17

Research on Culturing the Early Life Stages of Marine Ornamental Fish

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Introduction

The number of marine ornamental species that can be economically produced on commercial farms today is extremely limited. The future of marine ornamental fish farming, like marine food fish culture, depends on the ability to reliably produce eggs, raise large numbers of larvae, and transition them to juveniles. A large number of marine ornamental fish and invertebrate species have been spawned in captivity. Some species spawn naturally in large aquariums, and others have been induced to spawn by photoperiod and temperature cycles (Holt and Riley 2001) or by the use of hormones (Moe 1997). Many more species have been spawned in captivity than have been reared. The early life stages remain the critical bottleneck in the production of most marine ornamentals. Among the priority issues in hatchery technology are designing rearing systems that provide acceptable environmental conditions and identifying suitable prey for different stages during ontogenetic development.

Rearing Systems

Some major concerns in rearing the young are light, space, and water quality. The environmental conditions for raising larvae should mimic their natural planktonic habitat. This means very stable conditions of high salinity and oxygen concentrations, low nutrients, basic pH, light cycles of 12 hours light:12 hours dark, and warm temperatures. Some of these conditions are difficult to maintain once feeding is initiated because dense concentrations of live prey change the quality of the water. For example, high concentrations of algae deplete oxygen during the night and can alter the pH of the system, while large numbers of zooplankton added for food use up oxygen and add metabolites.

Creative designs are needed to maintain high-quality rearing water and at the same time avoid damaging the fragile larvae. Some systems that have overcome many of these problems use microcosms (Henny et al. 1995; Palmtag and Holt 2001), algae scrubbers, or flow-through water, but the last is highly location dependent. The author uses the microcosm approach, placing larvae and their prey (including algae) inside 10- to 18-liter baskets that are in turn placed in larger tanks (300–500 liters) connected to an external filter. Heating and aeration are carried out in the large tank, and water is slowly dripped into the baskets to replace the rearing chamber water several times a day. Mesh on the basket is small enough to retain prey. These types of rearing chambers have been used to rear a large number of fish and shrimp larvae at our lab (Table 17.1).

Feeding and Nutrition

A critical bottleneck continues to occur at first feeding, when larvae change over from internal yolk stores to exogenous feeds. Many ornamental fish (e.g. Chaetidontidae, Cirritidae, Serranidae, Labridae) spawn small pelagic eggs that hatch into small larvae with narrow mouth gapes. Rotifers and brine shrimp (***Arternia*** sp) are the most widely used live food items in marine fish culture, but they are not always acceptable food. First-feeding marine larvae (<3 mm standard length) feed on a wide variety of micro-zooplankton including protozoans (tintinids, ciliates, foraminiferans), dinoflagellates, larvae of barnacles and mollusks, and copepod eggs and nauplii (Holt and Holt 2000; Riley and Holt 1993). Diatoms occur in the diets as well,

Table 17.1 Marine ornamental fish and shrimp spawned in captivity at UTMSI, their early life characteristics and reproductive strategy

Closed life cycle	
Species	Characteristics
Lined seahorse *(Hippocampus erectus)*	Egg brooder, hatch as juveniles (g)
Jackknife fish *(Equetus lanceolatus)*	Pelagic eggs (1.O) hatch at 2.7 mm (g)
Cubbyu *(Equatus umbrosus)*	Pelagic eggs (1.2), hatch at 2.8 mm *(g)*
Comet *(Calloplesiops altivelis)*	Attached eggs, hatch at 2.7 mm (pg)
Fire shrimp *(Lysnzata debelius)*	Egg brooder, larval duration 75-158 days (sh)
Peppermint shrimp *(Lysmata wurdemanni)*	Egg brooder, larval duration 30-65 days (sh)
Spawned but life cycle not closed	
Species	Characteristics
Harlequin bass *(Halichores maculipinna)*	Pelagic eggs (0.75) hatch at 2.0 mm (sh)
Longnose hawkfish *Oxycirrhitus typus)*	Pelagic eggs (0.75) hatch at - 2 mm (pg)
Pygmy Angelfish *(Centropyge argi)*	Pelagic eggs (0.73) hatch at 1.2 mm (pg)
Lemonpeel *(Centropygejavissirnus)*	Pelagic eggs (0.71) hatch at 2.3 mm (pg)
Bluehead wrasse *(Thalassoma bifasciatunz)*	Pelagic eggs (0.56) hatch at 1.4 mm (pg)
Clown wrasse *(Halichores maculipinna)*	Pelagic eggs (0.59) hatch at 1.5 mm (pg)
Cuban hogfish[a] *(Bodianus pulchellus)*	Pelagic eggs (0.85) hatch at 2.2 mm (pg)
Firefish *(Nemateleotris magnijca)*	Attached eggs, hatch at 2.0 mm (pg)
Scarlet cleaner shrimp[b] *(Lysmata anzboinensis)*	Egg brooder, larval duration 180+ days (sh)

Note: Egg diameters at spawning are in millemeters in parentheses; (g), gonochoristic (separate sexes); (pg), protogynous hermaphrodite (female to male sex change); (sh), simultaneous hermaphrodite.

[a] Larvae reared to 21 days.

[b] Larvae reared to 6 months.

ranging from 2% of the total items in larval guts of temperate fish to 5% in tropical reef fish. The size of micro-zooplankton in the guts of small larvae varies from 3 to 100 ym, with the majority smaller than 60 ym. Wild zooplankton has been used to rear marine ornamentals (Danilowicz and Brown 1992), but it does not always provide a consistent quantity of prey items on a regular basis. The author reared *Bodianus pulchellus* on wild zooplankton but was not able to identify the prey consumed by the larvae; predators were often introduced into the rearing chambers (Holt, unpublished). Collecting specific organisms from plankton at times or places where appropriate prey is available has proven successful.

