

Final Report

Pacific Aquaculture Association

Culture of Tropical Marine Aquarium Fishes

By: Jeffrey A. Tellock

TECHNICAL REPORT NO. 020

JUNE 1996

TECHNICAL REPORT SERIES
OF THE
GUAM AQUACULTURE DEVELOPMENT AND TRAINING CENTER

Department of Commerce
Government of Guam
102 M. Street
Tiyon, Guam 96913



Sea Grant
Pacific

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PACIFIC AQUACULTURE ASSOCIATION

PROJECT REPORT

FINAL REPORT

PROJECT TITLE

Culture of Tropical Marine Aquarium Fishes

PROJECT PERIOD

September 1994 to March 1996

PRINCIPAL INVESTIGATOR

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SUMMARY OF FINDINGS

Many new species of marine tropical fish and shrimp have been cultured in recent years and some of these are beginning to be produced at a commercial level. However, clownfish are still the main product of marine aquarium fish hatcheries. Commercial efforts are becoming more successful from a biological standpoint, but some are still struggling financially to make this type of aquaculture profitable. Some commercial marine tropical fish hatcheries have discontinued operations after several years of activity, while others have started up in recent years or may start up in the near future.

Research activity in this area is also occurring at the university level. Literature on the culture of marine tropical fish and shrimp is increasing, but occurs mainly in the aquarium hobby literature, rather than the scientific literature.

A list of key marine aquarium fish and shrimp available in Micronesia and conducive for mariculture was compiled, and some of these species are particularly promising for commercial culture. Culture methods were examined for the Cinnamon clownfish *A.*

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melanopus, the gobies *Ptereleotris zebra* and *Valenciennesia strigatus*, and the Banded coral shrimp *Stenopus hispidus*. Several batches of *A. melanopus* were successfully reared to the juvenile stage. Regular spawning was obtained for the goby broodstock, but no larvae were reared beyond the first-feeding stage for either of the goby species. Larvae of *S. hispidus* were reared through the early larval stages, but none survived the long larval period to become juveniles.

Problems encountered in the culture of clownfish were mainly in the production of eggs. Successful larval culture of the goby larvae was not achieved due to a lack of a suitable first food for these tiny larvae. Progress was made in the larval culture of the Banded coral shrimp, but eventual contamination of larval tanks by hydroids and jellyfish medusae resulted in complete mortality before the larvae reached the juvenile stage. Suggestions are presented to overcome all of these obstacles in the rearing of these and other species of marine tropical fish and shrimp.

A growing trend of interest to commercial aquarium fish producers is the increase of restrictions on the importation and exportation of marine tropical species, with some island nations closing down or severely restricting exports of reef fishes and invertebrates. Guam is preparing to prohibit the export of marine fish and invertebrates from its reefs. This may be a growing trend among island nations as they seek to protect their resources. Some countries are also beginning to restrict imports of marine tropical fish and invertebrates.

An economic analysis of marine aquarium fish culture suggests that commercial culture of various marine ornamental species could be profitable in Micronesia. The most important factors affecting profitability are consistent production of marketable fish, market price and sales volume. Feed costs are not a major factor affecting economic return, although the unit cost of feed for marine ornamental fish is much higher than commercial diets for foodfish. Economic projections were made for a small hatchery and grow-out facility raising clownfish.

PROJECT OBJECTIVES

The project was designed to meet the following objectives:

1. Identify current private and government efforts to culture tropical marine aquarium fishes throughout the world.
2. Conduct applicable literature review for the culture of marine tropical fishes.
3. Identification and listing of key marine aquarium fishes readily available on Micronesian reefs and lagoons that are conducive for mariculture application.
4. Choose target fish species and explore culture methods.

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5. Identify current and anticipated international regulatory constraints to the import/export of various marine aquarium fishes.
6. Determine methods of mariculture certification of restricted species.
7. Identify/determine economic parameters that would make this new form of mariculture feasible on a commercial scale; from culture/grow out to export.

