtoxic doses for the fish are still stressing, and repeated doses can build up to toxic levels.



# **Trichodiniasis**

Trichodiniasis is the disease caused by infection of the gills and/or integument by peritrichous ciliated protozoa of the family Trichodinidae. Trichodinidae are collectively known as trichodinids. They are recorded from freshwater fish species worldwide. The majority are believed to be non-pathogenic, whereas a few species are known to be primary pathogens of fishes causing considerable mortality particularly among juveniles. Infection is common in native and introduced species, including aquarium species and occurs in both captive and wild populations. Although trichodinids are commonly found in low numbers on healthy fish in their natural environments, the presence of any trichodinids on aquarium fish warrants treatment.

In general, trichodinids are ectocommensal; however, some species are certainly primary pathogens and are parasitic on fishes. Some species may infect hosts other than fish, such as amphibians. They are characterised by the presence of a ring of interlocking cytoskeletal denitcles, which provide support for the cell and allow for adhesion to the surfaces of the fish. Trichodinids have a simple direct life cycle. That is, they have a single host and do not use alternation of generations or mass asexual replication off the host. The reproduce by binary fission, literally cell-splitting. This produces daughter cells with half the number of denticles of the parent cell. The full complement of denticles is restored by synthesis of new denticles from the outer edge of the cell, working inwards.

Trichodinids are typically found on the gills, skin and fins of fishes, though some species parasitise the urogenital system. A range of invertebrates is also host to trichodinid infections, including the surfaces of copepods and the mantle cavity of molluses. Transmission occurs by direct contact of infected and uninfected hosts, and also by active swimming of trichodinids from one host to another. Trichodina cells swim with the adoral surface facing forwards. On surfaces, they move laterally, with the adoral surface facing the substrate. Few trichodinids are host specific and have been reported to survive up to 1-2 days off their host.

Trichodiniasis is usually a relatively mild disease with a low morbidity and mortality, however, under certain conditions, especially confinement of infected fish in aquaria, clinical disease and high mortalities may occur. The length of incubation (the time from infection to the appearance of the first signs of the disease) depends on susceptibility, temperature, and severity of exposure. Infectious signs can appear within 2 hours of initial exposure and attain 100% prevalence after 10 days. Larvae and fry are particularly susceptible to infection.

In heavy infections, trichodinids attach firmly to the epithelium causing irritation and cell damage resulting in hyperplasia of the epithelium and mucous secretion. Infected fish are generally darker in coloration and may exhibit a whitish or bluish-grey colouration of the skin. Fish may exhibited listlessness and lack of appetite. At advanced stages of the infection, they adopt a vertical hanging position near the water surface with continuous lethargic swimming motions. A "tattered" appearance of the skin and fins may be apparent, with strands of mucous apparent. In mild infections, trichodinids graze over the epithelial surface feeding on particulate matter and detritus and cause insignificant damage. However, in combination with inappropriate aquarium conditions, secondary bacterial infection and intercurrent disease, high mortalities may occur. Because of the lack of information on protozoan parasites of rainbowfishes, most cases of trichodiniasis are not identified or more often, are simply misdiagnosed.

Although trichodinid protozoa commonly occur on native fish in Australia, most genera and species remain to be described. In a research project (Dove & O'Donoghue, 2005) freshwater fishes were surveyed across eastern Australia. Trichodinid ciliates were collected on an opportunistic basis during the survey for other parasites that encompassed 58 sites throughout Victoria, New South Wales, the Australian Capital Territory and Queensland. Twenty-one putative trichodinid species were recovered from the 33 fish species examined. *Trichodina heterodentata*, *T. mutabilis* and *T. reticulata* were the exotic species recovered regularly; a single specimen matched a fourth exotic species, *T. acuta*.

Two new native species were also described: *Trichodina cribbi* from *Hypseleotris galii*, *H. klunzingeri*, and *Hypseleotris sp.* (Lake's Carp Gudgeon); and *T. bassonae* from *Selenotoca multifasciata*. The native fishes of Australia harbour a species-rich fauna of native trichodinids, with at least 17 species of undescribed trichodinids detected from 33 species of native fishes examined from survey sources. It is estimated that the Australian trichodinid fauna may include up to 150 as yet undescribed species.

### Treatment

Trichodina can be controlled with any of the treatments used for ichthyophthiriasis. Correction of environmental problems is necessary for complete control. It is a good idea to do a 50-75% waterchange and a thorough cleaning of the aquarium to reduce the level of organic wastes before starting any treatment. This includes the removal of particulate matter (faeces, uneaten food, detritus, etc.); the removal of algae from tank walls and the removal of particulate matter from the filter (change filter wool or wash sponge etc.). Trichodina infections are often associated with monogenean trematode infections and treatment has to be applied for both.

Control of trichodiniasis has been successful using a long term bath of malachite green added to aquaria at 0.08 mg/L. A Formalin bath at a concentration of 250 ppm for one hour, or 40–50 ppm for a longer period has also been used with success. Common salt (NaCl) at 2–3% concentration is another successful treatment for infected fish.

However, a malachite green/formalin combination is probably the best treatment for aquaria. This combination exerts a mild anti-bacterial effect as well, but in most circumstances will not destroy biological filtration bacteria, although they may have a slight effect for a short period.



WS3, a commercial medication containing malachite green, acriflavine and quinine sulphate has also been used as a bath treatment combined with a salinity of 3‰.

One should follow the manufacturer's instructions for treatment, as different manufacturers use different concentrations of the active ingredients.

Garlic (*Allium sativum*) and Indian almond (*Terminalia catappa*) have been used as an alternative to chemicals to treat trichodiniasis. There is a fast growing interest in screening antiparasitic substances from plants to replace chemical and antibiotic alternatives. In laboratory trials, the use of crude extracts of garlic and Indian almond at 800 mg/L have been reported to significantly eliminated *Trichodina* infections in tilapia. However, the amount of activity varies widely among the different varieties of garlic and its use may prove to be unrealistic; the same may be said of Indian almond.

# **Gill & Body Flukes**

The monogeneans (flukes) are typically ectoparasites of the skin, fins and gills of many freshwater and marine fishes. Some species become endoparasitic by inhabiting the oral cavity, cornea, nasal tissue, pharyngeal tooth pads, urogenital system (cloaca, rectal gland, oviducts) and even the vascular system. In all cases they are attached to the host's surface by a characteristic opisthaptor (an attachment organ) provided with hooks, suckers or clamps. With the exception of the members of the viviparous family Gyrodactylidae, monogeneans usually have a simple life cycle involving hermaphroditic adults, eggs and larvae, and can proliferate rapidly on fish held in captivity.

Unlike the other monogeneans, the larva of the viviparous family Gyrodactylidae is retained in the uterus until it develops into a functional preadult, inside which a second larva is already formed, with a third larva inside that and a fourth inside the third. The steps of this sequential polyembryony are poorly understood. After its birth, this preadult larva begins feeding on its host and gives birth to the second larva remaining inside it. Only then may an egg from its own ovary become fertilised, repeating in a short time the development described above. Since this peculiar embryogenesis does not involve a free-swimming infectious larva, Gyrodactylidae depend on transmission of adults or pre-adults from one host to another by direct contact.

There have been very few studies of monogenean parasites from Australian freshwater fishes. The group is morphologically diverse and identifying them correctly is often impossible. Up until 1998 twenty-six species had been described from sixteen species of native freshwater fish. A study published in 2004 (Corlis, 2004) described another nineteen new species. These were found on Melanotaenia splendida, M. М. eachamensis, maccullochi, М. trifasciata, Rhadinocentrus ornatus, Cairnsichthys rhombosomoides, Craterocephalus stercusmuscarum, С. marjoriae, Pseudomugil signifer and P. gertrudae.

A very large group of fluke parasites infecting rainbowfishes have an intermediate live stage in molluscs and crustacea. Some of these parasites cause problems in both wild caught and captive rainbowfishes.

Monogenean parasites can be introduced into the aquarium from wild-caught fish. Although monogenean parasites are commonly found on wild fish, they are rarely a direct cause of disease or death in wild populations. Some destroy their hosts, but most are specific for one species of fish or for two or more closely related species. Because of high host specificity, most Monogenea do not spread to other fish species when they are introduced with their hosts into a new environment. However, they may become more dangerous to their hosts in the new surroundings. The establishment of exotic monogenean populations on Australian native fishes via host-switching is considered less likely than for other parasitic groups due to the generally high host specificity of monogeneans.

Transmission of monogenean parasites from fish to fish is primarily by direct contact. Oviparous monogeneans release eggs into the water which hatch and mature into adult forms within 7 days, depending on temperature. Viviparous monogeneans release live larvae which may attach to the same host as the parent or be carried by the water circulation to another. This direct life cycle can contribute to population explosions under aquarium conditions. Under the right conditions they can literally "bloom" like algae.

Monogenean flukes can be devastating to aquarium fishes. They appear on the gill filaments as tiny dark spots about 600  $\mu$ m long. They are a group of parasites best described as flatworms - the body is usually flat and oval. Most flukes are browsers, moving about the body surfaces, and feeding on skin mucus and gill tissue. Most species are host and site-specific, requiring only one host to complete an entire life cycle. In fact, some adults will remain permanently attached to a single site on the host. Large numbers of flukes on either the skin or gills may result in significant damage and mortality. Therefore, knowledge and understanding of monogenean parasites is important for aquarium hobbyists.

Mortality of aquarium fish caused by excessive parasite infestations is usually associated with inappropriate aquarium conditions, undernourished fish or have been subjected to temperature fluctuations. Heavy infestations may elicit epidermal hyperplasia due to disruption by parasite attachment and feeding; increased mucus production may also occur. Secondary infections by protozoa, bacteria and fungi often ensue and can cause deep wounds and ulcers. In severe infestations, death occurs, usually due to loss of osmotic regulation. A few flukes on a healthy mature fish are usually not significant; however, moderate numbers on a young fish can cause significant mortalities.

Freshwater fish infested with flukes become lethargic, swim near the surface or gather near the filter outlets, and subsequent loss of appetite. They may be seen rubbing the bottom or sides of the aquarium (flashing). The skin where the flukes are attached show areas of scale loss and may ooze a pinkish serous fluid. The gills may be swollen and pale, respiration rate may be increased, and fish will be less tolerant of low oxygen conditions, showing other signs of respiratory distress. Gulping air at the water surface may be observed in severe respiratory distress.

### Treatment

Prevention of fluke infestations by following appropriate quarantine practices is preferable to treating the parasites after they have become established in the main aquarium. This can be done by dipping fish prior to placing them into an established aquarium, and following a quarantine protocol whenever feasible. If quarantine is not possible, a simple way to minimise introduction of flukes, as well as other external parasites, is to dip rainbowfishes in a saltwater bath (30–35 g/L) for three- to five-minutes. Dipping rainbowfishes in a salt bath will eliminate many single-celled external parasites. This practice will not completely eliminate the risk of introducing parasites to an established tank or that no parasites will be introduced with new fish, but it will help minimise the numbers brought in.

Ideally, rainbowfishes should be quarantined for at least four weeks prior to being placed into a new system. While in quarantine, prophylactic treatment with a broad-spectrum parasiticide should be carried out. A quarantine system should be very simple so that fish are readily accessible for observation and handling, water can be easily changed, and treatments readily administered.

Treatment of flukes is usually not satisfactory unless the primary cause of increased fluke populations is found and alleviated. If the disease is not in the acute phase, a low salinity bath (5 g/L for 90 minutes) or (1 level tablespoon per 20 litres of water) is often enough to solve the problem if combined with water changes and general cleaning of the aquarium environment.

Praziquantel has been identified as the most effective "in water" treatment of infected fish. Praziquantel is harmless to fish of all species, is non toxic to plants, and has no negative filter impact. Praziquantel is a bitter tasting powder which shows good absorption directly from the treated water, and then admirable clearance of various surface and internal flukes and worms in fish. Praziquantel has been known to the hobby for many years. Praziquantel was traditionally available in the form of branded Droncit<sup>®</sup> tablets, for oral administration in dogs and cats, but is now available in a range of aquarium products.

Praziquantel used at 2–3 mg/L is very effective for control of both gill and body flukes and has a wide margin of safety for fish. Praziquantel is toxic to flukes on contact, paralysing the parasites within 15 seconds under laboratory conditions. Praziquantel preparations must be dosed high enough and long enough for effective treatment. Monogeneans can be persistent in aquarium systems necessitating regular treatments. In cool water, the parasites move through their life cycles slowly, so it is important to medicate long enough to intercept the emerging larvae. When temperatures are above 25° Celsius, treat once every 3 to 4 days for a total treatment time of 20 days. When temperatures are between 20 and 25° Celsius, treat once every 4 to 5 days for a total treatment time of 25 days.

The eggs can be resilient to chemical treatment, which make the use of multiple chemical treatments appropriate to control this group of organisms. Praziquantel can also be administered in food at a dosage of 35–125 mg/kg for up to 3 days or as a short-term bath treatment at a concentration of 10 mg/L for 3 hours.

Change 50–75% of the water in between the chemical treatments. Fish, which are obviously weak and heavily parasitised may not survive. Management to lessen the chance of infestation by these parasites includes maintaining the fish in a good nutritional state and avoiding water quality problems that might weaken the fish.

The effectiveness of the long-term use of Praziquantel has been evaluated in ornamental fish. Cumulative doses up to 10 mg/L water were tolerated without side-effects by Angel Fish (*Pterophyllum scalare*), Discus, and a variety of catfish species (*Ancistrus sp., Corydoras sp.*). It was found appropriate to start with a dosage of 2.5 mg/L and to add the same dosage every other day several times. All adult parasites and larvae were killed by this treatment. For the complete elimination of Dactylogyridae populations in a closed aquarium system, 3 therapy-cycles (duration: 5–6 days, accumulated dosage: 2.5 mg/L/day) proved to be effective. It was important to interrupt the therapy-cycles with intervals without medication (1 to 4 weeks). However, there are reports that kissing gouramis (*Helostoma temminckii*) have been adversely affected by Prazifish<sup>®</sup> (Praziquantel 98.5 mg/g).

Dactycid<sup>®</sup> is another effective treatment against flukes and all species of internal worms (hookworms and roundworms) including *Camallanus cotti*, hydras and planarians. However, it can be toxic to snails and is harmful to sharks and rays. Flubendazole (15%) is unique and specifically designed and developed for aquatic use and approved by the Veterinary Medicines Directorate under the small animal's exemption scheme to be sold over the counter for public sale. This medication will kill flukes, camallanus worms, tapeworms, anchor worms and other helminthics commonly found in tropical and coldwater fish. It has also been used successfully with 'skinny disease' in clown loaches.

Organophosphates can also be used to treat monogenean parasites, as well as leeches and crustacean ectoparasites Argulus (fish lice) and Lernaea (anchorworm). Organophosphates work by interfering with the nervous system and thus affect vital physiological processes. However, organophosphates are potentially dangerous to both fish and humans and, for a variety of reasons; their use in fish disease control has been banned in many countries, although the use of organophosphates in aquaria is still practiced.

The two most commonly used organophosphates are dichlorvos and trichlorfon. The concentrations of active ingredients vary in the different preparations. Historically, trichlorfon was used at a concentration of 0.25 mg/L active ingredient. When trichlorfon is added to water it degrades to dichlorvos, which then slowly degrades to less toxic by-products. The quicker it breaks down, the higher the initial dose needs to be.



The rate at which it degrades depends on various factors: light, high temperatures, aeration and high pH all speed up degradation. In mild temperatures and moderate pH the half-life can be several days, which means that it is active for longer. In alkaline conditions and high temperatures the half-life can be as short as a day. Also, organophosphate uptake and toxicity in fish is increased by low oxygenation of the water. These factors result in variable response of fish to exposure to organophosphates, with levels greater than 0.1 ppm being potentially toxic. Monogenean flukes can reinfest fish between 5 and 30 days after treatment with organophosphates, depending on water temperatures and degree of pond contamination with eggs. Up to three treatments, 21 days apart may be required to eradicate or reduce monogenean numbers to an insignificant level.

Spinal deformities have been reported in rainbowfishes (Melanotaenia pygmaea) after treatment with trichlorfon, at a rate of 0.5-2 ppm. Organophosphates exert their toxic effect by inhibition of acetylcholinesterase, an enzyme involved in terminating neurotransmission at cholinergic synapses in the central nervous system, some peripheral autonomic junctions and neuromuscular junctions. In intoxicated fish that don't die acutely from central nervous system dysfunction, the muscle spasms produced by excessive and prolonged stimulation of neuromuscular junctions of the muscles of the body are thought to be sufficiently severe to result in spinal fracture and lesions. Other choices include potassium permanganate, formaldehyde and maybe others. These chemicals can also be dangerous when used incorrectly and should only be used by experienced aquarists. There have been reports of flukes being resistant to certain types of treatment such as formaldehyde.



# **Bacterial Disease**

Bacteria can play both a beneficial and detrimental role in aquarium keeping. On the beneficial side they help break down ammonia, nitrite and organic wastes in the aquarium, which is a requirement for maintaining water quality. Conversely, bacteria can cause serious disease problems in aquarium fish. Only water quality problems exceed bacterial diseases in the area of aquarium fish mortality. Nevertheless, they are among the least known and understood elements of aquarium keeping.

The ubiquitous nature of bacteria in aquatic environments provides ample opportunity for rainbowfishes to come into contact with pathogenic and opportunistic bacteria. Such contact may lead to infection which, depending on the species and the virulence of the strains encountered, may have lifethreatening consequences. Some bacteria are even capable of causing illness in fishkeepers who may acquire infections from contaminated aquarium water.

Aquarium systems support large populations of bacteria. One study sampled water from an aquarium store and found a wide variety of potentially infectious organisms in every sample tested. Most of these are opportunistic and ubiquitous in aquatic environments. However, some bacterial growth is inhibited by the presence of other bacteria, suggesting that competition is a factor limiting the growth of some infectious bacteria.

Like other areas of aquarium keeping, bacteria have certain environmental preferences. However, most species tolerate a wide range of environmental conditions. An aquarium, with its warm water, aeration, nutrients and surfaces for colonisation, creates an environment where bacteria will flourish. Many of these bacteria live in biofilms that are located on all surfaces and in particular in the biofilter. Because the biofilter is full of different microniches, it can support the growth of a variety of microorganisms including pathogenic and opportunistic bacteria. The use of granulated activated carbon provides a readily available source of nutrient carbon for bacteria thus encouraging the growth of massive numbers of bacteria on its surface. However, bacteria are also present within the water column, and are capable of living independently away from their fish host.

Bacteria in biofilms are more resistant to a wide variety of antimicrobials including surfactants, antibiotics, phagocytic predators and drying. When conditions in the aquarium are optimum for the resident bacteria, planktonic cells are released from the biofilm and are capable of causing persistent and recurring exposure to disease and the presence of asymptomatic carriers.

In an aquarium, outbreaks of disease are usually associated with overcrowding, high or sudden change of temperature, handling, aggressive interactions, inferior water quality and poor nutritional status. All these conditions contribute to physiological changes and heighten susceptibility to infection. These management problems must be corrected for successful, long-term control of infections.