The nutritional requirements for long-chain n-3 highly unsaturated fatty acids (HUFA) for the normal growth and development for marine fish larvae are well established (Sargent et al. 1999; Watanabe 1982). Marine copepods typically have high levels of n-3 HUFA that reflect the fatty acid composition of their diet. These HUFA are transferred to marine larvae through feeding. There is a fundamental need for new and more nutritious prey species for rearing marine larvae that contain these essential nutrients. Although copepod eggs, nauplii, and copepedites are the natural food of young marine fish larvae, they have not generally been used extensively in aquaculture because they are difficult to culture on a continuous basis, and natural populations vary in abundance and size distribution. Various species of calanoid and harpticoid copepods have been raised in ponds and under controlled conditions, but it is thought that production is not high enough to sustain marine ornamental larviculture. Techniques have been recently described that reliably produce an intensive culture of a temperate water estuarine calanoid copepod *Gladioferens imparipes* (Payne and Rippingale 2001). They achieved 850 nauplii per liter per day for over 420 days in automated 500-liter culture chambers.

A recent breakthrough was the development of a protocol for producing resting stage or diapause eggs of the copepod *Centropages hamatus* (Marcus and Murray 2001). Initial tests of

the eggs as food for marine fish larvae were encouraging Larval comet Calloplesiops altivelis that were fed the copepod nauplii grew larger and survived better than larvae fed rotifers and brine shrimp (Table 17.2). Larvae grown on a mixture of wild zooplankton, rotifers, and brine shrimp were also smaller than the copepod-fed larvae. Copepod nauplii hatched from Centropages hamatus diapause eggs hold promise as an alternative live food for tropical ornamental fish. The next step needed is to develop a large-scale production protocol for these copepods, probably in collaboration with industry.

There has been considerable research on the fatty acid requirements of marine larvae, especially in temperate species that are of interest for aquaculture. Emphasis has been on ways to enrich live foods that do not contain adequate fatty acid profiles. It has been suggested that marine larvae require approximately 10% (of diet dry weight) as n-3 HUFA-rich phospholipids in their food, with optimum dietary levels of certain HUFA: docosahexaenoic acid (DHA) to eicosapentaenoic acid (EPA) ratios of 2:1 and DHA to aracadonic acid ratios of 10:1 (Sargent et al. 1999). Since rotifers and brine shrimp do not have this pattern of fatty acids, they can be enriched to increase the concentration of HUFA, but it is almost impossible to reach the desired HUFA ratios in brine shrimp. Many species of seahorses cannot be raised successfully on a diet of enriched brine shrimp unless they are fed copepods for the first few days of feeding (Payne and Rippengale 2000; Gardner 2001; Kucera and Holt, Hippocampus erectus, unpublished).

The ultimate goal for larviculture would be to develop artificial, inert diets as feed to replace live zooplankton. Several species of marine fish larvae have been successfully weaned to microdiets by cofeeding them with live prey for some length of time. Red drum Sciaenops ocellatus larvae have been successfully raised with only an inert diet and the algae Isocrysis galabana from first feeding (Lazo et al. 2000). Microdiets can be designed to provide all the nutritional requirements and energy needs of the developing larvae. So far this concept has seldom been applied to ornamental species. Two species of reef sciaenids, cubbyu (Pareques umbrosus) and jackknife fish (Equetus laceceolatus have been weaned onto microdiets at 2 weeks by cofeeding artificial diets with rotifers and brine shrimp. The major problem encountered in these studies was maintaining high water quality since some of the diet is uneaten and falls to the bottom of the culture chamber. With careful attention to biofiltration and occasional vacuuming and water replacement, recirculating systems can successfully be used to wean larvae. **A** great deal of research is needed before artificial diets will be of use in marine ornamental culture.

Table 17.2 Standard length and standard deviation (in parentheses) of comet *Calloplesiops altivelis* larvae raised on rotifers and brine shrimp (*Artemia* sp.), copepod nauplii, or a mixture of wild zooplankton, rotifers, and brine shrimp

Age	Rotifers + brine shrimp	Copepods	Zooplankton + rotifers + brine shrimp
Day 3	3.5	3.5	3.5
Day 7	3.97 (.177)	4.79 (0.115)	—
Day 14	3.83 (—)[a]	5.05 (0.297)	4.22 (0.469)

[a] Larvae survived to day 14 in only one of the replicates.

Uses of Cultured Ornamentals

There are many incentives for culturing marine ornamentals. Cultured organisms are positioned to play a more significant role in supplying the marine ornamental trade, but this is not a panacea. Research is needed to increase species diversity and availability and to establish a sound scientific background for culture. Open transfer of data and information would increase successful larviculture. It has been suggested that cultured animals could be useful for stock enhancement to decrease the recovery time of depleted or overfished populations (Zeimann 2001). Culture alternatives are listed in most conservation plans. Culturing fish in tropical regions by native islanders could provide alternative livelihoods as well as increased interest in conserving coral reef resources.

Moreover, culturing fish can provide valuable life history details such as size and age at spawning, measures of fecundity, morphological development, age at first feeding, stage durations, and growth rates. These life history traits are unknown for a large majority of fishes of the world, yet this information is critical for understanding fish population dynamics.

Knowledge of these vital rates is necessary for management of coral reefs and designing refuges and marine reserves. In addition, data on ontogenetic changes in larval feeding and physiological responses to environmental parameters would be invaluable for interpreting the response of coral reefs to human impacts. Thus, increased understanding of early life stages at all levels is crucial to understanding and mitigating anthropogenic impacts. Much of this information is available only through the controlled culture of marine ornamentals.

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