METHODS:

Objective 1. Private and government efforts to culture tropical marine aquarium fishes were identified through literature review and telephone contacts. In addition, database reports were obtained from the Breeder's Registry, a newsletter and database providing information on breeding and propagation of marine aquarium fish and invertebrates. A video from a conference on marine tropical fish culture and another video about a small commercial marine aquarium fish hatchery also provided information on previous efforts at propagating marine tropical fish.

Objective 2. A review of the literature was conducted for the culture of marine tropical aquarium fish and invertebrates throughout the study. A broader review of the literature on culture methods for other marine fish was conducted in the later stages of the study.

Objective 3. A list of key marine aquarium fishes available in Micronesia and conducive for mariculture application was created, using the following criteria: availability, popularity in the aquarium trade, wholesale or retail value, known adult and larval characteristics (e.g., size of spawners, spawn size, egg and larval size, larval duration), available information on spawning and larval rearing, reports of successful culture efforts, and ease of culture of related species. Not all the species in the list qualified favorably under every criteria, but each species qualified under at least some of the criteria and show potential for eventual commercial culture. Scientific and common names were checked and verified through two guide books on marine aquarium fishes (Burgess *et al.*, 1990; Myers, 1991).

Objective 4. The following marine aquarium species were obtained from local collectors for possible use in this project:

Species	Common name	Number
<i>Amphiprion chrysopterus</i>	Blue-stripe clownfish	3 pairs
<i>Amphiprion clarkii</i>	Clark's clownfish	2 pairs
<i>Amphiprion melanopus</i>	Cinnamon clownfish	30 fish
<i>Amphiprion perideraion</i>	Pink skunk clownfish	1 pair
<i>Ptereleotris zebra</i>	Zebra goby	1 pair
<i>Valenciennesa strigatus</i>	Blue-streak goby	1 pair
<i>Stenopus hispidus</i> *	Banded coral shrimp	8 pairs

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* Although this project originally targeted aquarium fish only, it was later expanded to include the Banded coral shrimp.

The majority of the fish and three pairs of the Banded coral shrimp were collected during the first six months of the project. The Cinnamon clownfish, *A. melanopus*, were collected as three groups of fish. Initial efforts were made to pair up the clownfish by putting together one large and one smaller fish. Several individuals were also cannulated to identify male and female fish and determine the level of maturation. Another group of Cinnamon clownfish collected later in the study was placed together with several other unpaired clownfish in a large (10-ton) fiberglass tank containing several spawning sites (i.e., bricks and tiles) and anemones. The other species of clownfish and gobies were collected as mated pairs. These fish were not cannulated to determine the level of maturation, so as not to risk damaging their reproductive tracts.

Various sizes of fiberglass tanks at the Guam Aquaculture Development and Training Center (GADTC) were used for holding clownfish broodstock, ranging from about 400 liters to 2000 liters. Some of the fish were held inside the hatchery and others were held outdoors. Ceramic tiles were placed in the tanks containing clownfish for use as spawning substrates. The tiles were examined daily for eggs, usually in the morning. Pairs of Banded coral shrimp were also placed in three of the broodstock holding tanks. Later in the study, five additional pairs were collected and all the Banded coral shrimp were moved to two shallow culture trays inside the hatchery, with each pair placed in a separate compartment (Figure 1). Water quality was maintained in all the broodstock tanks and grow out tanks by a continuous flow of seawater through the tanks. The seawater at the GADTC is obtained from saltwater wells, without any additional treatment or filtration. Indoor broodstock tanks received natural sunlight which came into the hatchery.

The pair of Blue-streak gobies, *V. strigatus*, were maintained in a 3000 liter tank inside the hatchery. The tank was provided with a shallow tray filled with coral sand and short pieces of PVC pipe stuck into the sand for a spawning site (Figure 2). The PVC pipe was checked on a regular basis for the presence of eggs. The behavior of the fish was also observed for indications of spawning. The pair of Zebra gobies, *P. zebra*, were maintained outdoors in a 1000 liter tank. Giant clams shells were provided on the bottom of the tank, forming small caves that the fish could go inside and use for a spawning site. The clam shell was checked on a daily basis for the presence of eggs.