Avoidance of exposure to the disease is the primary method of prevention. Keeping aquarium systems pathogen-free is an impossible task, but reducing levels of pathogens to below infective levels, should decrease the chance of fish becoming clinically infected.

Bacterial organisms may be the primary cause of disease, or they may be secondary invaders. Bacterial diseases can be transmitted by discharge from the intestinal tract and external lesions on the skin. Parasite damage and fungal infection may also allow entry and spread of the bacterial infection. In captivity, rainbowfishes will gradually develop resistance to some local strains of bacteria but may carry virulent organisms to another aquarium when transferred.

Fish can act as asymptomatic carriers of disease. In other words, they may be immune to a specific pathogen but still be able to shed the organism into the water or transfer it to other fish by contact. Sick and dead fish are often major reservoirs of disease-causing organisms. For this reason, sick, moribund (dying), and dead fish should be removed as soon as possible from the aquarium to prevent continued shedding of the bacteria into the water. Equipment, including nets, siphon hoses and buckets, can also transfer disease-causing organisms to another aquarium.

The most common bacterial infections in aquariums are caused by organisms such as Aeromonas, Pseudomonas, Mycobacterium, Flavobacterium and Vibrio. They can cause diverse pathological conditions that include both acute systemic and/or chronic diseases. External signs of bacterial infection are variable and include shallow reddened ulcers with irregular edges; loss of tail and finnage; missing or raised scales; haemorrhagic areas on the body, in the fins, and on the mouth; protruding eyes (exophthalmia); distended abdomen (dropsy); and a protruding and inflamed vent. Dropsy is a distention of the abdomen, giving the fish a "pot belly" appearance. This is a strong indicator of disease problems which may include swelling of internal organs (liver, spleen or kidney), the build up of body fluids, parasitic problems, or other unknown causes. At this stage, the infection has usually become systemic. External lesions expose the body surface to secondary invaders as well as serve as sites for the loss of salts and body fluids. Behavioural signs include a reduction in feeding and listlessness. Fish stop feeding and abnormal swimming may become pronounced. The effects on the fish are as varied as the signs.

### Treatment

Many bacterial diseases can be successfully treated with medication when diagnosed early. An experienced fish health professional should carry out treatment for all but the most common bacterial disease problems that your fish experience. Most cases will require scientific identification of the bacterial types involved and selection of a specific antibacterial agent. However, even veterinarians with laboratory diagnostic experience cannot make an accurate diagnosis of some problems without microscopic examination of the fish or cultivation for bacteria. For the detection and identification of bacterial pathogens in populations of fish showing disease signs, ideal samples are multiple (five or more) moribund fish or those showing clinical signs typical of the disease outbreak. For the detection of subclinical infections in populations of asymptomatic fish, larger sample numbers may be necessary. Fish that are found dead at the time of sampling are usually not acceptable for bacteriological examination, unless they are known to be very fresh. Contaminating bacteria can grow quickly in dead fish, particularly in warm water.

If the fish have a bacterial disease and the causative agent has been identified, a sensitivity test should be performed to ensure that the correct medication is used. The incidence of resistant bacteria is high and a sensitivity test will show the resistance of the diseasecausing bacteria to various antibiotics. When bacteria become resistant to a specific antibiotic, even high concentrations of that drug will not be effective. Certain antibiotics also have been shown to suppress the immune system, potentially making aquarium fish more susceptible to viral or parasitic infections.

Antibiotics are usually taken internally to control bacterial infections. Therefore, medicated feed or injection, are preferred for treating systemic (internal) bacterial infections. Dose rates are based on fish weight and are expressed as weight of chemical per weight of fish per day for a specified number of days. Improper doses may result in an ineffective treatment or mortalities. However, the effectiveness of oral antibiotic therapy has been inconsistent and, as a consequence, mortalities continue to occur.

Fish often stop eating as a bacterial disease progresses, so early diagnosis and treatment are essential to ensure that infected fish consume the medicated feed. Medicated feeds may be unpalatable to the fish or the fish may be too sick to consume the normal amounts. Withholding food for a few days prior to feeding medicated food may aid in acceptance. It has been suggested to enhance the food with cod-liver oil.

If commercial medicated feed is not readily available, it is possible to mix your own feed in small quantities. A qualified fish health professional should be contacted for help in calculating the appropriate quantity of medication needed before using this treatment.

The powdered antibiotic is combined with a binder and then added to the feed. Antibiotics can be introduced into artificial feed by making a solution of 5% gelation, cooling the solution to room temperature and adding the antibiotic. The gelatineantibiotic solution can then be mixed with the diet, which is then stored in a refrigerator at  $4-6^{\circ}$  Celsius. Vegetable or fish oil can work well as binders also. The feed and antibiotic must be mixed thoroughly to assure even distribution of the drug to all the feed. If you use the antibiotic to coat dry feed then the coated feed should be spread out to air dry. After several hours of drying, the feed can be re-bagged and stored under proper conditions. This can be a time-consuming process. In addition, a significant quantity of the antibiotic may leach out of the homemade medicated feed before being consumed by the sick fish.



# Fin & Tail Rot

Fin or tail rot is a common problem in aquarium fish. It describes a variety of infections including lesions, erosion, splitting of fin rays, nodular thickening and extensive loss of tissue. It can be caused by inappropriate aquarium conditions which may increase susceptibility to infections. Often it is the result of aggressive interactions that cause fin damage and secondary infection from a range of bacteria or fungal spores. Fish having mild cases can completely regrow the soft fin ray tissue if infection is resolved.

The most common bacteria involved are usually Aeromonas, Pseudomonas and Flavobacterium. These bacteria are often natural inhabitants in the skin mucus of fish. However, definitive diagnosis is by fish necropsy, culture and isolation of bacteria. Affected fish become anorexic, lethargic and dark in colour. They generally have a rather gross appearance with frayed fin edges and reddened caudal fin in the early stages. The tip of the fin becomes greyish, and then it becomes eroded and hemorrhagic. The lesions progress into fin rot or extensive fin loss, and in more advanced cases, the caudal fin will erode back to the peduncle, with ulcerated tissue often exposing the skeletal structure. Eventually, even the muscle fibres will be affected. Secondary fungal infection is common. A high mortality rate may be observed within a few days unless rapid identification of the disease-causing pathogen and treatment of the infected fish is undertaken immediately. Mortality increases significantly once bacteria enter the circulatory system.

A research project in 2004 investigating 170 naturally infected fishes with finrot revealed clinically progressive erosion, congestion and haemorrhages of the body fins especially the caudal and dorsal fins and oedema (dropsy) in some cases. The post-mortem results of the infected fishes showed abdominal ascites (accumulation of fluid in peritoneal cavity), enlargement and congestion of the liver, kidneys, spleen and intestine with distension and congestion of the gall bladder. The infected fishes revealed the presence of 468 bacterial isolates related to 8 bacterial genera and species, such as Aeromonas hydrophila (198), Pseudomonas fluorescens (102), Flavobacterium columnare (36), Klebsiella (48), Escherichia coli (24), Proteus (12), and Shigella (12). The pathogenicity of isolated strains revealed that Aeromonas hydrophila appeared to be highly virulent (100% mortality in infected groups) followed by Pseudomonas fluorescens (50%) and Flavobacterium columnare (37.5%).

Sensitivity test of isolated bacteria strains showed that Kanamycin and Nalidixic acid were the drugs of choice used for control and treatment of finrot disease. Kanamycin mixed with food has been reported as effective in curing finrot among ornamental fishes at 200–300 mg per 100 grams of food. It has also been reported that Kanamycin is absorbed from the water by fishes. Dose rate: 2–5 gm/L for 4–5 days. Afterwards make a 35–50% water change. Nalidixic acid (water soluble form) can be used as a one- to four-hour bath at a rate of 13 mg/L, repeat if needed. However, the use of antibiotics should be considered carefully before any application.

Bacterial finrot is very similar to fungal finrot with much the same symptoms. Fungal finrot looks like creamy or whitish, hairy-like growths. Bacterial finrot is whitish or gray, and although it causes the same amount of damage, the growths are not as prominent. Fungal finrot usually has a more uniform infection attacking all areas at the same rate. Bacterial finrot causes varying degrees of damage in some areas to little or no damage in other areas.

Bathing in a salt solution or various proprietary medications will usually prevent further development. There are several medications available commercially to treat finrot. One should follow the manufacturer's instructions for treatment, as different manufacturers use different chemicals and concentrations of the active ingredients. Malachite green at 1-3 mg/L of water for up to 1 hour has been effective in some cases. Another effective treatment is immersion in a salt bath. Salt (Sodium chloride NaCl) is one of the most commonly used treatments for aquarium fish and is sometimes referred to as "aquarium aspirin". Most tropical fish can tolerate a salt concentration of 1-3 g/L, and this level is not harmful to the biological filter. Short baths for 20-30 minutes at 10 g/L for fish species capable of tolerating certain salinity levels can be used. Most adult rainbowfishes seem to be able to tolerate salinities levels up to 17 g/L and juveniles 12 g/L. In a review conducted by Hart et al. (1989) the general conclusion was that most freshwater fish species in Australia appear quite tolerant of salinities up to 10 g/L.

## **Columnaris Disease**

Columnaris is the common name for disease caused by the bacterium *Flavobacterium columnare*, which often gives the appearance of white fungal growth on the fish, but is actually a bacterial infection. It is characterised by gill necrosis, greyish white spots on the body, skin erosion, and finrot. It is mostly an external infection but the bacterium is capable of entering the blood stream and is often isolated from the internal organs. Fish may die however, without any clinical symptoms. The biology of the disease is poorly understood even though it is one of the oldest known fish diseases. *Flavobacterium* is a genus of gram-negative, aerobic or facultatively anaerobic bacterium that consists of at least ten recognised species. They are highly heterogeneous and comprised of pathogenic and non-pathogenic species. Several species are known to cause disease in freshwater fish.

*Flavobacterium columnare* is ubiquitous (i.e., found in water and soil), and on the skin of healthy fish worldwide. It was first described by Herbert Spencer Davis in 1922. It has been a significant problem in many warm water fish species for decades and is among the most common pathogens in aquaculture, ornamental, and wild fish populations. Its name is derived from columnar shaped bacteria, which are present in virtually all aquarium environments. It may be opportunistic or of a secondary infectious nature. References to the disease can be confusing. It has been referred to by different names including *Bacillus columnaris, Chondrococcus columnaris, Flexibacter columnaris, Cytophaga columnaris,* and most recently *Flavobacterium columnare* (Bernardet *et al.* 1996).



*Flavobacterium columnare* is generally of low pathogenicity although the pathogenesis of the disease is not all that clear. In acute infections, hypoxia and death may result from extensive damage to the gills. Columnaris disease is usually transmitted by direct contact with infected fish or contaminated water. The infection can be expected to spread most rapidly if water conditions are less than ideal.

In most instances, bacterial infection occurs in fish that are exposed to stressful conditions, such as high water temperature (~28°C), overcrowding, excessive handling, and poor water quality, especially high ammonia or nitrite concentrations or increased organic waste content (~2 g/L) in the water. However, spontaneous, natural infections with Flavobacterium columnare have been reported in the absence of any obvious stressors. Spontaneous infections usually involve highly virulent strains and are associated with high mortality. Mortality rates can be extremely high, with 60-100% mortality common. Several days elapse before mortality results from infection by low virulence strains. High virulence strains cause death within 24-48 hours post exposure to the pathogen. High mortality (~100%) has been reported with acute Flavobacterium columnare infections in the absence of obvious clinical signs or lesions. The morbidity, mortality, and course of disease depend mainly on the water temperature and virulence of the bacterial strain.

In an experimental infectivity study, mortality did not occur at  $5^{\circ}$ C or  $10^{\circ}$ C. In contrast, 25% mortality was observed at  $15^{\circ}$ C, with a mean death time of 7 days, whereas 100% mortality was recorded at 20°C and higher, with a mean death time of 1 to 3 days. This study demonstrated that increasing temperature is associated with increased mortality during bacterial infection. Sudden changes in water temperature of  $5^{\circ}$ C or more pose significant stress, predisposing fish to infection by *Flavobacterium columnare*. The ideal temperature for the growth of *Flavobacterium columnare* is 20–25°C, although it can grow at temperatures ranging from 4–37°C. Good survival of the bacterium occurs over a wide range of water *p*H and hardness.

The initial clinical signs of columnaris disease are nonspecific and include listlessness, lethargy, inappetence, swimming near the water surface, and accelerated opercular movement. The disease can have different clinical manifestations, with various combinations of gill, skin, or fin lesions. Columnaris disease associated with primary gill involvement is acute, with mortalities occurring in the range from 2–5 days. Fish with peracute columnaris disease may be observed lying on their sides. Characteristic skin discoloration and ulcers are not usually observed. Sudden and vehement onset of columnaris disease is usually observed in younger fish that die within 1–4 days without visible lesions. Fish with established columnaris disease usually have lesions on the external body surface and gills.

The distribution of these lesions tends to vary with the species of the fish. In scaleless fish, the skin lesions begin as areas of discoloration, primarily at the base of the dorsal fin. However, lesions may also be seen on the head and craniodorsal part of the body. As skin lesions spread from the base of the dorsal fin, a pale white band extends laterally and encircles the body to form a characteristic, pale white, "saddleback" lesion. A yellowishwhite ulcer often develops in the centre of the "saddle" as the lesion progresses. In advanced disease, extensive and deep skin ulcers may develop, exposing underlying muscle and bone. In scaled fish, prominent gill and fin necroses are usually observed; however, skin ulcers may also be present in the absence of gill and fin lesions. Necrosis of gills and fins begins at the outer margins and extends from the distal end toward the body. Gill necrosis is observed as yellowish-white spots on the tips of the primary lamellae of the gills. Initially, the skin lesions are less prominent in scaled fish but become obvious as the skin lesions advance from mild hyperaemia to deep skin ulcers. The scales become loosened and slough off as the skin disintegrates. Grossly, bacterial mats can be seen attached to skin and/or fins and have a typical "cotton wool" appearance.

In aquarium fish species, inflammation of the mucous membrane of the mouth is common. The mouth and inner walls of the oral cavity may be covered with a yellowish-brown mucoid-like growth. This condition is popularly called cotton-wool mouth, and fungi are frequent secondary invaders. The infection may involve the opercula, teeth, maxillae, mandibles, and the spongy bones of the head.

The diagnosis of columnaris disease is based on cytological examination of smears from skin, gills, and fins; histopathology examination of tissues obtained during necropsy; and microbiologic culture, which is the most reliable technique for the definitive diagnosis of *Flavobacterium columnare* infection. The lesions are characterised by the presence of long, thin rods that exhibit flexing movement and are able to form columns. To verify the diagnosis, isolation of *Flavobacterium columnare* is required. However, isolation is often problematic, because the disease usually presents as a mixed infection with numerous other opportunistic bacteria belonging to the normal skin flora.

### Treatment

Columnaris disease can be treated successfully, provided it is diagnosed early. Avoidance of exposure to the disease is the primary method of prevention. An aquarium that has had a columnaris outbreak should be completely disinfected before restocking if all the fish have been lost. In addition, all equipment that has been in contact with the infected fish should be disinfected. This disease can be spread easily between tanks from contaminated nets, shared equipment, etc.

Because *Flavobacterium columnare* primarily attacks the skin and gills, infections in the early stages usually respond to treatment using surface-acting disinfectants. The in vitro growth of *Flavobacterium columnare* is reported to be inhibited at 10 g/L Sodium chloride (NaCl). A salt bath at 10 g/L of water for 20–30 minutes can be used. Stop the treatment earlier if the fish show signs of stress.

When treating systemic (internal) infections, medicated antibiotic feed or injection, are the preferred methods. In such cases an appropriate antibiotic can be given as an in-feed preparation. The U.S. Food and Drug Administration have approved the use of Florfenicol as a medication feed additive



for the treatment of *Flavobacterium columnare*. Dose rates are based on fish weight and are expressed as weight of chemical per weight of fish per day for a specified number of days. However, the effectiveness of oral antibiotic therapy has been inconsistent and, as a consequence, mortalities continue to occur.

Antibiotics are only effective in treating bacterial diseases if treatment is applied very early during the course of the disease. *Flavobacterium* have shown resistance to chloramphenicol, streptomycin, ampicillin, tetracycline, chlortetracycline, oxytetracycline, neomycin, nitrofurazone, nalidixic acid, kanamycin and penicillin G. The resistance of *Flavobacterium columnare* to polymyxin and neomycin has also been reported by Fijan, Griffin, Bullock *et al.*, Bernardet and Grimont. When bacteria become resistant to a specific antibiotic, even high concentrations of that drug will not be effective.

# **Fungal Disease**

Mycologists and others have given much study to aquatic fungal diseases infecting fish species, but the results have not always been in agreement. They comprise tens of thousands of species and an extensive number are associated with health problems in fish. The name saprolegniasis is used because it relates particular disorders of fish much more succinctly to fungal infections than do such names as "fungused fish". The identity of the various organisms associated with fungal diseases in fishes has long been a problem.

Fungi are a group of organisms called heterotrophs, meaning that they obtain their energy and carbon compounds from organic nutrients. They are saprophytic (feed on decaying organic matter) and parasitic organisms that require living or dead matter for growth and reproduction. Unlike plants, they are incapable of manufacturing their own nutrients by photosynthesis. Fungi are present everywhere – in most cases, they serve a valuable ecological function by processing dead organic matter.

Almost every freshwater fish will be exposed to at least one species of aquatic fungi during its lifetime. All fungi produce spores - and it is these spores which readily spread disease. The spores of the fungus are always present and will take any opportunity to grow and develop. Fungal spores are freeswimming in water and can therefore be introduced or transferred between aquariums. Aquatic fungal infections are considered difficult to prevent and treat, and are reported to be second only to bacterial disease in importance. Fungal infections are generally restricted to chronic losses. The diseases they cause are almost always external in nature, rarely becoming systemic to include internal organs.

It is still widely believed that fungal infection of fishes is largely a secondary development. The hyphae (fungal branches) start their growth on skin or gill lesions that are usually initiated by conditions such as bacterial infections, poor water quality, injuries associated with handling and social interaction, sudden changes in temperature, infestation by parasites and nutritional deficiencies. If these factors weaken the fish or damage its tissue, fungus may infest the fish and can, without treatment, lead to the death of large numbers of fish. However, there is supporting evidence that some fungi may affect healthy fish in certain circumstances. They are commonly known to colonise plant and animal debris in freshwater.

The most commonly identified fungal pathogens of fish are Oömvcetes (commonly called water moulds). Oömvcetes are like fungi. They have the same filamentous, branching, indeterminate bodies and absorb food by excreting digestive enzymes and absorbing the resultant mixture (absorptive nutrition). However, Oömycetes are not considered to be 'true fungi' taxonomically, but have been placed in the phylum Oomycota. Within this phylum is the family Saprolegniaceae, containing among others the genera Achlya, Aphanomyces and Saprolegnia, with some species being pathogens of fish, crustaceans and plants. The Oömycetes are true aquatic organisms, largely saprophytic and are considered ubiquitous in freshwater systems throughout the world. Although little quantitative information is available, their cosmopolitan distribution and ability to colonise a wide variety of substrates suggest a role in the decomposition of organic materials in freshwater ecosystems.

Fungal infections of fish by water moulds are widespread in both wild and captive fish populations. It is known that the members of at least six genera are natural parasites of fish, fish eggs and crustaceans, including *Saprolegnia*, *Achlya*, *Aphanomyces*, *Pythium*, *Leptomitus* and *Allomyces*.

The most widespread species parasitising fish include members of the genera *Saprolegnia* and *Achlya. Saprolegnia* is ubiquitous in freshwater ecosystems and is the main genus of water moulds responsible for significant mycoses of freshwater fish and eggs. *Saprolegnia parasitica* is common on fish eggs and on fish skin. *Saprolegnia ferax* and *Saprolegnia parasitica* most frequently cause death of fish both in aquariums, and their natural environment. *Saprolegnia torulosa* has been encountered on the eggs of freshwater fish species. *Saprolegnia* infects the eggs by adhesion and penetration of the egg membrane, and can spread from dead eggs to live eggs. In warmer climates the role of *Saprolegnia* is largely taken over by species of *Achlya. Achlya* is commonly found on wild-collected rainbowfishes, which have had skin or scale damage during the collecting process.

Saprolegniasis affects all stages in the life cycle from eggs through to adult fish. The fungus produces long filamentous strands called hyphae, which grow on the surface of fish, eggs and organic material. The fungus looks like greyish-white wool-like growths of different sizes. New growths may be difficult to distinguish; the older ones are usually greyishgreen. Microscopically, individual hyphae are evident that are nonseptate and about 20 microns in diameter. Older segments of hyphae often terminate in zoosporangia containing zoospores.

Reproductive motile spores are released from the ends of the hyphae into the water and these quickly find other sites to colonise. The rate of development depends on water temperature and the condition of the fish. Up to 40 or 50% of

the body surface may be covered and the gills, nasal openings and eyes may be infected. The tissue degeneration resulting from the invasion of the fungus disrupts the osmotic balance of the fish. Diseased fish become increasingly lethargic and lose equilibrium shortly before death. Mortalities can range from 10 to 50%. As the fungus radiates away from the focus of the infection, the hyphae penetrate and destroy the layers of skin, and in some cases extend into the muscle. Very severe cases have been reported where the fungus blocked the pharynx of first feeding fry and grew out over the gill lamellae preventing feeding or normal respiratory functioning.

Members of the genus *Pythium* are soil- or water-dwelling organisms. More than 200 species of this genus have been described. They usually live as saprophytes, but several species have been reported to cause disease in plants, fish and crustaceans. *Pythium rostratum* invades the eggs of many freshwater fish species. The genus *Aphanomyces* contains two specialist aquatic animal pathogens, *Aphanomyces astaci* and *Aphanomyces invadens*.

The latter species is associated with the tropical and subtropical fish disease Epizootic Ulcerative Syndrome (Red Spot Disease). In Australia, red-spot disease has been reported in the Northern Territory, New South Wales, Queensland and Western Australia. It begins as a small area of reddening over a single scale, which subsequently spreads to involve a number of adjacent scales; this is the characteristic 'red spot'. As the condition progresses, the 'red-spot' expands and deepens, giving a deep ulcer, which sometimes extends into the abdominal cavity. Some fish, especially Lates calcarifer develop unilateral or bilateral cloudiness of the cornea; these changes in the eye may or may not be accompanied by lesions in the skin. Some cases of red-spot disease heal spontaneously, but many affected fish, especially juveniles, die. Aphanomyces invadens has been identified in rainbowfishes from a number of river systems in the Northern Territory.

*Aphanomyces astaci*, the causative agent of crayfish plague, is an exotic disease to Australia. Few if any susceptible crayfish survive outbreaks of crayfish plague. The fungus produces a motile spore called zoospore. When released, the zoospore can swim for up to two days, seeking another crayfish to infect. If a spore reaches a crayfish, it germinates, penetrates the hard, outer skeleton - the exoskeleton - and forms a new infection.

While precise identification of fungi species from lesions on fish requires a considerable level of familiarity with taxonomy of the aquatic phycomycetes, the detection of a significant level of fungal infection does not require rigorous classification. Once recognised, the growth of aquatic phycomycetes on lesions is difficult to confuse with any other aquatic disease.

### Treatment

Avoidance of exposure to the disease is the primary method of prevention. The most effective strategy for controlling and preventing fungal infections is good fish keeping practices. It is imperative that aquariums are maintained under conditions conducive to good health. Well-nourished fish reared in highly favourable environmental conditions will be resistant to most pathogens. Your veterinarian can make a diagnosis of fungal disease based on microscopic examination. Diagnosis is quick, accurate and generally inexpensive. A biopsy sample is examined using a microscope and the presence of thick (10–25  $\mu$ m), nonseptate, branching hyphae (fungal stalks) confirms fungal infection. A number of other pathogens and saprophytes (opportunistic pathogens) including algae, crustaceans, helminths (worms) and protozoa commonly colonise the fungal tuft.

After evaluating the environment and history of your fish, your veterinarian may or may not decide to implement a treatment protocol. Several chemotherapeutic (drug treatment) options are available. Some lesions are treated topically with a disinfectant like povidone iodine after the water mould and necrotic tissue have been surgically removed. If the infection is not severe, many fish will heal with supportive care (good nutrition and clean water). In severe cases, death is frequently, due to impaired osmoregulation and the fish's inability to maintain fluid balance.

For many years infections have been largely controlled with malachite green. Unfortunately, the potential teratogenic or mutagenic properties of malachite green have resulted in its limited or curtailed use in many countries. The search for alternative chemical treatments or other means of controlling fungal infections has resulted in many investigations being conducted. However, to date, only a limited number of chemical compounds show any potential as fungicides and none are considered as effective as malachite green.

Malachite green is extremely toxic to *Saprolegnia parasitica*, as this species is unable to infest new seeds when treated with as little as 1 mg/L for one minute. This undoubtedly explains why malachite green is very effective in controlling certain fungus infections, many of which are due to this particular pathogen. Malachite green treatments for 24 hours at 5 mg/L are sufficient to control most fungal infections. However, when fish are exposed for longer periods, even to much lesser concentrations, malachite green can be highly toxic to some fish. Malachite green has also been recommended for use concomitantly with another antifungal agent, formalin. However, the chemical evidently is not without its own harmful elements. Some strains of water moulds implicated in fungal infections supposedly are more resistant to malachite green than are others.

Another common dye, methylene blue, is also effective against fungal infections of fishes and may be used as an alternative to malachite green. It is particularly effective against *Saprolegnia* by applying 3 mg/L in a long duration bath. Methylene blue can be used for the treatment of fungal infections on all ages of freshwater fishes at 2 to 3 mg/L in a permanent bath. It is safe for use with fish eggs and fry. Methylene blue has a wide safety margin and is non-toxic when used as recommended. Fish tolerate relatively high dosages without side effects. However, it should not be used in recirculation systems that utilise biological filtration, as it will interfere with the normal biological processes of nitrifying bacteria. It can also interfere with normal plant growth. Also, be aware that this material will stain almost everything with prolonged contact.



Formalin (a solution of 37% formaldehyde) treatments are an alternative to malachite green but are not as effective, and it may have detrimental effects on the fishes it is intended to cure. The dose has to be adjusted according to water pH. Low doses should be used at low pH and higher doses at higher pH values. Oxygen depletion of the water is rapid at high temperatures. However, formalin is for specialist use only. It is inadvisable to use this compound where other treatments are available.

A successful treatment formulated by Dr. Gerard Bassleer, a wellknown fish disease expert, in 1983 consists of the following combined ingredients:

Formaldehyde (37%) = 1 litre [or 100 ml] Malachite Green (oxalate) = 3.7 gm [or 0.37 gm] Methylene Blue = 3.7 gm [or 0.37 gm]

Dosage: 1.0-1.2 ml/100 litres water (25 drops/100 L).

Change the water (50%) after one day treatment and add another dose. This medication can be more toxic in soft acid water, and also at higher temperatures

The use of ordinary salt (or sea water) was among the first of the methods proposed to combat fungal disease. Often, the application of salt either directly onto the diseased part of individual fish, or as a solution in which to bathe the fish. Salt can be used at 10g/L for 20 minutes for young fish and 25g/L for 10 minutes for older fish. A continuous well-aerated salt bath of 2–5g/L may assist in the recovery from fungal infections. However, there appear to be significant differences among species and possibly families as well in the tolerance of the larval and fry stages to salt treatment.

### Worms

Diagnosing worm infestation is not always a simple task for most fishkeepers and is at best, just guesswork. When a fish is eating well yet is still not putting on weight, intestinal worm infestation may be suspected. Particularly when a fish eats regularly yet actually looses weight. This is usually seen as thinning along the back on either side of the dorsal fin. In an extreme case, this may result in a well-fed fish actually starving to death.

Most worms found in aquarium fishes live in the intestine and can readily be removed with various drug treatments. However, others have larval stages that live in lymph ducts and blood vessels, and they can be difficult to treat. Most worms do not pose a serious health risk for rainbowfishes because they often have complicated life cycles in which the fish may serve as only one of possibly several intermediate hosts. Since the necessary intermediate hosts are not usually found in an aquarium, transmission of the worms to the fish does not readily occur. Therefore, worms are more likely to appear in wild-caught specimens or in rainbowfishes that are bred or maintained in outdoor ponds.

However, *Camallanus* is one such species that can be found in aquarium fishes and can cause considerable damage to the host fish. Several species of *Camallanus* have been reported from freshwater aquarium species around the world. The most common

species are *Camallanus cotti* and *Camallanus lacustris*, both of which produce live larvae. Female specimens can attain a length of up to 10 mm, while males usually only grow about 3 mm long. They are commonly found infecting rainbowfishes maintained in outdoor ponds. Even if successfully treated, most rainbowfishes do not recover fully and often die from other complications.

*Camallanus* are ovoviviparous with mature females releasing motile infective larvae. Juvenile worms are vented with the fish's faeces and ingested by other fish. This can result in heavy infestations in some situations. Camallanus infection will initially be noticed as red-coloured worms protruding from the anus of the fish, most noticeable after feeding. Other clinical signs include wasting (anorexia), abdominal swelling, and extensive lesions in the rectum mucosa. The worms attach to the wall of the fish's intestines with their tiny jaws and suck its blood. This can cause considerably damage to the intestinal lining, and a reduced capacity of the fish's ability to readily absorb nutrients.

Most *Camallanus* species reproduce by means of an intermediate host, which can be small crustaceans such as copepods or various insect larvae. However, the species' relatively frequent and persistent occurrence in aquaria worldwide strongly indicates flexibility in its life cycle, i.e. the ability to infect the final host directly. Research has shown that under aquarium conditions, without any presence of an intermediate host, *Camallanus cotti* is able to infect various fish species directly for at least three generations. It was further shown that the infective free-living firststage larvae may survive for more than three weeks in the aquarium environment and that their host-attracting behaviour is not precluding direct transmission to the final fish host. Therefore, any treatment for Camallanus worms should be directed towards both individual infected fish hosts as well as the free-living larvae on the substrate.

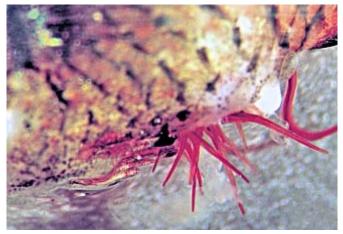
### Treatment

The simplest solution is to never get them in the first place by eliminating all possible sources of infection. Take care when feeding your fish any wild-collected live food, sterilise anything that you put in your aquaria (if possible), and be very careful from where and whom you obtain your fish. Also, make sure you quarantine any new fishes for 30 days before you put them in your regular aquaria. If they are infected, you should eventually see the red worms protruding from the fish's anus.

There are numerous recommended chemical treatments for internal worms. Among them are Piperazine, Levamisole, Fenbendazole, and some specific aquarium medications. However, mixed results have been reported using these drug treatments and it is sometimes difficult to remove *Camallanus spp*. from infected aquaria. Some success has been reported using the following treatment regimes:

Fenbendazole has been used to control intestinal worms in fish. A dosage of 25 mg/kg, delivered in food for 3-5 days, has been commonly recommended, but this regimen has not been evaluated in controlled trials. Flubendazole 5% mixed with fish food at 100 mg/100 gm of food. Feed every second day for 5 days. On those days feed only once with the regular diet. Treatment should be repeated after 21 days.





Camallanus worms protruding from the anus of a fish **A** 

It is suggested to enhance the food with cod-liver oil and bind it with gelatine or agar, since fish are quick to refuse food, which has been medicated. Withholding food for a few days prior to feeding medicated food may aid in acceptance. As a bath, it can be used at 2 mg/L of water once a week for three weeks.

Mebendazole is chemically related to flubendazole and fenbendazole. It is a benzimidazole derivative, and is a useful broad spectrum anthelmintic; the drug of choice for mixed worm infestations. Some sources suggested that water soluble ivermectin might overtake mebendazole as the drug of choice for mixed worm infestations.

Ivermectin 1% - Some studies have shown that ivermectin added directly to aquarium water has been useful in treating *Camallanus spp.* in fish. The dose used was 0.7 millilitres of a 1% injectable solution per 76 litres of water. The dose was added over a period of four days (0.1, 0.2, 0.2, and 0.2 millilitres). A solution of 1 part ivermectin 1% in 19 parts distilled water can be made and administered as a split dose of 2 ml on day one, 3 ml on day two, and 3 ml on day three followed by a water change on day four. However, because this drug has a narrow margin of safety, some veterinarians advise against any use of ivermectin for aquarium fishes.

Concurat L (10% Levamisole) mixed with food at 1 gm/100 gm of food. Feed once daily over 5 days. If the fish are already weakened and refuse to eat then you can try 30 mg/L of water dissolved in a little water beforehand. After 3 days do a 50% water change and dose again. After another 3 days repeat the waterchange. Levamisole is a widely used drug to treat nematode infections in fish and has been reported to kill both the infective free-living larvae and the fish parasitic stages of *Camallanus cotti*.

Praziquantel at 2-10 mg/L for 1 to 3 hours in a bath is effective in treating adult worm infections in fish. Praziquantel has been identified as the most effective "in water" treatment of infected fish. Praziquantel can also be administered in food at 35-125 mg/kg for up to 3 days. Praziquantel was traditionally available in the form of branded Droncit® tablets, for oral administration in dogs and cats, but is now available in a range of aquarium products. Praziquantel is usually harmless to fish of all species, is non toxic to plants, and has no negative impact on nitrifying bacteria. However, there are reports that Kissing Gouramis (*Helostoma temminckii*) have been adversely affected by Praziquantel (98.5%).

# Hydra

Hydra are small aquatic invertebrates commonly found in most unpolluted freshwater environments in both tropical and temperate regions of the world. They belong to the same group of animals as the marine jellyfishes, corals, and sea anemones. Hydra are commonly found hanging inconspicuously from drifting and stationary vegetation or from the waters surface film in most freshwater habitats. They are often unintentionally introduced to the aquarium with live foods, snails, driftwood, plants, gravel and water collected from natural freshwater environments.

Hydra are about 1 to 25 mm long and have a flexible cylindrical body secured by a simple adhesive basal disc by which the animal anchors itself to a surface. Gland cells in the basal disc secrete a sticky fluid that allows for its adhesive properties. On the opposite end are from five to twelve arms or tentacles, gently swaying in the water current. If the hydra is disturbed the tentacles can be retracted to small buds and the body column itself can be retracted to a small gelatinous sphere, making them almost impossible to see. They are generally sedentary or sessile, but do occasionally move quite readily. They do this by bending over and attaching themselves to the substrate with mouth and tentacles and then release the foot, which provides the usual attachment. The body then bends over and makes a new place of attachment with the foot. By this process of "somersaulting", hydra can move several centimetres in a day. Hydra may also move by amoeboid motion of their bases, or by simply detaching from the substrate and floating away in the current. They float at the surface of the water buoyed up by gas bubbles given out by the basal disc. They remain in a free-floating stage until they find a spot that is suitable for their needs.

Hydra are strictly carnivorous, feeding mainly on small crustaceans, insect larvae, worms or other tiny aquatic animals. In aquaria, they will thrive on any similar live food being fed to rainbowfish larvae - including the rainbowfish larvae. Any aquarists who feed copious amounts of live brine shrimp nauplii or similar live foods will eventually find colonies of hydra in their aquarium. To capture these minute forms of life hydra extend their body to maximum length and then slowly extend their tentacles. Despite their simple construction, the tentacles are extraordinarily extensible and can be four to five times the length of the body, sometimes extending to a length of 5 cm. Once fully extended, the tentacles are slowly swept around waiting for contact with a suitable prey animal. Each tentacle, or cnida (plural: cnidae), is embedded with highly specialised stinging cells called enidocytes. Cnidocytes contain specialised structures called nematocysts which look like miniature light bulbs with a coiled thread inside. At the narrow outer edge of the cnidocyte is a short trigger hair. Upon contact with prey, the contents of the nematocyst are explosively discharged; firing a dart-like thread containing neurotoxins into whatever triggered the release. Once stimulated, the nematocysts take only three milliseconds to release fully.

Judging from its incredible potency and speed of paralysis and death, the poison is believed to be a nerve toxin. They have



four different types of nematocysts, some stinging and paralysing the prey, whilst others discharge threads which coil round the prey and hold it. Small animals paralysed by the stinging cells are brought to the digestive centre by the tentacles and devoured. The mouth of the hydra is located at the centre of the tentacles. The body is hollow and the inner layer of cells digests the food by engulfing it. With prey almost as large as itself, the hydra stretches so thin in getting its digestive gut around it that the prey animal appears to be covered with a thin transparent film; only the tentacles tell you that you are looking at a hydra, although they generally keep their tentacles coiled up. A hydra that has recently fed will be shorter and more rounded than one that is hungry.

In aquaria, hydra can be found attached to plants, gravel, stones, the sides of the aquarium, filter equipment, etc. They are most commonly found attached to the stems of waterplants or the undersides of floating leaves or plants such as duckweed. Unless they occur in large colonies or are coloured, most hydra in a normal aquarium will go unnoticed. However, within the confines of a small fry-raising tank, these little monsters can be deadly, and can ingest a large batch of newly hatched rainbowfish larvae in less than a week. Hydra can kill larvae up to a size of around 10 to 15 mm. Newly hatched larvae of rainbowfishes and blue-eyes are bite-size for the average hydra. Larger larvae may often pull away from the stinging tentacles, but will usually die in any case. Juveniles over 15 mm, however, generally don't seem to have any problems.

Hydra only seems to appear in fry-raising tanks being fed brine shrimp nauplii or similar live foods. They don't seem to appear in fry-raising tanks that are fed primarily a dry or liquid diet. However, once you start feeding large amounts of brine shrimp nauplii, it isn't long before hydra appear. Hydras have however, been known to feed on the organic material off the substrate when live food supply is insufficient. This behaviour however, is not considered normal. Usually you do not notice them unless a heavy infestation has become established. They are usually brown in colour or transparent and are not readily seen against a background of natural coloured gravel or on plants. The best way to avoid introducing them into fry-growing tanks is to sterilise the tank and filter equipment with a chlorine solution before they are used.