The broodstock were primarily fed a frozen diet prepared at the GADTC. The recipe for this diet was similar to a gelatin diet used at the Waikiki Aquarium, with some modifications. Ingredients used in the diet and instructions for preparing it are listed in Appendix 1. The composition of the diet varied slightly from batch to batch, depending on local availability of ingredients. The broodstock were usually fed *ad libitum* twice daily with the frozen food. In the beginning of the study, this diet was occasionally supplemented with live adult *Artemia* or small penaeid postlarvae. Later in the study, brine

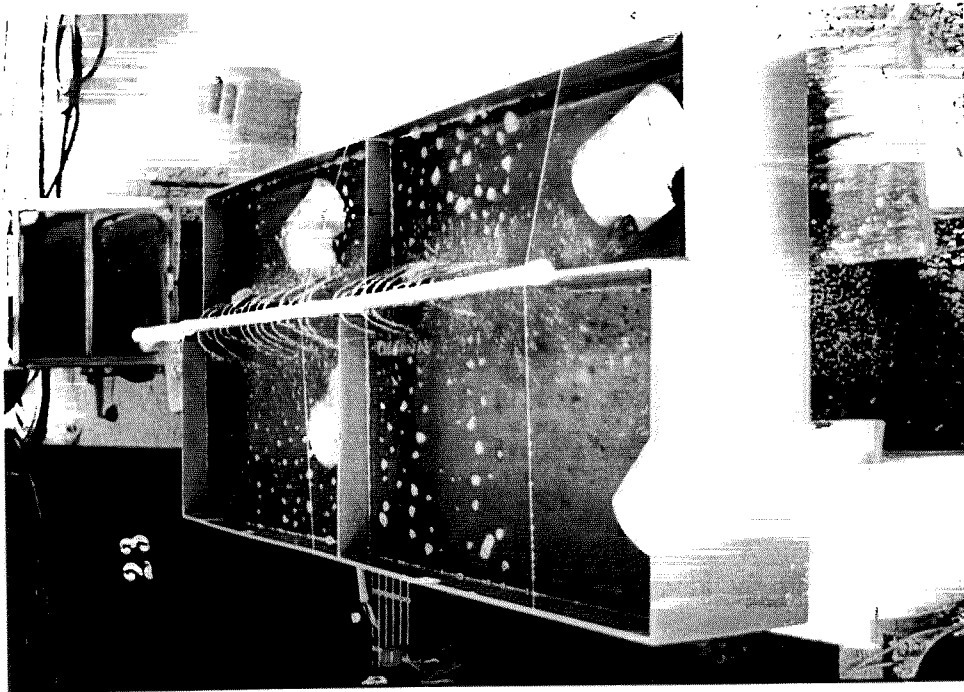


Figure 1. Banded coral shrimp broodstock tanks. It was determined that these 20 and 30 liter tanks were adequate to maintain spawning pairs of Banded coral shrimp.



Figure 2. Blue-streak goby broodstock and pvc pipe spawning site.

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shrimp flakes were occasionally used as a supplemental feed. The banded coral shrimp were also fed a shrimp maturation pellet on a regular basis in addition to the frozen food.

Clownfish egg development was monitored visually and by length of incubation period. Previous experience and reports in the literature indicated that the egg incubation period of clownfish is usually between six and nine days, depending on temperature and condition of the broodstock. Late in the afternoon on the day when a batch of eggs appeared to be fully developed and ready to hatch, the tile on which the eggs were laid was removed from the broodstock tank and put into a larval tank. The tile was placed on edge and leaning against the inner standpipe, with an airstone placed near the egg mass, so that a stream of air bubbles passed close to the egg mass. The following day after the eggs hatched, the tile was removed and returned to the broodstock tank. A few spawns were deposited on the side of the broodstock tank or hatched earlier than expected. In these cases, it was necessary to remove the larvae from the broodstock tank after hatching. Newly-hatched clownfish larvae are positively phototactic and were concentrated with a light and siphoned into a bucket of seawater.

Banded coral shrimp egg development was monitored regularly based on the appearance of the egg mass. New egg masses are a blue-green color and become a creamy white as they develop. When the eggs are ready to hatch, dark eyespots can be seen in the white eggs. A female with mature eggs was placed into a bucket of clean seawater with an airstone in the afternoon preceding hatching. The next morning, the female was returned to its holding tank and larvae were counted and stocked into a larval rearing tank.

Spawn size in clownfish was estimated by counting the "egg spots" on the spawning site after hatching. These egg spots usually appeared as lighter colored spots on the tile where the eggs were attached. Spawn size was estimated in Banded coral shrimp by counting the number of larvae in several 6-ml samples taken from a bucket containing the hatched larvae, calculating an average number of larvae per ml, and multiplying that number by the total volume of the bucket.

Three types of live foods were examined in larval rearing trials, including rotifers (both s-type and ss-type), sea urchin larvae and oyster trocophores. Rotifers were mass cultured outdoors in batch culture as described by Tamaru et al. (1993). The GADTC originally cultured the s-strain of rotifer (*Brachionus plicatilis*), but in April 1995 production was switched over to the ss-strain of rotifer (*B. rotundus*).

Sea urchins (*Echinometra mathaei*) were collected from local waters and maintained in outdoor tanks, where they grazed upon the algae growing on the sides of the tanks. Fertilized sea urchin eggs were obtained by induced spawning after injecting approximately 0.5 ml of a 0.5M KCl solution into the body cavity of the sea urchins using a 1 cc tuberculin syringe. The injected individual was then placed upside down in a 250 ml glass beaker filled with seawater and observed for the release of gametes. Males and females were identified by the release of sperm or eggs, respectively. Males were removed

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petri dish so that milt could be collected without mixing with sea water. Females were allowed to spawn until eggs no longer were released. The spawned eggs were collectively placed into a small beaker containing seawater. The sperm was activated with seawater and then a few ml of diluted sperm was added into the container of spawned eggs and mixed to fertilize the eggs. The eggs were then sieved through a 150 micron screen to remove large debris and placed into a 3-liter pitcher of seawater and aerated until used. Tamaru and Carlstrom-Trick (1995) provide additional details of the spawning procedure and developmental sequence of the sea urchin larvae used in this study.

Oysters (*Saccostrea cucullata tuberculata* and/or *Crassostrea echinata*) were collected in Sasa Bay, Guam and held in tanks at the GADTC. The adult oysters were occasionally fed algae cultured at the GADTC. Some of the oysters were held in tanks containing juvenile mullet or tilapia. Oyster trochophores were obtained after natural spawning (probably following handling stress) or by placing them in an aquarium and adding approximately four ppm hydrogen peroxide (Figures 3 and 4). Oyster trochophores were harvested with a 23 micron screen about 10 to 12 hours after spawning and added to the larval tank.

Clownfish larvae were reared in 500 liter black fiberglass tanks. Larval tanks were initially filled to about 250 to 300 liters volume with seawater and provided with light aeration. Several liters of a dense culture of microalgae (*Chaetoceros gracilis* or *Nannochloropsis oculata*) was added to the larval tanks daily during the first two weeks of the larval period to provide food for the rotifers and to help maintain water quality. The amount of algae added (usually 5% to 10% of the tank volume) depended on the density of the algae culture, the amount of algae already present in the larval tank, and the volume of the larval tank. *Artemia* nauplii was added to the larval tanks, beginning about seven days posthatch. Artificial diets (i.e., ground up artemia flake, larval fish diets, or frozen food) were usually added beginning at about day 12 to 15 posthatch. Daily water exchanges were initiated after the first week, and a continuous flow of water was started after the first two weeks to maintain water quality.

Juvenile clownfish were grown out in fiberglass tanks of 1000 to 3000 liters volume both indoors and outside. Juveniles were fed twice daily with the frozen diet prepared at the hatchery. Occasionally, this diet was supplemented with brine shrimp flakes. Fish were occasionally hand-sorted by size and marketable fish selected for sale to a local retail pet store or to a commercial fish collector for export.