Finding a few hydra in your regular aquarium doesn't mean that the aquarium is unhealthy. Actually, it means you have a normal healthy aquarium, since hydra will not live in water of poor quality.

The name "Hydra" is derived from a monster in Greek mythology. One of the twelve tasks imposed on Hercules was to slay the Hydra of Lerna. It had nine serpent-like heads, one of which was immortal. So potent was its venom that even its smell was fatal to those who passed too close. Worst of all was that for every head Hercules cut off, two more grew in its place. He finally triumphed by cauterising the wounds with a firebrand as he chopped away. Just like the legendary monster, a single hydra may be cut into many pieces and if each piece contains a portion of the two body layers, ectoderm, and endoderm, it will develop into a complete animal. A number of different species of hydra are known, and they come in a variety of different colours such as green, grey, black, brown, pink, orange, and transparent.

*Hydra viridissima* (formally *Chlorohydra viridissima*) is referred to as the green hydra due to the presence of a symbiotic green alga (*Chlorella vulgaris*) in the gastrodermal cells of the animal. The algae make use of those substances that the hydra would normally excrete, such as carbon dioxide, phosphates and nitrogenous substances in this symbiotic relationship. For its part, hydra benefits by receiving the oxygen produced by photosynthesis. If there is a food shortage, the hydra will digest the plant cells and later ingest more living algae, which it does not digest. Due to the presence of these symbiotic algae, this hydra is attracted by light, contrary to other species and can survive long periods without food. Their distribution is considered ubiquitous throughout Australian freshwater systems.

Hydra are considered to be hermaphrodites and reproduces both asexually and sexually. Asexual reproduction takes place in a process known as budding; the hydra simply forms a bud on the side of its body. Buds are produced every two to three days under favourable conditions. Hydra often has four buds at a time. These simple organs are no more than lumps attached to the body of the female hydra. The embryo secretes around itself a hard, sticky shell and is carried by the female until the young hydra develop tentacles, and then breaks off, fully prepared to exist in its own. All hydra species have been observed to go through occasional sexual phases. One thing that can cause sexual reproduction is when environmental conditions become unsuitable. They produce eggs which form on the external surface of the stalk. The fertilised eggs secrete a tough outer coating and, as the adult dies, these resting eggs fall to the bottom of the lake or stream to await better conditions, whereupon they hatch into miniature adults.

The growth rate of hydra populations is closely related to conditions such as temperature, water quality and quantity of food. Habetha *et al.* (2003) analysed the population growth of *Hydra viridissima* fed *Artemia salina*. They observed that the population fed daily doubled in 3 days; those fed once a week, in 14 days, and those fed once a fortnight, in 31 days.

Freshwater hydra as a whole, are relatively little known in Australia in spite of being common organisms in most freshwater habitats. In Australia there are at least 4 species: *Hydra attenuata, Hydra hexactinella, Hydra oligoactis* and *Hydra viridissima*.

### Treatment

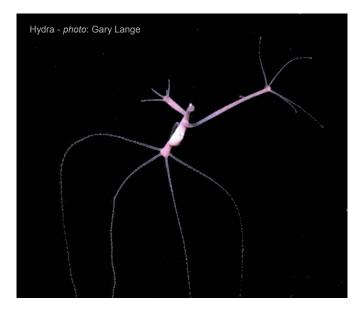
In the past, the only way I found to eradicate hydra successfully was to sterilise the tank with a chlorine solution. Some copper medications claim that they will kill hydra but they can also be deadly to the fry. Other treatments usually recommended include ammonium nitrate, quinine sulphate, raising the temperature to 40°C for 15 minutes or more, lowering the *p*H, or adding salt until the hydra are killed. An even more elaborate treatment I read once was the use of a 9V battery connected to leads that fed into the aquarium water. Most of these treatments simply do not work and in addition, they can be harmful to the young rainbowfish fry.



Dactycid<sup>®</sup> is a very effective drug treatment against hydra and all species of internal worms (hookworms and roundworms) including Camallanus cotti, flukes and planarians. Use according to manufacturer recommendations. Flubendazole is another veterinary drug and can be found under a number of synonyms: Fluvermal; Flubenol; Flumoxal and Flumoxane. Flubendazole 5% is very effective in killing hydra and is very safe to use with small rainbowfish fry used at 1-2 mg/L. I have used both these products with rainbowfish fry only 10 days old, without any problems. Pre-dissolve the flubendazole 5% in a container with some aquarium water, and then pour it in the aquarium. After 5-7 days, following treatment, do a 50% waterchange. Flubendazole is a "wettable powder" and as such does not completely dissolve. It may leave a white residue in the aquarium, but this can easily be removed with following waterchanges. Remove any deposit of white powder left on the aquarium walls with a sponge and siphoning the bottom with a gravel siphon. I did not have any problems with the residue and it also had no effects on brine shrimp nauplii when fed to the fry. However, these chemicals can be very toxic to snails.

Flubendazole (15%) is specifically designed and developed for aquatic use. This medication will kill flukes, camallanus worms, tapeworms, anchor worms and other helminthics commonly found in tropical and coldwater fish. It has also been used successfully with 'skinny disease' in clown loaches. However, it can be toxic to snails and is harmful to scaleless fish species. One should follow the manufacturer's instructions for treatment, as different manufacturers use different chemicals and concentrations of the active ingredients.

Flubendazole may not be available in Australia; I obtained the chemical directly from Europe. However, a number of other products have been reported to have similar effects on hydra and other worms. Panacur (fenbendazole) a similar chemical, has also been reported as an effective control for hydra used at 2 mg/L. This chemical is available in various formulations and trade names. Praziquantel is another chemical that is very effective for control of hydra used at 1–2 mg/L, while having a wide margin of safety for fish. It is available as an aquarium formulation under a number of brand names. However, before using any of these products consult a veterinarian.



# Planaria

Planaria are a free-living flatworm found in almost every kind of environment, on land and in fresh and salt water. They are a very small black or brown flatworm that look very similar to leeches and often appear in freshwater aquariums. They are generally around 3 to 5 mm long, but some grow as large as 10 mm. In a normal aquarium situation they usually don't cause any problems and probably even go unnoticed. However, if you get them in a breeding aquarium, they can destroy a whole spawning of eggs within hours. They are usually seen crawling around on the front of the aquarium after the lights have been turned off. They feed on anything organic but can also infect the mucous membranes of the fish's gills.

Planaria belong to the Phylum Platyhelminthes, Class Turbellaria, and Order Tricladida. The genus Planaria, has apparently has been replaced by an older name, Dugesia. Planaria come in a number of species and strains, some of which are albino. The following species have been reported from freshwaters, Dugesia tigrina, Dugesia dorotocephala, and Dugesia tigra. Dugesia notogaea has been recorded from northern Queensland.

They are carnivores and feed by sucking food through a tube called the pharynx located on the mid-ventral surface that opens into a blind gut, or gastrovascular cavity. Excretion is via protonephridia. Planarians have two protonephridia composed of branched tubules that empty wastes through excretory pores on their surface. The protonephridia contain numerous bulblike flame cells with clustered, beating cilia that propel fluid into the tubules. These structures function in waste excretion and osmotic regulation. The body is cylindrical in the small species, dorsoventrally flattened in the large ones. The epidermis is ciliated. There is no blood vascular system and transport is via diffusion, a mechanism made possible by the small body size and/or flattening. Food is delivered to the tissues by the gastrovascular cavity. Oxygen diffuses across the body surface to the tissues, none of which are more than 1 mm from it. The nervous system consists of a brain and longitudinal nerve cords. There may be light-sensitive ocelli in various positions on the body.

Planaria typically reproduces asexually by architomy. This is a type of fission is which the worm divides into two fragments without any prior differentiation of new parts. A transverse cleavage just posterior to the pharynx divides the worm into an anterior, nearly normal, worm with head, mouth, pharynxm and most of the gut, and an incomplete, headless posterior mass of tissues, which must produce the missing parts. The posterior portion remains immobile until it replaces most of the missing structures whereas the anterior end behaves normally and moves about.

Planaria are also able to reproduce by regeneration, producing an entire new worm from a piece that has been cut off. If the head is split, both halves will grow back the other half. Movement may be accomplished either by muscular contractions of the body wall or the use of cilia along the ventral surface. When using the cilia the planarian appears to be gliding across the surface.





Spawning mops are commonly used by hobbyists for breeding many rainbowfish species. However, before using them in different breeding tanks, it is advisable to always sterilise the mops to kill any planaria, or other pests that may be present. This can easily be accomplished by washing them in boiling water. Another method is to soak them in a chlorine solution, then thoroughly rinsing them before being sun-dried. This is where spawning mops offer a significant advantage over live plants as a spawning medium.

#### Treatment

Flubendazole is a veterinary drug and can be found under a number of synonyms: Fluvermal; Flubenol; Flumoxal and Flumoxane. Flubendazole 5% is very effective in killing planaria and is very safe to use with small rainbowfish fry used at 1–2 mg/L. I have used both these products with rainbowfish fry only 10 days old, without any problems. Pre-dissolve the flubendazole 5% in a container with some aquarium water, and then pour it in the aquarium. After 5–7 days, following treatment, do a 50% waterchange.

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If you are unable to obtain Flubendazole there are a number of other products that have been reported to have similar effects on planaria. Panacur (fenbendazole) has also been reported as an effective control for planaria used at 2 mg/L. This chemical is available in various formulations and trade names. Praziquantel is another chemical that is very effective for control of planaria used at 1–2 mg/L, while having a wide margin of safety for fish. It is available as an aquarium formulation under a number of brand names. However, before using any of these products consult a veterinarian.

A proprietary product called Dactycid<sup>®</sup> is also very effective, use according to manufacturer recommendations.



There are several diseases which can affect rainbowfishes. Opportunistic bacteria and parasites can cause dermal and systemic infections. Intestinal nematodes can cause chronic wasting (anorexia) and considerably damage to the intestinal lining. Additionally, water moulds and fungi may also present disease problems in poorly managed aquariums. However, one of the most common and problematic diseases of captive rainbowfishes is mycobacteriosis.

The increasing popularity of rainbowfishes has resulted in a significant increase in the number of commercial operators breeding, rearing and distributing rainbowfishes. This increases the potential for dissemination and exacerbation of infectious diseases, such as mycobacteriosis. Rainbowfishes are a highly susceptible species based on numerous reports and anecdotal observations. Perhaps rainbowfishes differ from other aquarium fishes in their immunological response to mycobacterial organisms.

Two terms are used to describe the disease, either "piscine tuberculosis" or "mycobacteriosis". Mycobacteriosis is usually a sub-acute to chronic disease of fish where the etiologic agent is an acid-fast bacillus in the genus Mycobacterium. Chronic proliferative mycobacteriosis is characterised by the formation of granulomas, while subacute and acute forms of the disease are associated with necrosis and acid-fast bacilli scattered diffusely among the kidney, liver, spleen, and often all visceral organs. Under pathology examination, mycobacteria have been found in apparently healthy rainbowfishes. Often no external signs are present until advanced stages of the disease occur, at which time non-specific signs present including emaciation, hemorrhagic and dermal lesions, lethargy, and death. Many Mycobacterium species are ubiquitous in the aquarium hobby and trade, making control by avoidance of these pathogens very difficult. Furthermore, there is currently no effective treatment for mycobacteriosis in rainbowfishes.

### Source of Infection

It is generally believed that infected fishes are the main source and reservoir of mycobacteria in aquaria. The potential for infection among aquarium fish depends on the contact rate among infectious fish. Under aquarium conditions, the frequency of that contact is vastly increased. Mycobacteriosis can also be acquired through the ingestion of mycobacteria present in the aquarium environment, which usually have their origin in detritus derived from dermal lesions, faecal material or exudates etc., shed by diseased animals that contain mycobacteria. The sources and modes of transmission in fish may be related to the infection of invertebrates, such as freshwater snails, daphnia and shrimp. The entry of mycobacteria through skin and gill lesions caused by injury or parasitic infection should also be considered. After the organisms enter the body, they may cause skin lesions or spread to other organs through the circulatory or lymphatic system.

It is suspected that ovarian transmission from parent to offspring may occur. A report from an Australian fish hatchery in 1977 provided evidence of mycobacteriosis transmission from eggs to the F1 generation. This observation does not confirm that ovarian transmission takes place, as the egg surface may have been contaminated by peritoneal fluid containing mycobacteria. However, research in 1994 confirmed the transmission of mycobacteria in Siamese fighting fish (*Betta splendens*), via transovarian passage. Acidfast bacteria were found in the ova of diseased female Siamese fighting fish, using the fluorochrome technique. Transovarian transmission has also been reported in *Xiphophorus maculatus*. The observation of mycobacteria in the fish's eggs and tubercle granulomas in the ovary wall suggests that transovarian transmission is a definite possibility.

Mycobacteriosis disease outbreak in aquarium fish is often reported to be related to management factors. However, even the healthiest aquarium can harbour the bacteria. A variety of bacterial pathogens are always present in an aquarium, even if the system is maintained in optimal condition. Most of them are ubiquitous in aquatic environments and the non-expression of their virulence could be ascribed to a good management of the system and to a good physiological status of the fish. Moreover, the presence of bacteria described as producers of inhibitory compounds, suggests that the indigenous microbiota can control pathogenic organisms in aquarium systems.

Although there is no firm evidence to confirm that environmental stress can cause mycobacteriosis infection, it has been suggested that an unnatural environment, such as an aquarium, may actually promote the disease. Fish should be maintained under optimal conditions. Inappropriate aquarium management can result in abnormal stress and a reduction in the normal resistance of the host. Overcrowding, accumulation of waste and organic matter in the water and increasing water temperature (above 28°C) may all be predisposing factors. Beyond overcrowding and confinement, aquarium fish are also subjected to other stressors such as handling, fluctuating temperatures, poor water quality, and social stresses. High numbers of mycobacteria have been correlated with warmer temperatures, low dissolved oxygen and pH. Such factors exacerbate the susceptibility of fish to disease and thus further increase morbidity and mortality in the population. Attention to water quality and good nutrition will assist the fish in fighting these chronic infections.

Poor nutritional health can greatly enhance the progression and severity, and reactivation of disease. Once present in an aquarium, infection rates can vary from 10 to 100%. The severity of the disease is influenced by a number of interrelated factors, including bacterial virulence, the kind and degree of stress exerted on the population of fish, the physiologic condition of the host, and the degree of resistance inherent within specific populations of fishes.



### Prevention

Rainbowfishes should be obtained from specific pathogen-free sources and quarantined when received. In addition, knowledge of the origin and aquarium practices of your source can help you prevent future potential problems. Breeders should maintain separate young brood fish populations and avoid using older brood fish. Breeders who breed from wild stock don't generally have a problem.

A better understanding of the pathogenesis of mycobacteriosis, including factors affecting host susceptibility, may enable aquarists to manage this pathogen and prevent potential disease outbreaks through effective aquarium practices. The implementation of preventive measures in controlling chronic mycobacteriosis is particularly relevant, due to difficulties in treatment, and different fish species probably have different levels of sensitivity.

Clearly, prevention and appropriate routine disinfection should be viewed as the primary means to control mycobacteria in aquarium systems. The chronic nature of mycobacteriosis means that it is often too late for any remedial action to be taken once the first cases have been observed and diagnosed. The same protocol can be used in quarantine systems, at least on a periodic basis, to prevent potential concentration of mycobacteria. Although fish should be quarantined for at least 4–8 weeks before being placed in their main aquarium, most fish become clinically affected after a longer period of time. Direct lethal sampling of a quarantined population, with histopathology and culture, may be necessary to detect subclinical infections.

Ideally all equipment such as nets, hoses, buckets, etc. that comes into contact with stock (diseased or healthy) should be immersed into a strong biocide regularly (such as hypochlorite or iodophores), ideally after each use, to achieve sterilisation. Quaternary ammonium compounds can also be used. All this products however, must be rinsed adequately prior to reuse because all these compounds are toxic to fish.

### **Clinical Signs**

Early signs of mycobacteriosis may be subtle or unapparent, and visual clinical signs often do not develop until the disease has become widely systemic. Clinical signs of mycobacteriosis are not specific to the disease and often resemble other diseases. They can vary in occurrence and severity and infected fish may manifest few or no external signs of disease. Clinical signs can vary between fish species and the species of mycobacteria can also influence the clinical symptoms observed.

Mycobacteriosis is generally a chronic, slowly progressive disease. The acute form of the disease occurs rarely. It is characterised by rapid morbidity and mortality with few clinical signs. The chronic form of the disease is most commonly seen and it may take months to years for the number of organisms to grow to readily detectable numbers. There is ample evidence that these organisms are capable of adapting to prolonged periods of dormancy in tissues, and that this dormancy is responsible for the latency of disease. Chronic mycobacteriosis infections manifest themselves primarily as swollen white patches or lumps on the body that turn into red or pale lesions. Fish with only skin infections may have several types of concealed lesions. Both the dermis and epidermis are eroded and the underlying musculature becomes severely necrotic. At this stage, the infection has usually become systemic and the infection on the surface of the skin may occur throughout the peritoneum and musculature. Internally the liver, kidney, and spleen may be impaired.

Because of the slow progression of the disease, younger fish infected with mycobacteriosis show no external signs. As fish age or are stressed, the infection becomes more serious. Nevertheless, mycobacterial lesions have been observed in rainbowfishes as young as three months old. It is difficult to specify the length of incubation (the time from infection to the appearance of the first signs of the disease). The incubation period varies greatly and depends on susceptibility, temperature, and severity of exposure. With rainbowfishes kept in substandard conditions and at higher temperatures, it may last only a few weeks or months.

If clinical signs develop, emaciation, cachexia (wasting, loss of weight), exophthalmia (pop-eye), ascites (dropsy), skeletal deformities (curvature of the spine), haemorrhagic and dermal ulcerative lesions or loss of scales may be observed. Other signs of infection can be seen in the gills, which are paler than normal and show thickened areas on some filaments. Small lesions may be observed around the mouth and vent. Changes in cutaneous pigmentation include a fading of normal colour in aquarium fish or change in colouration. Affected fish generally exhibit lethargic behaviour, isolation, abnormal swimming behaviour, floating impassively on the surface of the water, with concurrent loss of appetite. Poor growth, panophthalmitis and retarded sexual maturation may also occur.

Affected fish populations may show chronic low-level mortality, and increased susceptibility to parasitic infection. Any group of rainbowfishes showing chronic, low-level mortality and spawning difficulties, regardless of whether they show external signs of the disease should be submitted to a fish-health laboratory for investigation. Mycobacteriosis probably predisposed the fish to other pathogens commonly found in the aquarium fishes.

### Diagnosis

Unfortunately, there is no non-lethal method available to identify infected individuals, especially those in early to mid stages of disease. Slow mycobacterial growth rates contribute to the late onset and chronic effects of mycobacteriosis. By the time clinical signs and low-level mortality are observed, the disease may already be entrenched in the aquarium population. Methods for detection of infected individuals have yet to be developed. The techniques for diagnosing mycobacteriosis in fish are continually evolving, but clinical signs and gross pathology may give an initial indication of infection with mycobacterial species. Most cases of mycobacteriosis are not identified or more often, are simply misdiagnosed. It is therefore recommended that infected fish be submitted to a laboratory for identification.