Several larval rearing trials were conducted with the Banded coral shrimp. In the beginning of most of the trials, rotifers were maintained in the larval tank at a density of 10 to 20 rotifers/ml and the green algae *Nannochloropsis oculata* was added daily to maintain a concentration of about 500,000 to 1,000,000 cells/ml in the larval tank. After the first two to three weeks, the diet was gradually changed from rotifers to *Artemia* nauplii and eventually enriched 24-hour old *Artemia*. Daily water exchanges were begun after about the first week, and a slow continuous flow of seawater was introduced to the larval tank after about three weeks.

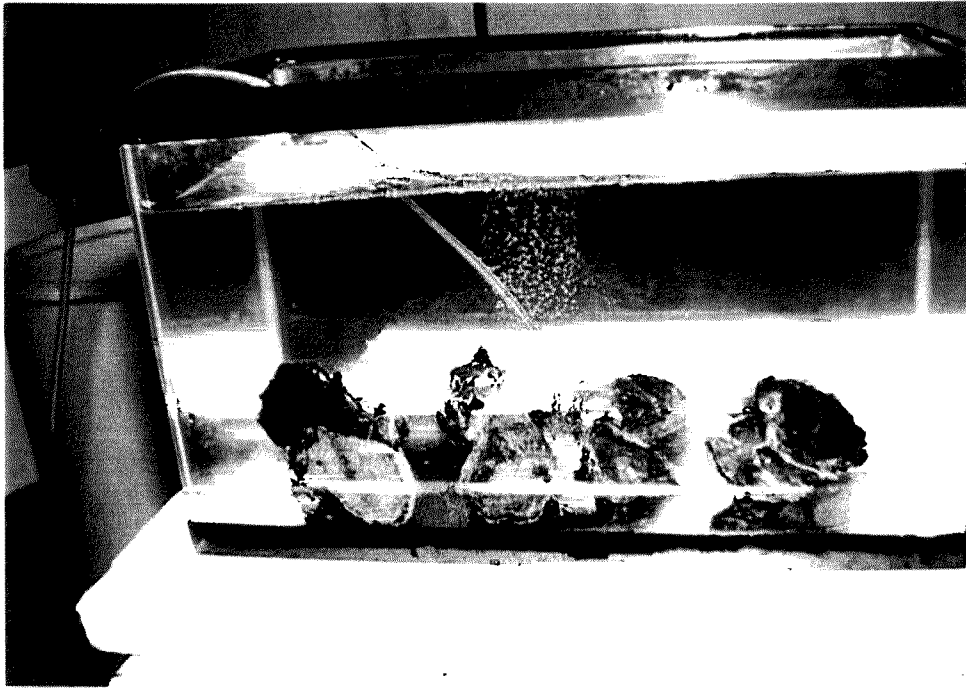


Figure 3. Oyster spawning tank before the release of gametes.