#### Hobbyists Diagnose

The most common bacterial infections in aquarium fish are caused by organisms such as *Aeromonas*, *Pseudomonas*, *Mycobacterium* and *Flavobacterium*. Aeromonas has been found to be the most common. All of them cause opportunistic skin infections often caused by injury or parasitic infection. Mortality increases significantly once bacteria enter the circulatory system. *Aeromonas*, *Pseudomonas* and *Flavobacterium* generally have short incubation period and rapid progression of infection. Clinical signs are generally reached within one week of the initial infection of the disease.

On the other hand, mycobacteriosis is a chronic disease and it may take a long time for infected fish to show any clinical signs. *Aeromonas* infections can cause 100% mortality amongst fish in 21 days. The average mortality rate of *Pseudomonas* can be as high as 50% during the first 21 days with continued mortality for another 7–14 days. Within 36 hours of infection with *Flavobacterium*, fish will show areas of greyish discoloration. Once established, the infection can spread quickly and cause high mortality rates. In contrast, mycobacteriosis infected fish populations generally show low-level mortality. Therefore, if you have an infected rainbowfish with a lesion that has not changed that much for more than 21 days, then I would suggest that in all probability it will be a case of mycobacteriosis.

### Treatment

Control of mycobacteriosis in aquarium systems is extremely difficult once an infection has occurred. Unlike most other bacterial fish diseases, there is no cure for mycobacteriosis and it will progress despite your best efforts, resulting in chronic health problems and eventually, mortality in the whole population.

Currently there are no satisfactory treatments for mycobacteriosis and infections of aquarium fish should be considered non-treatable. There are several reasons why systemic mycobacteriosis should not be treated. There is a lack of information on the bioavailability of most chemotherapeutic agents in aquarium fishes, as are successful well-documented clinical trials. In fact, no chemotherapeutic agent is approved for the treatment of mycobacteriosis in aquariums or aquaculture. Efforts to eliminate infection in affected populations with antibiotics have not been successful as mycobacteria are mostly resistant to conventional antibiotics. Finally, mycobacteriosis has zoonotic potential.

In most situations, the customary treatment for infected fish or populations is euthanasia of the entire stock (especially in breeding facilities), and the disinfection of the aquarium before restocking. If several fish become infected in the same aquarium, it is usually assumed that the others are carriers and that they be treated accordingly. Fish that have survived an epizootic disease and have recovered may be latent carriers, posing a significant risk to the entire population. Following depopulation, the entire system, especially the filters and substrate, must be thoroughly disinfected with a biocide. In addition, all equipment that has been in contact with the infected fish should be disinfected. Gloves should be worn when handling infected fish or cleaning contaminated tanks or other equipment. Hands should be washed thoroughly afterwards with 70% isopropyl alcohol and a bactericidal soap. Break down the original infected aquarium and any other tank use as a treatment or quarantine tank and disinfect them with a strong chlorine solution. Use Calcium hypochlorite 65% to disinfect any tanks, which are in the vicinity of others housing live fish. Granular chlorine does not volatilise as readily as liquid chlorine (Sodium hypochlorite). In a poorly ventilated fishroom, fumes from liquid chlorine can cause fish kills in adjacent tanks. Concentrations of 200–1000 mg/L available chlorine for 60 minutes should be effective for disinfections of tanks, substrate, and submersed equipment (keep filters running during treatment).

Always use chlorine with caution as repeated use and extended exposure of the silicon sealant to strong chlorine solutions will destroy or render the adhesive bond ineffective on glass aquariums with disastrous results. Chlorine will dissolve synthetic material like sponge filters, but most plastics are unaffected. Calcium hypochlorite is an oxidising agent and should not be exposed to intense heat, acids, or organic compounds because it is a fire hazard, particularly if wet. In some cases, explosion may occur. Always wear eye protection and rubber gloves when handling large quantities of chlorine. Chlorine can be neutralised by adding Sodium thiosulfate to the solution (7.5 grams of Sodium thiosulfate will neutralise the chlorine present in 5 litres of a solution of 200 mg/L).

However, disinfection is not always successful due in large part to the resistance of many species of mycobacteria to common disinfectants. Mycobacteria are resistant to many commonly used bactericidal agents at standard dosage rates, including chlorine and quaternary ammonium compounds. Mycobacteria can be highly resistant to chlorine disinfection. As much as 10,000 mg/L available chlorine has been reported necessary to kill some species of mycobacteria. Bacterial biofilm in an aquarium can harbour the organism even after aquariums and equipment are disinfected; indeed, biofilm bacteria appear to be more resistant to disinfection than free organisms.

Veterinarians at the National Aquarium in Baltimore, USA recommend using chlorine to clean the tank and substrate, etc., and then spray 65–90% isopropyl alcohol onto the glass, and allow it to dry. They recommend the alcohol as they found that chlorine does not kill all mycobacteria. They use chlorine to remove/oxidise organic material to assure the alcohol contacts all mycobacteria in/on the tank. Remove all residues of disinfectant from the aquarium before reuse. (Denise Petty DVM, *pers. comm.* 1998).

### **Personal Experience**

#### Case # 1:

In early 1997, I transferred some 4-year-old Goyder River rainbowfishes from their present aquarium to a larger 600-litre aquarium. I had raised 30 individuals and decided to split them in half. The 600-litre aquarium contained a mixture of fullygrown rainbowfishes. Most of them were more than 4 year old with some specimens as old as 9 years. Over the ensuing weeks the Goyder's, one by one, stopped feeding and started to 'hang' just below the surface of the water. Apart from laboured breathing, and looking as though they just had a very large meal, no other symptoms were apparent - death followed soon



after. When only 3 individuals were left from the original 15 that had been transferred, I decided that I needed some confirmation of their disorder. I took the remaining three fish to veterinarian, Dr. Stephen Pyecroft BVSc of Aquatic Diagnostic Services International Pty Ltd for examination.

Stephen's diagnose showed that "Disseminated caseating pyogranulomatous inflammation possibly due to Mycobacterium infection" and "Hepatic Lipidosis" (fatty liver disease). He commented, "The severity and chronicity of the pyogranulomatous inflammation suggests this is the primary disease process. Special stains have shown the presence of acid-fast bacilli consistent with *Mycobacterium spp*. These organisms were found in the macrophages in the liver and kidney. The hepatic lipidosis is quite severe and could well be associated with hepatoencephalopathy although histological evidence of this was not detected in the brain sections examined. The lipidosis was found in all those examined."

"The fish I concentrated on for the histopathological examination definitely showed the greatest degree of pathology and because of the diagnose of mycobacteriosis we must suspect that the total clinical picture observed is due to this problem. There is no ignoring the fact that these fish on the whole were over weight and that the hepatic lipidosis present would have eventually caused their demise had the TB not caused their final problems. The picture is still not that clear and I personally believe that the nutritional imbalance leading to the lipidosis is the major management factor that will need to be addressed. However the fact that a mycobacterial infection is present must, in these fish, be accepted as the primary cause of disease."

What that means in layman terms is that the fish were overweight and infected with mycobacteria. My conclusion from all this was that the 600-litre aquarium was the culprit and knew somewhere down the track that I would have to destroy all the fish and sterilise the tank with chlorine. This belief was confirmed as I continued to have disease outbreaks in this aquarium with some fish displaying similar symptoms while others also developed external lesions. This aquarium was treated with a strong chlorine treatment and all fish and plants destroyed. It is interesting to note that the remaining 15 fish, two years later and 6-years old, in the original aquarium were still doing well, albeit on a somewhat reduced and modified diet, and showed no external signs of the disease whatever.

#### Case # 2:

About 8 months after the above episode I presented Stephen Pyecroft with six young (1-year-old) specimens of *Melanotaenia oktediensis*. All six specimens had what I refer to as "Blackhead Disease" in varying degrees. This disease exhibits itself as a black darkening of one side of the head only. Two of specimens also had small skin eruptions on one side of the body and one also showed the darkening skin colouration along one side of the posterior portion of its body. The most severely affected fish would swim with their head up and tail down and showed an increased respiratory rate. This disease (blackhead) seems to be common among rainbowfishes as I have seen it often and many other hobbyists have spoken to me about this problem. It also seems to be particularly prevalent among Goyder River rainbowfish. Stephen found that most of the fish had enlarged kidneys, which had a granulated pale colour and protruded beyond their normal position. Granulomas were also present in the spleens and around abdominal organs. Acid fast (Ziehl-Nielsen) stains were preformed on impression smears from most of the affected organs and the presence of acid fast bacteria was confirmed. The Diagnose: Disseminated granulomous inflammation - nephritis, hepatitis, and peritonitis.

Stephen comments were "As we have discussed before, the dark areas on the skin are most likely due to a malfunctioning in either the pigment cells or the nerves that control the pigment cells in that area of the skin. The findings of a generalised infection with *Mycobacterium* species would be suggestive that the localisation of the dark pigmentation is due to the formation of local abscesses, which are then causing the expression of the major clinical sign. Most of these cases of "blackhead syndrome" in rainbowfish that I have investigated have had a primary infection with mycobacteria. There may be other primary causes of this distinct clinical sign but in these fish it was piscine TB."

### Zoonosis

Mycobacteriosis is different from most other fish diseases that you are likely to experience in your aquarium. This is because mycobacteria are capable of causing a wide range of dissimilar symptoms in infected fish and its ability to cause disease in humans. Human infections caused by mycobacteria transmitted from fish or the aquatic environment is quite common.

Mycobacteria have a well-documented zoonotic history. In 1951 it was found that this bacterium was able to infect people who frequented swimming pools. For this reason, the skin infection was termed swimming pool granuloma. Since then, several authors have noted the association of the skin infection with aquariums and tropical fish and today is generally referred to as "fish keeper's disease".

Although several hundred cases have been reported in the scientific literature, transmission from fish to human is rare. In most cases the infected individual has been in contact with high numbers of mycobacteria, with a break in the skin at the site of original infection. The most commonly infected sites involved are the fingers and hands. Allergic dermatopathies have also been reported on the skin of aquarists handling water in which affected fish have been reared. Anyone who suspects they may have been exposed to mycobacteriosis from handling infected fish should contact their physician and inform them of the nature of the exposure. Diagnosis and treatment may be difficult, especially in view of emerging antibiotic resistance in fish pathogens.

However, "fish keeper's disease" is not a focal infection of the skin. A case of mycobacteria infection contacted from mouth syphoning water from a fish tank has been reported. It concerned an individual who experienced a throat infection that wouldn't get better, and was eventually diagnosed as fishtank granuloma (Practical Fish Keeping, Jan. 1998). So next time you do a water change and take a big suck on the end of the syphon hose - just think of this article.



A number of products are manufactured and marketed for the therapy and prophylaxis of fish diseases. Some of these chemicals can be dangerous if applied incorrectly and should only be used by experienced aquarium specialists. A very thin line separates effective treatment levels from overdoses that will kill the fish.

ACRIFLAVINE - this dye has both antibacterial and antiprotozoal activity at a dose of 2 to 3 ppm. It will also kill aquatic plants, colour the water yellow. Many organisms are resistant and it has been largely replaced by more specific treatments. 5-10 mg/L in water for several hours to several days.

ACRIFLAVINE NEUTRAL -- 5–10 mg/L in water for several hours to several days.

ALKA-SELTZER -- several tablets in 17 to 34 ounces of water for euthanasia of fish by  $CO_2$  toxicity.

ALUM (ALUMINIUM SULPHATE) -- dose "to effect" to decrease pH in pools and aquariums.

ACETIC ACID -- 1000 to 2000 ppm dip for 1 to 10 minutes as a parasiticide for fish.

BENZALKONIUM CHLORIDE -- this blend of quaternary ammonium compounds is used at a concentration of 1 mg/L as a one-hour bath to treat susceptible bacterial diseases, especially gill conditions where excess mucus production is a problem; it is also utilised as a disinfectant for nets and other equipment.

BENZOCAINE -- used for sedation of fish at a dose of approx. 10 to 40 mg/L of aquarium water; for anaesthesia at a dose of approx 50 to 500 mg/L in the water or sprayed as an aerosol on the gills; and for euthanasia "to effect".

CALCIUM CHLORIDE -- used to increase water calcium concentration to insure proper egg hardening. Dosages used would be those necessary to raise calcium concentration to 10 to 20 ppm.

CALCIUM CARBONATE  $(CaCO_3)$  -- up to 150 ppm indefinitely to increase the hardness of water for holding and transporting fish in order to enable fish to maintain osmotic balance.

CALCIUM OXIDE -- used as an external protozoacide for fingerlings to adult fish at a concentration of 2000 mg/L for 5 seconds.

CARBON DIOXIDE GAS -- for anaesthetic purposes in fish.

CHARCOAL (ACTIVATED CARBON) -- used in filtering systems to eliminate chlorine, as well as antibiotics and other impurities. Charcoal is sold by many aquarium supply companies in bags which fit inside individual filters, or as blocks which are inserted into the lines of multi-tank filtering systems (200 mg/L).

CHLORAMINE-T -- at a dose of 0.5 to 2 mg/L, this disinfectant has been reported as a successful treatment for a variety of bacterial infections of aquarium fish; its action is based on the fact that it slowly breaks down to hypochlorous acid, releasing oxygen and chlorine. Chloramine-T can be administered in a bath at the following dose rates for 2–3 days, using lower dose rates in soft water with a low pH:

pН	Soft Water	Hard Water
6.0	2.5 ppm	7.0 ppm
6.5	5 ppm	10 ppm
7.0	10 ppm	15 ppm
7.5	18 ppm	18 ppm
8.0	20 ppm	20 ppm

This chemical is best used by aquarium hobbyists simply as a disinfectant for equipment.

CHLORAMPHENICOL -- Columnaris, Enteric Red Mouth, Finrot, Furunculosis, Haemorrhagic Septicaemia, Pasteurellosis, Ulcer Disease, Vibriosis.

(a) 50-70 mg/kg of food/day for 5-10 days(b) 10-50 mg/L of water, as a bath

This drug should not be used for home aquaria because it is unstable in water, and poorly absorbed by target fish; and, it can cause fatal human aplastic anaemia if touched by a person who is allergic to the compound.

CHLORMON -- used to neutralizes ammonia, chlorine and chloramines. Chlormon deals with ammonia instantly, destroying it completely leaving the tap water safe for immediate use. Chlormon used in fish transport water, eliminates ammonia as it is produced during transit, allowing for a longer and safer journey. May be added directly to a bag of fish. Chlormon is also safe for invertebrates, no effect on marine salts. Quantitative dose rate 5 mL of Chlormon per 20L neutralizes 0.75 ppm ammonia. 5 mL/20L also neutralises chlorine and chloramines in tap water.

CHLORINE -- use household bleach equal to 5.25% sodium hypochlorite as a disinfectant for aquarium equipment; chlorine can then be rinsed off with sodium thiosulfate wash.

CHLOROQUINE DIPHOSPHATE -- this anti-malaria drug is effective against Amyloodinium at a dose of 40 mg/gal used as a prolonged immersion of three weeks' duration.

COPPER SULPHATE (CuSO<sub>4</sub>) -- this "old-time" medication has seen decades of use both as an effective algaecide and an external parasite treatment, but there are much better preparations available today. Copper sulphate has also been used with success as an algae control by aquarium owners at the 0.1 to 0.2 ppm level, but some find the resulting water chemistry too harsh to grow decorative plants and are forced to remove it through charcoal filtration. Copper sulphate is extremely toxic, particularly in water of low



alkalinity. Never use copper sulphate without testing the alkalinity of the water, carefully measuring the volume of the aquarium or pond to be treated, and weighing the amount of chemical to be applied.

The concentration of copper sulphate to apply is often calculated by determining the total alkalinity of the water and dividing that number by 100. For example, if the total alkalinity of the aquarium is 100 mg/L, then  $100 \div 100 = 1$  mg/L copper sulphate. Do not use copper sulphate if the total alkalinity is less than 50 mg/L. If you are unsure how to measure the alkalinity of your water, or have never used copper sulphate, then do not use it.

Because of its algicidal activity, copper sulphate can cause dangerous oxygen depletions, particularly in warm weather. Emergency aeration should always be available when copper sulphate is applied to your aquarium systems. Copper sulphate should not be run through the biofilter on a recirculation system, as it will kill the nitrifying bacteria. If possible, tanks should be taken "off-line" during treatment with copper sulphate. If necessary, clean the biofilter manually to decrease organic debris and residual parasite load.

When using a commercially formulated copper cure, always follow the label instructions for dosage rates. Chelated copper will stay in solution longer than copper sulphate and appears to be safer to fish. You can create your own chelated copper by using two parts citric acid to one part copper sulphate, by weight. Combine both in distilled water and dissolve them together. It is important to remember that you will be treating with the copper sulphate and not the citric acid, so when weighing the formula, use only the weight of your copper sulphate in calculating dosages.

Most fish are extremely sensitive to copper. Concentrations of copper as low as 42 and 17  $\mu$ g Cu/L were found to be acutely toxic to the Penny-fish (*Denariusa bandata*) and the Eel-tailed Catfish (*Porochilus rendahli*) respectively. *Melanotaenia inornata* and *Ambassis spp*. have been found to be sensitive to copper; half of the individuals tested died at copper concentrations between 120 and 200  $\mu$ g Cu/L. The atyid shrimp, *Caridina sp.* is extremely sensitive to copper, dying at levels of only 2  $\mu$ g /L. River prawns, *Macrobrachium sp.* were found sensitive to copper with half the individuals dying at 160  $\mu$ g Cu/L. Snails are also known to be very sensitive to copper. For example the snail *Physatra gibbosa* (of New South Wales), succumbs at 31  $\mu$ g Cu/L after 7 to 9 days.

Copper sulphate is for specialist use only as it is highly toxic and requires removal. It is inadvisable to use this compound where other treatments are available.

DICHLORVOS -- (see Organophosphates)

DIFLUROBENZURON -- used to treat crustacean copepods as a prolonged immersion at a dose of 0.11 mg/gal.

DOXYCYCLINE and MINOCYCLINE -- used against susceptible bacteria as a prolonged immersion at a dose of 2 to 3 mg/L.

EPSOM SALTS -- (see Magnesium Sulphate)

FENBENDAZOLE -- used to control intestinal helminths in fish. A dosage of 25 mg/kg, delivered in food for 3-5 days, has been commonly recommended, but this regimen has not been evaluated in controlled trials. Also been reported as an effective control for hydra used at 2 mg/L. This chemical is available in various formulations and trade names.

FLUMEQUINE -- used for infections caused by susceptible bacteria; most effective as a dip if the water pH is near neutral; as a dip, use 50 to 100 mg/L for three hours.

FORMALIN -- used as a water treatment to control external parasitic infections. It is extremely effective against most protozoans, as well as some of the larger parasites such as monogenetic trematodes. Formalin effectively kills parasites on the gills, skin, and fins. It is not generally considered the preferred treatment for external bacterial or fungal infections. Formalin can be used in a short-term bath at a concentration of 175–250 mg/L if water temperature is greater than 20° Celsius for no more than 30–60 minutes. Treatment should never exceed 1 hour even if the fish show no signs of stress. It can be used as an indefinite bath at a concentration of 15–25 mg/L for up to 12 hours.

Formalin has a high level of toxicity and fish under treatment must be followed closely for toxic signs such as respiratory difficulties. Under some conditions, fish may be stressed by normal treatment concentrations. Heavily parasitised or diseased fish often have a greatly reduced tolerance to formalin. Such fish do not tolerate the normal tank treatment regime the first time they are treated, and the time or dosage or both may need to be reduced. If adverse reaction is observed, fish should be removed from the treatment tank at once and placed in clean well-aerated water. Careful observations should always be made throughout the treatment period whenever tank treatments are made.

Formalin is a generic term, which describes a solution of 37-50% formaldehyde gas dissolved in water. Formaldehyde is a colourless gas with a pungent, suffocating odour at room temperature; the odour threshold for formaldehyde is 0.83 ppm. The chemical formula for formaldehyde is CH<sub>2</sub>O and the molecular weight is 30.03 g/mol. Solutions of formalin for use as a fish medication should contain 10–15% methanol, which inhibits formation of paraformaldehyde, a highly toxic substance.

Formalin can be combined with malachite green (0.1 mg/L malachite green mixed with 25 mg/L formalin) to treat external protozoans diseases. The two chemicals work well together and are very effective for the control of various external parasites of freshwater fishes.

The toxicity of formalin and therapeutical success is influenced by water parameters and is falling out of favour as an aquarium treatment because of its undesirable qualities. It is a reducing agent and thus lowers the available oxygen level in the water, which is hardly favourable to fish being treated. Each 5 mg/L of formalin applied removes 1 mg/L of dissolved oxygen. However, this can be avoided in aquarium systems by always supplying adequate aeration whenever formalin is used.



Formalin toxicity is increased at high water temperatures. If water exceeds 21° Celsius, the concentration of formalin delivered in a prolonged bath should be decreased. Formalin is carcinogenic to laboratory rodents and causes contact dermatitis and lung damage in people; it is volatile; and, it is a direct irritant to fish gills. Laboratory experiments have shown that juvenile fish exposed to high concentrations died and fish embryos exposed to low concentrations were unable to hatch.

Formalin is for specialist use only. It is inadvisable to use this compound where other treatments are available.

FULLER'S EARTH -- used to reduce the adhesiveness of fish eggs to improve hatchability.

FURALTADONE -- related to nifurpirinol with which it shares antimicrobial activity; as an immersion, use 20 to 50 mg/L and treat fish for one day.

FURAZOLIDONE -- related to nifurpirinol with which it shares antimicrobial activity; as a prolonged immersion, use 1 to 10 mg/L and treat for 24 hours.

HYDROGEN PEROXIDE -- use as a treatment of acute oxygen insufficiency at a dose of 0.25 ml of a 3% H<sub>2</sub>O<sub>2</sub> solution per litre of water. It can also be used to treat external protozoans at a dose of 10 ml of a 3% solution per litre of water as a 10 to 15 min bath. Used at 250–500 mg/l to control fungi on all species and life stages of fish, including eggs.

There are many different doses suggested in the literature for use in aquarium fish. However, because there are hundreds of species of ornamental fish, certain factors must be taken into account when using Hydrogen peroxide. It can be very toxic to some species, and certain life stages may be more sensitive. Increasing temperature seems to increase the potential toxicity. Dosage and duration of treatment will also determine whether fish being treated will live or die. Hydrogen peroxide can cause mortalities primarily by damaging the gills. Therefore, toxic effects will often be seen related to gill damage, as indicated by gasping near the surface, or increased ventilation rates.

Contrary to popular belief, in water with relatively low organic content, the concentration of Hydrogen peroxide does not decrease significantly. Of course, any increase in organic loading will change this factor, but the bottom line is that Hydrogen peroxide does not break down as quickly as some may think. Water changes are required after treatment.

More work has to be conducted on the use of hydrogen peroxide, especially its safety, efficacy, and effects on biofiltration. More organics in the system lessen the likelihood that biofilter bacteria will be damaged or killed by these chemicals. However, too high an organic load will render this chemical ineffective as a treatment.

IVERMECTIN (1%) -- Some studies have shown that Ivermectin added directly to aquarium water has been useful in treating Camallanus worms in fish. The dose used was 0.7 millilitres of a 1% injectable solution per 76 litres of water. The dose was added over a period of four days (0.1, 0.2, 0.2, and 0.2 millilitres). A solution of 1 part Ivermectin 1% in 19 parts distilled water can be made and administered as a split dose of 2 ml on day one, 3 ml on day two, and 3 ml on day three followed by a water change on day four. However, because this drug has a narrow margin of safety, some veterinarians advise against any use of Ivermectin for aquarium fishes because it can cause neurologic signs and death in fish at therapeutic doses.

LEVAMISOLE HCL -- used for the treatment of susceptible nematodes at a dose of 10 mg/L of water as an immersion. 1 mg/L 1 to 2 days for skin and gill flukes; 2 mg/L once per week for 3 weeks for Camallanus.

MAGNESIUM SULPHATE -- used to treat external monogenetic trematode infestations and external crustacean infestations in fish at all life stages. Used in all freshwater species. Fish are immersed in 30,000 mg/L MgSO<sub>4</sub> and 7000 mg/L NaCl solutions for 5 to 10 minutes.

MALACHITE GREEN -- used for treatment against parasitic protozoans infecting the skin of freshwater aquarium fishes. When used as directed, the medication will control or prevent the following common protozoan parasites: Ichthyophthirius, Ichthyobodo, Chilodonella, Ambiphyra, Cryptocaryon, Epistylis, Piscinoodinium and Trichodina. Malachite green is also effective against common external fungal infections of fishes and eggs, which include Achlya and Saprolegnia.

Malachite green was originally developed in the 1920s as a textile dye. In its original form, it contained zinc, which is toxic to fish. However, later variants came in a zinc-free form, making it more applicable as a fish therapeutant. In the 1960s, malachite green proved to provide the most effective treatment against protozoan ectoparasites, particularly Ichthyophthirius multifiliis. It became even more important when its effectiveness against Saprolegnia in fish eggs were demonstrated. Since then, malachite green has been extensively used in controlling infections due to bacteria, fungi, protozoans and monogenetic trematodes on eggs, fry and adult fish. It is used by itself, or in combination with formalin, salt or dimethyl sulphoxide (DMSO). Malachite green is also used in multicomponent treatment baths (malachite green in combination with formalin, brilliant green, crystal violet, methylene blue, etc.). Malachite green combined with formalin (0.1 mg/L malachite green mixed with 25 mg/L formalin) work well together and are very effective for the control of various external parasites of freshwater fishes. The most favourite multi-component preparation for aquarists is a mixture of formalin, malachite green and methylene blue known as FMC.

In recent years, however, there have been strong moves against malachite green application, especially with respect to its use in food fish. In 2000, the use of malachite green for food fish was banned in the EU because the general public may become exposed to malachite green through the consumption of treated fish. This is because the chemical is believed to have potential teratogenic, mutagenic or carcinogenic attributes. While there has been no evidence actually linking malachite green with any carcinoma, its use in food fish has been banned in many countries. Studies have also indicated that malachite green may have very long withdrawal times. Residues of malachite green have been found in fry some 30 days after eggs were disinfected.



Despite its toxicity, it is commonly used to control parasitic protozoans on ornamental fish. Malachite green is used for treatment against parasitic protozoans infecting the skin of freshwater aquarium fishes. When used as directed, the medication will control or prevent the following common protozoan parasites: *Ichthyophthirius, Ichthyobodo, Chilodonella, Ambiphyra, Cryptocaryon, Epistylis, Piscinoodinium* and *Trichodina.* Malachite green is also effective against common external fungal infections of fishes and eggs, which include *Achlya* and *Saprolegnia.* An extensive body of literature supports its use as an effective agent in the control of the above mentioned parasites.

Malachite green is quite effective when used at concentrations of 0.05 to 0.10 mg/L as an indefinite bath or 1–3 mg/L of water for up to 60 minutes. It is also used to treat eggs against fungal infections as a 2 mg/L wash for 30 to 60 minutes. This chemical can be extremely harsh on fish, particularly on gill tissue, so be careful not to overdose the fish. Lethal concentrations for fish and recommended therapeutic concentrations are sometimes very close to each other. Dosage calculations should be double-checked before applying treatment. Therapeutic concentrations could be different for distinct species as well as for different developmental stages of fish. It is usually applied at 0.01 ppm for postlarvae and 0.1 ppm for juveniles. It also seems to be more toxic to scaleless fish than fish with scales when used at the same concentration, and should be avoided on these species.

The progress of intoxication is very rapid. Typical clinical symptoms include restlessness and uncoordinated movements of the fish in the tank. The fish move in the upper half of the tank, leap above the water surface, and gasp for air, which is followed by the loss of balance, apathy, agony and death. The pathological anatomical picture of fish intoxication with malachite green is characterised by greenish tinge of their skin and increased production of skin slime. The gills are oedematous, with excessive amounts of mucous matter, and are discoloured by the agent. Vessels in the body cavity were dilated, and muscle tissues and internal organs were often light-green in colour.

Large differences in the toxicity of malachite green in dependence on its purity and varying concentrations of residual impurities that could render more or less toxic are another major obstacle in its application. High-quality grades of malachite green can be produced by the inclusion of additional purification stages in production, but even a nominal 100% malachite green dry powder by analysis can only contain 82% (oxalate) or 95% (hydrochloride), the rest of the weight being the acid component. Different toxicological properties of malachite green were confirmed in a series of acute toxicity tests on common carp and rainbow trout in which 10 types of malachite green obtained from different sources were investigated. It follows from these experiments that lethal concentrations of some types of malachite green are very close to therapeutic concentrations. In one case the recommended therapeutic concentration of malachite green was even higher than its lethal concentration.

When treating fishes, it should be borne in mind that malachite green toxicity is significantly influenced by the quality of used

water. The toxicity and hence of course its effectiveness are primarily influenced by the reducing substances present in water, for example organic substances, calcium-ions, pH and temperature values. The toxicity of malachite green is reduced for example by humic substances in soft water. At lower water temperatures, the fish can tolerate slightly higher concentrations of malachite green than at higher temperatures. During the hot summer months the exposure time for malachite green treatment should be decreased. Malachite green is also more toxic at low *p*H. Malachite green has two forms depending on pH. The initial strong green coloured prevails at low pH (acidic), while in alkaline water it is converted to a colourless carbinol form. So in alkaline water it may seem that it has disappeared, but it is still present, but invisible!

For the above reasons, it is important to pay great attention to the selection of malachite green to be used, take water parameters and temperature into account and also observe recommended dosages and exposure periods. One should follow the manufacturer's instructions for treatment, as different manufacturers use different concentrations of the active ingredients.

MEBENDAZOLE -- Mebendazole is chemically related to flubendazole and fenbendazole. It is a benzimidazole derivative, and is a useful broad spectrum anthelmintic, the drug of choice for mixed worm infestations.  $\sim$  used to treat monogenean flukes; as a 24 hour immersion use 1 mg/L.

METHYLENE BLUE -- used against ciliates infecting the skin at 1 to 3 mg/L in a bath for 3 days, or 30 mg/L for a short duration bath. Methylene blue may be used for the treatment of Ichthyophthiriasis (white spot disease), skin and gill flukes, velvet disease, Costiasis, Chilodonelliasis, Trichodina and as a palliative medicine in all cases of disease of the gills, where fishes suffer from difficulty in breathing.

Methylene blue is a redox dye which raises the oxygen consumption of cells. This means that the hydrogen to be oxidised is passed on to the oxygen. Each molecule of the dye is oxidised and reduced about 100 times per seconds. Thus, while disinfection results from this, methylene blue is also excellent against methemoglobin intoxication. The therapeutic action of methylene blue on bacteria and other parasites is probably due to its binding effect with cytoplasmic structures within the cell and also its interference with oxidationreduction processes.

Methylene blue is also effective against superficial fungal infections of fishes and may be used as an alternative to malachite green for the control of fungus when it is known that the fish to be treated are sensitive. It is safe for use with fish eggs and fry for the prevention of fungal infection. It is particularly effective against Saprolegnia by applying 3 mg/L for long duration. At a concentration of 2–3 mg/L, it can be used as an indefinite bath for fish at all ages. When used against gill rot and other bacterial disease, the rate is at 8–10 mg/L as a bath treatment. It can also be applied during quarantine treatment of aquarium fishes by using an indefinite bath of 1 mg/L.

Methylene blue has a wide safety margin and is non-toxic when used as recommended. Fish tolerate relatively high



dosages without side effects. However, it should not be used in recirculation systems that utilise biological filtration, as it will interfere with the normal biological processes of nitrifying bacteria. It can also interfere with normal plant growth. Methylene blue is best used in bare aquariums as porous materials such as rock and driftwood will absorb the dye and it may permanently discolour the silicone sealant. At the conclusion of treatment, a partial or complete water change should be made to remove any chemical residues or use activated carbon in the filter.

Methylene blue comes in various fish medication preparations available at pet shops, and these are more convenient to use than the pure form. One should follow the manufacturer's instructions for treatment, as different manufacturers use different concentrations of the active ingredient.

METRONIDAZOLE -- used to control flagellated protozoans and can be delivered in a medicated food or as a bath if fish are not eating. A concentration of  $\sim$ 7 mg/L can be administered daily for 5 days. A daily water change a few hours after treatment is recommended. Metronidazole can be administered at 50 mg/kg delivered in food, for 5 days. Anecdotal information suggests that excessive treatment (10 times the recommended dosage for 30 days) with metronidazole may be associated with reproductive failure in some fish.

#### MINOCYCLINE -- (see Doxycycline)

MS 222 (3-aminobenzoic acid ethyl ester methanesulfonate salt) -- in carbonate buffered aquaria water. Rainbowfish can be anaesthetised by bathing for 3 mins in a concentration of 150 mg/L.

NIFURPIRINOL -- this nitrofuran compound is commonly used, and effective, against many aquarium microbes: as a dip, at 1 to 2 mg/L for 5 min to 6 hours; as an immersion, at 0.1 mg/ L for three to five days.

ORGANOPHOSPHATES -- drugs of this group are used to treat a wide assortment of metazoan ectoparasites; there are a number of such compounds in this classification, but the ones practicing veterinarians are most likely to find useful are dichlorvos and trichlorfon in a variety of concentrations and combinations; trichlorfon is used as an ectoparasiticide effective against flukes, fish lice, and anchor worms at a dose of 0.2 mg/L active ingredient as a permanent treatment, or as a 2 to 2.5%, five to ten minute dip; trichlorfon degrades to dichlorvos (the antiparasitic entity) and further to dimethyl-hydrogen-phosphate in aquarium water—this chain is water pH and hardness dependent—faster in hard, alkaline water than soft, acid water.

PAPAIN -- use of a 0.2% solution in removing the gelatinous matrix of fish-egg masses in order to improve hatchability and decrease the incidence of disease.

PHENOXYETHANOL -- this drug has been used as an anaesthetic at doses of 0.1 to 0.5 ml/L [100 to 500 mg/L], and is also claimed to have antibacterial action; the biological activity is temperature-dependent and lower doses can be used at lower water temperatures; there is a narrow margin of safety and 2X doses will kill fish.

POTASSIUM CHLORIDE -- used as an aid in osmoregulation; relieves stress and prevents shock. Dosages used would be those necessary to increase chloride ion concentration to 10–2000 mg/L.

POTASSIUM PERMANGANATE -- for use against ciliates infecting the skin; use 4 mg/L in a bath for 30 to 60 minutes.

POVIDONE IODINE -- 100 mg/L solution for 10 minutes as an egg-surface disinfectant during and after water hardening.

PRAZIQUANTEL -- useful against tapeworms and monogenetic flukes; at a dose of 2 mg/L, this drug has been shown to remove tapeworms within one hour, and external flukes within one day.

Praziquantel has been identified as the most effective "in water" treatment of infected fish. Praziquantel is harmless to fish of all species, is non toxic to plants, and has no negative filter impact. Praziquantel is a bitter tasting powder which shows good absorption directly from the treated water, and then admirable clearance of various surface and internal flukes and worms in fish. Praziquantel has been known to the hobby for many years. Praziquantel was traditionally available in the form of branded Droncit<sup>®</sup> tablets, for oral administration in dogs and cats, but is now available in a range of aquarium products.

Praziquantel used at 2–3 mg/L is very effective for control of both gill and body flukes and has a wide margin of safety for fish. Praziquantel is toxic to flukes on contact, paralysing the parasites within 15 seconds under laboratory conditions. Praziquantel preparations must be dosed high enough and long enough for effective treatment. Monogeneans can be persistent in aquarium systems necessitating regular treatments. In cool water, the parasites move through their life cycles slowly, so it is important to medicate long enough to intercept the emerging larvae. When temperatures are above 25° Celsius, treat once every 3 to 4 days for a total treatment time of 20 days.

When temperatures are between 20 and 25° Celsius, treat once every 4 to 5 days for a total treatment time of 25 days. The eggs can be resilient to chemical treatment, which make the use of multiple chemical treatments appropriate to control this group of organisms.

Praziquantel can also be administered in food at a dosage of 35-125 mg/kg for up to 3 days or as a short-term bath treatment at a concentration of 10 mg/L for 3 hours.

Change 50–75% of the water in between the chemical treatments. Fish, which are obviously weak and heavily parasitised may not survive. Management to lessen the chance of infestation by these parasites includes maintaining the fish in a good nutritional state and avoiding water quality problems that might weaken the fish.

The effectiveness of the long-term use of Praziquantel has been evaluated in ornamental fish. Cumulative doses up to 10 mg/L water were tolerated without side-effects by Angel Fish (*Pterophyllum scalare*), Discus, and a variety of catfish species (*Ancistrus sp., Corydoras sp.*). It was found appropriate to start with a dosage of 2.5 mg/L and to add the same dosage every other day several times.



All adult parasites and larvae were killed by this treatment. For the complete elimination of Dactylogyridae populations in a closed aquarium system, 3 therapy-cycles (duration: 5-6 days, accumulated dosage: 2.5 mg/L/day) proved to be effective. It was important to interrupt the therapy-cycles with intervals without medication (1 to 4 weeks). However, there are reports that kissing gouramis (*Helostoma temminckii*) have been adversely affected by Prazifish<sup>®</sup> (Praziquantel 98.5 mg/g).

SODIUM BICARBONATE -- 142 to 642 mg/L for 5 minutes [or to effect] as a means of introducing carbon dioxide into the water to anaesthetise fish [higher doses to euthanize fish].

SODIUM CHLORIDE (NaCl) -- also known as salt, has many potential applications in fish keeping. It effectively controls some parasites and minimises osmoregulatory stress. Immersing rainbowfishes in a salt concentration of 30 g/L of water for 10-30 minutes may effectively eliminate some parasitic infestations (stop the treatment earlier if the fish show signs of stress). Weaker solutions containing 5 to 10 grams per litre of water may be used as a bath for several hours to eliminate some freshwater parasites.

The use of ordinary salt (or sea water) was among the first of the methods proposed to combat fungal disease. Often, the application of salt either directly onto the diseased part of individual fish, or as a solution in which to bathe the fish. Salt can be used at 10 g/L for 20 minutes for young fish and 25 g/L for 10-20 minutes for older fish. A continuous well-aerated salt bath of 2–5 g/L may assist in recovery by preventing fungal infections. However, there appear to be significant differences among species and possibly families as well in the tolerance of the larval and fry stages to salt. 0.5 to 1% solution for an indefinite period as an osmoregulatory aid for the relief of stress and prevention of shock in fish. A 0.3 to 0.5% solution will control Hydra; a 10 to 15 min bath in a 2 to 3% solution facilitates the removal of leeches. Salt is very effective against Trichodina, at a rate of 0.3%, added 0.1% every 12 hours for 3 treatments. Some strains have been found to be resistant against salt, so another choice of treatment would be Quick Cure<sup>®</sup> or any other medication containing Formalin.

Care must be exercised to avoid over treatment, which will place the fish in the same condition of osmoregulatory shock. Water constantly enters the body of freshwater fish because their body fluids have a higher salt content than the surrounding water. Salts will move from areas of high concentration. (blood), to low concentration. (fresh water), by diffusion. While the skin is moderately watertight because of a mucus coating, the gills and oral membranes allow water to pass through passively. Therefore, although these fish drink very little water, by controlled elimination they must excrete large volumes of urine and take in salt to maintain an osmotic balance within the narrow limits necessary for life. Any physical damage to the external tissues allows increasingly more water to enter the body, (and salt to escape), placing an additional burden on the kidneys. With just moderate injuries, this can become too much and the kidneys will fail causing death.

The blood salt content of rainbowfishes is approximately 9 g/L (0.9 percent), and an average pH 7.4. Almost 80 percent of this

blood salt is sodium chloride, (NaCl), the remainder made up of bicarbonate, potassium and calcium. Sodium and potassium are vital for normal heart, muscle, and nerve function, excessive loss causing heart failure plus muscle and nerve spasms. Damage and stress caused by capture and handling of fish results in the loss of salt which must be replaced. This replacement is an active process requiring body energy from an already stressed and weakened animal.

The salt addition must be exact and monitored: 10 g/L of NaCl is 10 percent higher than the total blood salt content and may cause some water loss and salt diffusion into the blood resulting in dehydration. 7 g/L is slightly lower than normal blood and is probably optimal for holding and shipping water: dehydration will be avoided, salt loss will be low, and the kidneys will be active but not overloaded. However, those species not adapted to elevated salt content may not tolerate this concentration and more dilute salt solutions should be trialed. By understanding the need to maintain a water balance in freshwater fish, one can understand why using salt during transport is beneficial. Most freshwater fish can tolerate a salt concentration of 1-3 g/L, and this level is not harmful to the biological filter.

Salt Solution 1 gram/Litre water = 0.1% 10 grams/Litre water = 1%

SODIUM SULPHITE -- use a 15% solution for 5 to 8 minutes on fish eggs to improve their hatchability.

SODIUM THIOSULFATE -- used to remove chlorine from aquarium water; there are many commercial preparations available (follow package directions); in instances where chlorine and chloramine are bonded and both contained in the source water, sodium thiosulfate will break the bond and detoxify the chlorine but leave the ammonia, which then must be removed.

TOLTRAZURIL -- for use against ciliates infecting the skin and gills; reported to be active against trophozoites if used at a dose of 10 mg/L for two hours on day 1, then 20 mg/L on days 2 and 3.

#### TRICHLORFON -- (Case Report)

An ornamental fish and aquarium plant producer noted approximately 50% of fish in a pond to be swimming in erratic circles, apparently due to having bent bodies varying from subtle to extreme. The pond was primarily used for plant production, and contained mixed species and sizes of rainbowfishes. All affected fish were the pygmy rainbowfish (*Melanotaenia pygmaea*) that were greater than 5–6 cm in length. One affected fish was submitted to Berrimah Veterinary Laboratories for evaluation.

At gross necropsy, spinal curvature in the dorso-ventral plane of the proximal tail region was noted. Histological examination revealed severe alteration in the normal size, shape and cellular organisation of one region of the spinal column. There was fragmentation and collapse of a vertebral body with associated fibrosis and irregular bony proliferation, consistent with earlier fracture of the vertebral column and attempts at regeneration. Adjacent muscle fibers were necrotic. There was no evidence of an infectious cause, such as bacterial, fungal or parasitic infection of the affected tissue to explain the lesions.

Subsequent close questioning of the producer revealed that the pond had been treated repeatedly in the past weeks with an organophosphate pesticide containing trichlorfon, at a rate of 0.5–2 ppm, to control aquatic invertebrate pests. Organophosphate pesticide exposure of fish occurs relatively commonly, either inadvertently, due to environmental contamination, or deliberately, for treatment for fluke, leech and crustacean fish ectoparasites.

Organophosphates commonly used to treat fish are trichlorfon and dichlorvos. When added to water, trichlorfon degrades to the more toxic dichlorvos, a process that is influenced by light, high water temperature and high pH. Also, organophosphate uptake and toxicity in fish is increased by low oxygenation of the water. These factors result in variable response of fish to exposure to organophosphates, with levels greater than 0.1 ppm being potentially toxic.

Organophosphates exert their toxic effect by inhibition of acetylcholinesterase, an enzyme involved in terminating neurotransmission at cholinergic synapses in the central nervous system, some peripheral autonomic junctions and neuromuscular junctions. In intoxicated fish that don't die acutely from central nervous system dysfunction, the muscle spasms produced by excessive and prolonged stimulation of neuromuscular junctions of the muscles of the body are thought to be sufficiently severe to result in spinal fracture and lesions as seen in this case.

UREA AND TANNIC ACID -- used to denature the adhesive component of fish eggs at concentrations of 15 g urea and 20 g NaCl/5 litres water for approx. 6 minutes, followed by a separate solution of 0.75 g tannic acid per 5 litres of water for an additional 6 minutes.

### Chemical Effect on Nitrifying Bacteria

Chemicals used to treat fish diseases and parasites can be toxic to nitrifying bacteria at therapeutic levels for fish. Research has shown that there are some differences in the inhibitory effects of formalin, malachite green, methylene blue, copper sulphate, and potassium permanganate on the biofilter bacteria. Different studies show different effects.

Formalin used in one study, at 25 mg/L had no effect, whereas another study showed reduction of biofilter bacterial activity by 27% when used at 15 mg/L. As a rule of thumb, most aquaculturists do not consider the use of formalin at 15–25 mg/ L to have a major impact on the biofilter. However, when testing for ammonia levels, formalin will react with Nessler's Reagent (a component of most ammonia test kits) and can give a falsely elevated ammonia reading. In systems treated with formalin, the salicylate reagent test for ammonia is recommended because it does not react with aldehydes (e.g., formaldehyde found in formalin).

Malachite green has been shown to have no effect on the biofilter at 0.1 mg/L, combined with or without formalin at 25 mg/L. Copper sulphate at 1 and 5 mg/L likewise had no effect on biofiltration.

By contrast, potassium permanganate experiments have been mixed. In one study, a 4 mg/L dosage resulted in no inhibition of the biofilter, whereas in another study, a 1 mg/L dosage resulted in an 86% inhibition. The actual impact on an individual system will most likely depend upon many factors, such as chemical concentration, length of time in treatment, organic load, pH, temperature, alkalinity, filtration, oxygen levels, and stocking density; and, although this will most likely be true for most chemicals, this may better explain the differences in effect by potassium permanganate.

Antibiotics should never be used in aquarium systems because of severe detrimental effects on the bacteria within the biofilter. If a population of fish in a recirculating aquarium system must undergo a specific antibiotic treatment, the biological filtration system should be shut off during treatment. After treatment, a large water changes for the treated aquarium is recommended.



# Antibiotics

Aquarium systems support large populations of bacteria. Nevertheless, they are among the least known and understood elements of aquarium keeping. They can cause diverse pathological conditions that include both acute systemic and/or chronic diseases. One of the most common means of treating these bacterial infections is to administer antibiotics. However, this is a grossly misunderstood part of aquarium and pond keeping and is rarely presented correctly to the aquarist or pondkeeper. The belief that antibiotics will solve all your problems is a common mistake often encountered in fish keeping. Antibiotics are of limited use for treating aquarium fishes because none of the antibiotics have been originally developed for fishes. Their requirements were originally quite different, with only some being adaptable to be used for aquarium fishes.

Since they were first discovered, antibiotics have revolutionised the treatment of a whole range of previously deadly diseases in man and animals. However, as a traditional strategy for aquatic disease management, antibiotics have been extensively criticised for the potential development of antibiotic-resistant bacteria, as well as having marginal effect in most cases. Antibiotics are generally not a hundred percent effective for treating aquarium fish against bacterial infections; some cannot be controlled with antibiotics. The success of chemotherapy with bacteriostatic compounds often depends on the ability of these compounds to control bacterial growth until the immune response can cope with the invaders. However, certain antibiotics also have been shown to suppress the immune system, potentially making aquarium fish more susceptible to viral or parasitic infections. Furthermore, the question remains whether these treatments completely eliminate infection. A likely scenario is that treatments only eliminate overt clinical signs and the treated fish become asymptomatic carriers.

A number of aquarium products are manufactured and marketed for the treatment and prevention of bacterial diseases. However, medications sold and used in the aquarium hobby vary in quality and effectiveness. In fact, some fish medications simply do not work. In 1974, Trust and Chipman tested eight products marketed for the treatment of bacterial diseases in aquarium fishes. The products contained erythromycin, neomycin, nitrofuran, penicillin, sodium sulfathiazole, sodium sulfamerazine, sodium sulfamethazine, streptomycin or tetracycline. When used at the concentration recommended by the manufacturer, the products failed to inhibit the growth of bacterial species known to be potential pathogens of aquarium fishes. Furthermore, one of the more effective antibacterial formulations was toxic to the fish. The results also showed that markedly higher levels of the formulations also failed to significantly decrease the numbers of viable bacteria in the aquarium water. It is worth noting that control of bacterial growth in aquariums may further be complicated by the presence of filtration (biological and chemical), sand, gravel, plants, and organic matter, since these will reduce the efficiency of the antibacterial compound by direct inactivation or by mechanically protecting the bacteria from attack.



The method most commonly used to treat bacterial diseases of aquarium fishes is to bathe the fish in a water-soluble antibacterial compound. Although few studies have directly compared drug levels attained by different routes of administration, practical experience suggests that therapeutic levels can rarely be attained by bathing fish in chemotherapeutics. In fact many antibiotics are not suitable, being ineffective for use in water against bacteria, because the antibiotics are not readily soluble in water. This is because the antibiotics are for human and animal oral ingestion, and remain insoluble until the internal organs do the dissolving. If the antibiotics are added to the water, many will only moderately dissolve. At best, for those antibiotics that do fully dissolve in the water, they will be effective for at most about an hour or less in the aquarium or pond water. Therefore, bath treatments should only be considered when treating primarily external bacterial infections of the skin and gills of fish.

Currently, most of the therapeutic dosages used for aquarium fish have been extrapolated from the aquaculture literature. This is because there has been relatively little research related to the pharmacology for aquarium or ornamental pond fishes. Much of the literature dealing with antibiotic usage in aquarium fish is empirical and anecdotal. Therefore, it seems undesirable to continue to allow the unrestricted sale of antibiotics for aquarium use.

Nevertheless, despite these limitations, there are times when antibiotics can be successfully used in treating certain bacterial infections of aquarium or ornamental pond fishes. The problem is in knowing in each case what the bacterial infection is and which antibiotic is the right one to use. There are not a lot of specialists in aquarium and ornamental pond keeping who can readily provide this answer. Without expert testing of the antibiotic on the infecting bacterium, it is impossible to know beforehand whether it will be effective or not as a treatment.

Most cases will require rapid scientific identification of the bacteria involved and selection of a specific antibacterial agent. However, even veterinarians with laboratory diagnostic experience cannot make an accurate diagnosis of some problems without microscopic examination of the fish or cultivation of bacteria. If the fish have a bacterial disease and the causative agent has been identified, a sensitivity test will need to be performed to ensure that the correct medication is used. The incidence of resistant bacteria is high and a sensitivity test will show the resistance of the disease-causing bacteria to various antibiotics. However, when multiple tests are done on a bacterial infection of a particular fish to find out which antibiotic will kill the infection, it is common that the bacteria involved will be found to be resistant to most commercially available antibiotics.

For the detection and identification of bacterial pathogens in populations of fish showing disease signs, ideal samples are multiple (five or more) moribund fish or those showing clinical signs typical of the disease outbreak. For the detection of subclinical infections in populations of asymptomatic fish, larger sample numbers may be necessary. Fish that are found dead at the time of sampling are not suitable for bacteriological examination, unless they are known to be very fresh. Contaminating bacteria can grow quickly in dead fish, particularly in warm water.

Antibiotics are only effective in treating bacterial diseases if treatment is applied very early during the course of the disease. Medicated feed or injection, are the preferred method for treating systemic (internal) bacterial infections. Dose rates are based on fish weight and are expressed as weight of chemical per weight of fish per day for a specified number of days. Improper doses may result in an ineffective treatment or mortalities. However, the effectiveness of oral antibiotic therapy has been inconsistent as infected fish generally have a reduced appetite and, as a consequence, mortalities continue to occur.

Regardless of the antibiotic used, treatments should always be the maximum recommended dose and should be used for the total number of days recommended even if the fish appear to have recovered. If the dose is too high or treatment times are too long, there is a danger of toxicity to the fish, frequently causing liver, kidney, or other organ damage that may or may not be reversible. On the other hand, if the dose of antibiotic is too low or treatment time is too short, the bacteria will not be killed or weakened enough for the immune system of the fish to remove them, and this greatly increases the risk of the bacteria developing resistance to the antibiotic. When bacteria become resistant to a specific antibiotic, even high concentrations of that drug will not be effective.

Over the last number of years the veterinary use of antibiotics has been the subject of media attention. The apparent increase of the occurrence of antibiotic resistance among bacteria from various areas of animal production and its possible implications for public health have in many countries lead to an intensified surveillance of bacterial resistance. Bacteria carrying transferable drug resistance factors have also emerge in household aquaria and in the aquaria of the distributors of ornamental fishes. This pool of multidrug-resistant bacteria may have considerable health implications since fishkeepers can be exposed to aquarium-borne, drug-resistant bacteria. This exposure can be by either direct contact with the water or indirect contact mediated by the spread of a bacterial aerosol formed by the action of aquarium aeration. When ornamental fish are sold by retail stores to the public, the fish and a volume of their aquarium water is generally transferred to a plastic bag. Although this allows the fish to be transferred to the purchaser's aquarium, it also represents one means by which the aquarium hobbyists could be exposed to pathogenic organisms. The aquarium water supplied with ornamental fish purchased at retail outlets contains significant numbers of a wide variety of bacteria. Since the efficacy of antibiotics in protecting the health of ornamental fish is not proven and since the incidence of human infections by drugresistant bacteria is on the increase, compounds used in the therapy of fish diseases should not include antibiotics used in human medicine.

Most countries have a regulatory approval and control system for all antibiotic agents and products containing antimicrobial agents used in animals, including aquaculture. However, the conditions under which antibiotics are supplied varies around the world. In some areas they are available "over the counter" from retailers. In other regions they are available only by prescription from a veterinary surgeon.

# Antibiotic Resistance

Antibiotic resistance is a property of bacteria that enables them to grow in the presence of antibiotic concentrations that would normally kill or suppress the growth of susceptible bacteria. Antibiotic resistance occurs naturally in some genera of bacteria and in others it is acquired. The antibiotic resistance of greatest concern is that which is acquired by bacteria through genetic mutations or through movement of antibiotic resistance genes from one bacterium to another. Resistant bacteria can transfer the resistance to other bacteria (even to bacteria of different genera) that have never been exposed to the antibiotic. At the same time, the fact that one microorganism acquires resistance against an antibiotic seems to help it in becoming resistant against others. Genes that confer resistance to antibiotics let bacteria become resistant to many related antibiotics all at once. Therefore, the continued use of an antibiotic in the presence of resistance allows those resistant bacteria to survive and become dominant within the bacterial flora.

An aquarium contains a mixed bacterial culture growing in a liquid medium, and it is in these conditions in which microorganisms of different species and strains coexist and multiply simultaneously that there is ample opportunity for the transfer of genetic information, including that concerned with drug resistance. The use of antibacterial products at subtherapeutic levels will certainly provide the selection pressures necessary to increase the numbers of these drug-resistant strains in aquaria and hence in the environment.

Antibiotic efficiency has been declining for various reasons, not least the development of bacterial resistance. The causes for appearance of antibiotic resistance can be many and varied. Disease in commercial ornamental fish farms can cause great economic losses and breeding facilities are rarely supervised by fish health services. This has resulted in the indiscriminate use of antimicrobials and chemicals to control infection. These substances are applied prophylactically under uncontrolled



conditions and often using wrong dosages. The uncontrolled use of antibiotic substances leads to the situation that fish come in contact with different antibiotics and chemotherapeutants at an early age and bacterial resistances are built up. Among the wide spectrum of bacteriostatic drugs, the following are used most frequently: nitrofurazone, neutral acriflavine, oxytetracycline (Terramycin), Combiotic (veterinary penicillin) and neomycin sulphate.

Furthermore, antibiotic substances are also used in shipping water. However, while they may strengthen the resistance of fish, they are probably of little value in shipping water. After shipment, the fish undergo further prophylactic treatments to prevent disease outbreaks in the importers holding facilities. Most fish importers use antibiotics in fish tanks as a preventative measure against illness from aquatic fish pathogens. The most common antibiotics used are chloromycetin, tetracycline, metronidazole and sulphadiazine. Therefore, the very high rate of antibiotic resistance in ornamental fish is probably mainly due to the uncontrolled application of antibiotic drugs.

The resistance of bacteria to antibiotics and other synthetic chemotherapeutic agents has been recognised for many years. There is also clear evidence that the use of antibiotics in the ornamental fish industry has been accompanied by the emergence of resistant variants of bacteria associated with fish disease. The emergence of antibiotic multi-resistant bacteria in the ornamental fish industry in Southeast Asia was detected as early as the mid 1970's. Shotts et al (1976) found antibiotic multi-resistant isolates of the '*Aeromonas hydrophila* complex' in water and ornamental tropical fish imported from Southeast Asia. A high percentage of the *A. hydrophila* isolated were resistant to ampicillin, an unnamed tetracycline, sulphamethoxazole-based drugs and streptomycin.

In 1976, a number of aquarium fish were purchased from 14 Canadian retail outlets. The fish were supplied in plastic bags containing water taken from the aquaria in which the fish had been housed. The water in each container was sampled for bacteriological examination immediately upon arrival at the laboratory, and the fish were transferred to holding aquaria. There was a total of 40 water samples, each representing a single aquarium and including a single fish species. Isolated bacteria included 57 strains of *Aeromonas*, 57 *Pseudomonas* and 51 strains of *Citrobacter*. Other bacteria included species of *Acinetobacter*, *Flavobacterium*, *Proteus*, *Providencia*, *Serratia*, *Staphylococcus*, and *Vibrio*. It should be noted that mycobacteria were not isolated in the study, but this may have been due to the relatively short incubation times and the nonselective media used.

A total of 70 different patterns of resistance were demonstrated, with 47% of the isolates being resistant to five or more of the antibacterials employed. The majority of strains were resistant to penicillin, tetracycline, streptomycin, ampicillin, cephaloridine, sulfonamide, and kanamycin, but resistance to chloramphenicol, furadantin (Nitrofurantoin), and nalidixic acid was not uncommon. Resistance to gentamicin and polymyxin was not observed. Water samples containing the aquarium fishes purchased at the retail outlets also contained bacteria capable of growth on media

supplemented with antibiotics or chemotherapeutic agents. This data indicates that water containing ornamental fishes frequently contains bacteria that are resistant to more than one antibiotic or chemotherapeutic agent. Many of the multidrug-resistant species isolated in this study, such as *Pseudomonas fluorescens*, are probably naturally resistant to many of the antibiotic agents tested.

During 1999–2000, a screening program on fish samples collected from two German ornamental fish importers newly imported from four different Asian countries of origin (Hong-Kong, Thailand, Singapore and Sri-Lanka) were examined within two weeks of importation. Two fish species were examined from each country of origin. For each sample, 15 fish for each group were sacrificed, dissected and examined. After differentiation of the bacteria found, antibiogrammes were established on Müller-Hinton-agar. The following substances were involved in the testing procedure: Chloramphenicol, Trimethoprim-Sulphonamide, Oxytetracycline, Furazolidone, Chlortetracycline, Enrofloxacin, Flumequine, Oxolinic acid, Amoxicillin, Gentamicin, Neomycin, Colistin and Florfenicol. The inhibition test was carried out at three pH-ranges: pH 6.0/7.2/8.0.

A total of 250 positive bacterial agents could be detected. Most of the findings were of facultative fish pathogenic nature. Only a few specific bacteria could be identified. A total of 13 cases of mycobacteriosis were detected after Ziehl-Neelsen staining. In ten samples, *Flavobacterium columnare* was identified. Two cases of *Aeromonas salmonicida subsp. achromogenes* and of *Vibrio anguillarium* infection were recorded. In the case of the facultative fish pathogenic bacteria, mainly motile aeromonads were found, most of them *Aeromonas sobria*. Furthermore, *Pseudomonas* (33 cases) and *Myxobacteria* (30 cases) were identified. Another 35 other bacterial and 38 mycotic (fungal) agents were also isolated.

High disparities were observed between the rates of resistance of the different antibiotic substances tested. The lowest rate of resistance was found for Florfenicol (13.4%) while Oxytetracycline showed the highest rate (90.1%). In general, the resistance situation for substances used frequently in ornamental aquaculture (Tetracyclines, Furazolidone, potentiated Sulfonamides) is very unfavourable. In contrary, the rate of resistance for Florfenicol, Colistin and also for Enrofloxacin was low.

In 2002, isolates of *Aeromonas* species from tropical fish imported into the U.S. from Singapore were found to be resistant to ampicillin (94.9%), with variable resistance to cephalexin (76.3%), trimethoprim (37.3%), tetracycline (11.9%), cefuroxime (5.1%), and ceftazidime (1.7%). All strains tested were susceptible to gentamicin, chloramphenicol, and ciprofloxacin. In another study, antibiotic susceptibility tests were performed on 164 strains, and resistance to ciprofloxacin, nalidixic acid, furazolidone, streptomycin and norfloxacin were recorded.

A research project on antibiotic susceptibility with three isolated strains of *Pseudomonas*, found that the bacteria were resistance to eleven out of fifteen antibiotic drugs that were tested. All the strains were highly sensitive to gentamycin while chloramphenicol and cefotaxime ranked second. Resistance to pefloxacin, kanamycin, streptomycin, erythromycin, ampicillin/sulbactam, olfloxacin, amikacin, piperacillin, ciprofloxacin, ceftizoxime and tetracycline were shared by all three strains. Only a few antibiotics are effective against *Pseudomonas*. These have included fluoroquinolone, gentamicin and imipenem, but even these antibiotics are not effective against all strains.

In 2007, approximately fifty diseased aquarium fish were collected from an aquarium shop in Kuala Terengganu, Malaysia. In the laboratory, 25 isolates were successfully isolated from the diseased fish. One isolate of each *Edwardsiella tarda, Flavobacterium sp., Stenotrophomonas maltophilia, Serratia marcescens, Acinetobacter baumannii, Acinetobacter iwoffi, Yersinia sp.* and *Enterobacter sp., 3* isolates of *Chromobacterium violaceum* and 15 isolates of *Aeromonas hydrophila.* 

The result of this study showed that the majority of the isolated bacteria were *Aeromonas hydrophila*. Although *Stenotrophomonas maltophilia* and *Serratia marcescens* are rarely reported in ornamental fish, several studies claimed these types of bacteria have been isolated from diseased fish. In this study, 41.8% cases of antibiotic resistance were recorded. On the contrary, 23.7 and 34.5% cases of intermediary sensitivity and susceptible, respectively against the tested antibiotics were noted. Most of the present isolates were resistant to sulphamethoxazole except for *Acinetobacter iwoffi* which was found to be intermediary sensitive. In the present study, kanamycin was found to be effective in controlling the present isolates because only one isolate showed resistance to it.

Most of the imported bacterial diseases from Asia have multiple antibiotic resistances and are totally immune to any of the legal treatments allowed by the U.S. Food and Drug Administration. In the U.S., more than 90% of strains are susceptible to third-generation celphalosporins (cefotaxime, ceftriaxone, ceftazidime and cefoperazone) and aminoglycosides (gentamicin, tobramycin, amikacin, sisomicin, netilmicin, kanamycin, and neomycin). Nearly all aeromonads are susceptible to quinolones (ciprofloxacin, norfloxacin, ofloxacin, levofloxacin, sparfloxacin, moxifloxacin and gatifloxacin. Most U.S. strains are susceptible to chloramphenicol, tetracycline, minocycline, doxycycline, and nitrofurantoine, but resistant to clindamycin, vancomycin, and erythromycin. Imipenem was found effective for treatment of *Aeromonas* infection.

At present, the ornamental fish trade is running out of chemotherapy options due to the emergence of resistant strains and the strict control on the licensing of antimicrobials for use in fish therapy. There seems to be no clear way to stop the emergence of resistant bacteria as long as antibiotics are used. This could be achievable however, if communication between fish health specialists and the ornamental fish trade is increased and more scientific research on chemotherapy or alternative treatments are addressed to this particular group of fish.

The subject of diseases in the aquarium trade, in general, puts the industry in a difficult position. Representatives of the pet-fish industry are frequently reluctant to talk about problems with diseases for fear that it will result in additional regulations. Privately, they however admit that diseases of their stocks during confinement in close conditions, during culture, transportation, or holding facilities are a serious problem.

Some researchers have suggested ways of dealing with the problem. One suggestion is the use of probiotic bacteria to balance bacterial populations (harmful and harmless bacteria), and the shifting of the ecological balance from resistant to susceptible bacteria by sensible chemotherapy programmes. Probiotics may protect their host from pathogens by producing metabolites which inhibit the colonisation or growth of other microorganisms or by competing with them for resources such as nutrients or space. Recent studies have found that Spirulina algae functions as a probiotic, allowing the fishes own immune system to function at a higher level of activity.

# Antibiotics use in Aquariums

Antibiotics are of limited use for treating aquarium fishes because none of the antibiotics have been originally developed for fishes, but were developed for man and farm animals. Their requirements were originally quite different than for fishes, with only some being adaptable to be used for fishes.

Aminoglycosides are toxic to fish and should be used with caution. Severe kidney lesions have been reported in goldfish treated with gentamicin. Toxicity may be exacerbated by a high ammonia concentration in the water. Aminoglycosides include amikacin, gentamicin, kanamycin, neomycin, netilmicin, paromomycin, rhodostreptomycin, streptomycin, tobramycin, and apramycin. ~ The Merck Veterinary Manual (2008).

# Amoxycillin

Furunculosis, Gill-disease 60–80 mg/kg body weight of fish/day for 10 days

## Chloramphenicol

Columnaris, Enteric Red Mouth, Fin-rot, Furunculosis, Haemorrhagic Septicaemia, Pasteurellosis, Ulcer Disease, Vibriosis

(a) 50–70 mg/kg of food/day for 5-10 days(b) 10–50 mg/L of water, as a bath

(0) 10–30 mg/L of water, as a bath

This drug should not be used for home aquaria because it is unstable in water, and poorly absorbed by target fish; and, it can cause fatal human aplastic anaemia if touched by a person who is allergic to the compound. (N. Frank, AquaVetData editor, 1998)

## Ciprofloxacin

Ciprofloxacin is a synthetic broad spectrum antibiotic that is effective against gram-negative and some gram-positive bacterial pathogens of fish. It can be used for finrot, skin lesions and other systemic disease. Because it inhibits unique target enzymes needed for bacterial replication and DNA repair, it may be effective against bacteria unresponsive to other antibiotics. Treatment is usually by immersion bath. Ciprofloxacin activity decreases with pH above 6.9. It can be bacteriostatic or bactericidal depending on the effective concentration at the target site.

# Difloxacin

Furunculosis 5 mg/kg body weight/day for 5–10 days



# Doxycycline

Streptococcosis -2 mg/kg body weight of fish/day for an unspecified duration. Used against susceptible bacteria as a prolonged immersion at a dose of 2-3 mg/L.

## Enrofloxacin

Bacterial Kidney Disease, Furunculosis 10 or 20 mg/kg bodyweight/day for 10 days

The compatibility and efficacy of enrofloxacin, an antibiotic belonging to the quinolones, was tested for five ornamental fish species (*Herotilapia multispinosa*, *Pterophyllum scalare*, *Symphysodon discus*, *Brachydanio rerio* and *Melanochromis johanni*). The results prove that enrofloxacin has a high tolerance level for ornamental fishes and high efficacy against important bacterial diseases. 30 mg/L water over 5 hours is recommended for the treatment of ornamental fishes.

## Erythromycin

Bacterial Kidney Disease, Streptococcosis (a) 25-200 mg/kg of fish/day for 4 -12 days (b) 20 mg/kg as an Injection

## Florfenicol

Florfenicol is a structural analogue of chloramphenicol similar to thiamphenicol, but with more activity against some organisms than chloramphenicol. It differs importantly from chloramphenicol in that it lacks the para-nitro group that is believed to be responsible for the problems of aplastic anaemia. The U.S. Food and Drug Administration have approved the use of florfenicol as a medication food additive for the treatment of *Flavobacterium columnare*. Recommended dosage is 10 mg of florfenicol per kg of body weight of fish for 10 consecutive days. Florfenicol is a drug that has great potential to control and reduce mortalities associated with a number of fish diseases.

## Furanace

Coldwater Disease, Columnaris, Fin-rot, Gill Disease, Haemorrhagic Septicaemia, Vibriosis (a) 2–4 mg/kg of fish/day for 3–5 days (b) 0.5–1 mg/L of water for 5–10 min, as a bath

## Kanamycin

Fin rot, Haemorrhagic Septicaemia, Mycobacteriosis, Vibriosis 50 mg/kg of fish/day for 7 days

Kanamycin has been used with some success to treat bacterial diseases of ornamental fish. It can be administered orally at 20 mg/kg, by injection at 20 mg/kg, or in a bath at a concentration of 750 mg/L for 2 hours. Anorectic fish can be medicated with a bath treatment or by injection, repeated daily, until fish begin to eat, at which time the drug can be incorporated into the feed to complete the treatment period. Treatment should be continued for 7 days beyond the alleviation of clinical signs.

Kanamycin mixed with food has been reported as effective in curing fin and tail rot among ornamental fishes. It has also been reported that Kanamycin is absorbed from water by fishes. Dose: 2-5 gm/L for 4–5 days. Afterwards make a 35–50% water change. Also mixed in the food at 200–300 mg into 100 gm food.

#### Nitrofurazone

Dose is 20 mg/L as a 5-hour bath; 100 mg/L as a 30 minute dip; Used as a prolonged immersion at 2 mg/L for 5 to 10 days. Several soluble forms exist.

## **Oxolinic Acid**

Columnaris, Enteric Redmouth Disease, Furunculosis, Haemorrhagic Septicaemia, Vibriosis (a) 10 mg/kg of fish/day for 10 days (b) 1 mg/L of water as a bath for 24 hours (c) 25 mg/L of water as a dip for 15 minutes – two times daily for three days

## **Oxytetracycline (Terramycin)**

50-75 mg/kg of fish/day for 10 days.

Terramycin is usually incorporated into the feed at 0.5 gm/100 gm food. Terramycin must be fed for 10 days to control the infection. An additional consideration when feeding Terramycin is the drug can be broken down by high temperatures, and it doesn't work very well in very hard water. As much as 95% of the efficacy of the drug is inactivated.

Sulphonamides (sulphisoxazole, sulphamerazine, sulphamethazine) 100-200 mg/kg of fish/day for 10–20 days

Sulfonamides inhibit the growth and multiplication of certain bacteria, but do not kill them. Because of their toxicity and increasing resistance of bacteria to them, sulphonamides are of limited use in fish disease control.

## Tetracycline

75-100 mg/kg of fish/day for 10-140 days

## **Feed Additives**

In the aquaculture industry, antibiotics have been added to feed as growth promoters, to treat specific diseases or as prophylactics. Investigations using chromatographic methods have determined that many artificial larval feeds, including shrimp flakes and micro-encapsulated diets, are adulterated with various antibiotics such as oxytetracycline, oxolinic acid and even chloramphenicol.



# **Volume Calculations**

Chemical treatments can be ineffective if volume is underestimated and potentially lethal if it is overestimated. Before determining the concentration or amount of chemical to be used the water volume must first be calculated. Therefore, all fishkeepers should know the volume of their aquarium or pond.

Exact measurement of volume is essential in order to calculate any chemical applications and should be calculated before a problem occurs. Ponds preferably should be calculated when they are filled with water for the first time. The information is then recorded so it is immediately available when needed. In small ponds, depth should be measured across the pond in at least two directions. The number of different directions that will be needed will depend on the shape and bottom uniformity of the pond and will have to be determined on site. If water depth is not uniform it is important that average depth is measured. Greater number of depth measurements will result in greater accuracy.

Most aquariums used for holding fish are rectangular and the volume of rectangular aquariums is calculated by the formula: Volume (litres) = length × width × depth in centimetres  $\div$  1000. When measuring a tank, take inside measurements of length and width and the depth at the appropriate water level. If the bottom of the tank is sloped, an average depth measurement should be used. To get the average depth of the tank, take three measurements: at the shallow end, in the middle and at the deep end. Add these depths together and divide the total by 3.

Circular pond or container volume is determined by the formula: Volume (litres) =  $3.14 \times \text{radius}^2 \times \text{depth}$  in centimetres  $\div 1000$ . The radius is measured as  $\frac{1}{2}$  the inside diameter of the container. The radius is squared or multiplied by itself. For example, a circular container with an inside diameter of 180 cm and depth of 60 cm has a volume of 1526.04 litres ( $3.14 \times 90 \times 90 \times 60$ )  $\div$ 1000

# Aquarium Medication Calculations

All aquarium medications must be applied at a prescribed rate. Accurate application of this prescribed rate is necessary to achieve adequate control of the target organisms, and to avoid unwanted results such as mortality of non-target organisms. Chemical application rates for aquariums are generally given as a final concentration of active ingredient in the water, usually in parts per million (ppm).

To calculate the dose rate of a chemical required in a given volume of water the formula is:

Dose rate = (required ppm  $\times$  litres of water to be treated)  $\div$  percent of active ingredient.

[1 ppm = 1 mg/L; 1 million milligrams = 1 litre; 1000 milligrams = 1 gram; 1000 grams = 1 litre] The easiest way to find out the total amount of chemical required is to convert the rates into something understandable like milligrams or grams. For example, to calculate the dose rate of 25 ppm (mg/L) of a chemical with an active ingredient of 400 grams/litre in 100 litres of water:

25 ppm x by total litres to be treated =  $25 \text{ mg} \times 100 \text{ litres} = 2500 \text{ mg}$ . 2500 mg divided by percent (40%) of active ingredient =  $2500 \div 0.40 = 6250 \text{ mg}$ .

## Example:

- $= (25 \text{ ppm} \times 100 \text{ litres}) \div 40\%$
- $= (25 \text{ mg} \times 100 \text{ litres}) \div 40\%$
- $= 2500 \text{ mg} \div 0.40$
- = 6250 mg or 6.25 grams.



# Miscellaneous (parts per million and percent)

0.0038 grams per US gallon = 1 ppm 1 milligram per litre = 1 ppm 0.001 gram per litre = 1 ppm

ppm = mg/L 1 mg/L = 1,000 milligrams per litre

1 Percent (%) = 10,000 parts per million (ppm) = 10 grams per litre (g/L)

How to convert ppm into percent: Percentage is parts per 100. 1 part per hundred is 10 parts per thousand or 10,000 parts per million. So to get from ppm to percentage you have to divide by 10,000.

# Percent <--> Grams/Litre Conversions

This is a simple conversion. Since percent is parts per hundred, and grams/litre is parts per thousand (ppt), we simply need to multiply percent by 10 to get grams/litre, or: grams/litre = 10 percent

grams per litre g/L = pptmilligrams per litre mg/L = ppm

The two most common elements in sea water, after oxygen and hydrogen, are sodium and chloride. Sodium and chloride combine to form what we know as table salt. Sea water salinity is expressed as a ratio of salt (in grams) to litre of water. In sea water there is typically close to 35 grams of dissolved salts in each litre. It is written as 35%. The normal range of ocean salinity ranges between 33-37 grams per litre (33% - 37‰).

# Converting ppm to ppt:

To convert ppm readings to ppt, divide the ppm reading by 1000. For example a reading of 5000 ppm = 5000 ppm/1000 = 5.00 ppt.

To convert ppt readings to ppm, multiply the ppt reading by 1000. For example a reading of  $4.00 \text{ ppt} = 4.00 \text{ ppt} \times 1000 = 4000 \text{ ppm}$ .

## Converting µS to mS:

To convert  $\mu$ S readings to mS, divide the  $\mu$ S reading by 1000. For example a reading of 5000  $\mu$ S = 5000  $\mu$ S/1000 = 5.00 mS.

To convert mS readings to  $\mu$ S, multiply the mS reading by 1000. For example a reading of 4.00 mS = 4.00 mS x 1000 = 4000  $\mu$ S.

# Units of Concentrations

Parts per million: Assuming the density of water is 1.00 g/mL, 1 litre of solution = 1 kg and hence, 1 mg/L = 1 ppm. This is generally true for freshwater and other dilute aqueous solutions.

ppt-parts per thousand (used for common ions in sea water)

 $ppm = mg/L = \mu g/mL$ 



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