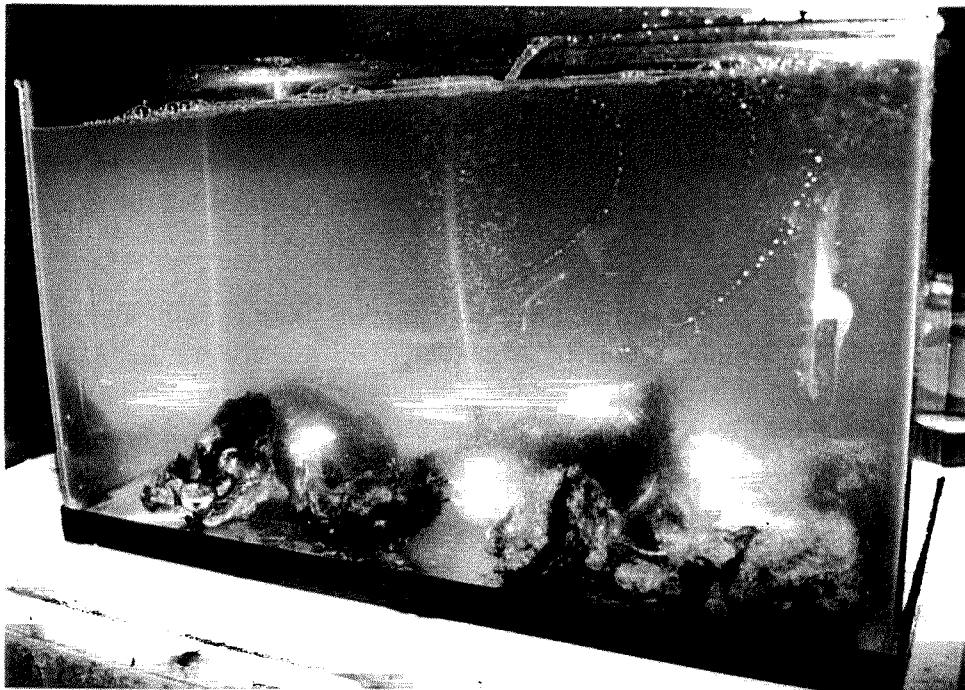


Figure 4. Oyster spawning tank after the release of gametes into the water. This spawn produced viable oyster larvae.

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Objective 5: Regulatory constraints were investigated by personal communications with Gerry Davis, Fisheries Supervisor at the Division of Aquatic and Wildlife Resources (DAWR), Guam Department of Agriculture, Robert Meyer (formerly with the DAWR), and Richard Pyle (Bishop Museum, Honolulu, HI).

Objective 6: Methods of mariculture certification were investigated by personal communications with Gerry Davis of the DAWR and by review of the literature and other mariculture operations.

Objective 7: Cash flow projections were made for a small clownfish hatchery and grow-out facility.

RESULTS:

Objective 1. A few hatcheries are known to be currently producing marine aquarium fish commercially. The largest hatchery is C-Quest, Inc. in Puerto Rico. Another is Reef Propagations, a small basement hatchery in Chicago. A third company is Desert Fisheries in Utah. A fourth hatchery (believed to be the Tropical Marine Centre Ltd.) is located in England. Three other hatcheries are presently operating at a research and development or pilot stage and are supplying small numbers of tank-raised fish to the marine aquarium market. These include a hatchery owned by Red Sea Fish pHarm, Ltd. in Israel, and two closed system hatcheries in the United States owned by SeaPhiz. Four commercial hatcheries formerly produced marine aquarium fish, but are no longer in operation. These include Aqualife Research Corporation in the Bahamas, Dynasty Marine Associates in the Florida Keys, Aquaculture Development A/S in Denmark, and Instant Ocean Hatchery in central Florida.

C-Quest, Inc. completed construction of a 15,000 ft² marine fish hatchery in 1991. The hatchery contains twelve hundred 40 gallon aquariums used for broodstock, larval rearing, and research. The facilities have since been expanded to include four hundred 300 gallon outdoor larval rearing and grow-out tanks, nearly doubling the size of the original hatchery. In addition, there are four 25,000 gallon concrete ponds used for large-scale culture of microalgae and grow-out of brine shrimp. Filtration includes biofilters, sand filters, cartridge filters, diatomaceous earth filters, and UV sterilizers.

Commercial production efforts initially focused on clownfish, but have since included two species of goby and three species of dottyback. Several other species have also been cultured at the C-Quest hatchery. The list of species cultured by C-Quest includes the following:

Species	Common Name
<i>Amblygobius rainfordi</i> *	Rainford's goby
<i>Amphiprion akallopisis</i>	Skunk clownfish
<i>Amphiprion akandynos</i>	Barrier Reef clownfish

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<i>Amphiprion bicinctus</i>	Two-banded clownfish
<i>Amphiprion clarkii</i>	Clark's clownfish
<i>Amphiprion ephippium</i>	Red saddleback clownfish
<i>Amphiprion frenatus</i>	Tomato clownfish
<i>Amphiprion melanopus</i>	Cinnamon clownfish
<i>Amphiprion ocellaris</i>	Common clownfish
<i>Amphiprion percula</i>	Percula clownfish
<i>Amphiprion perideraion</i>	Pink skunk clownfish
<i>Amphiprion sandaracinos</i>	Orange skunk clownfish
<i>Calloplelesops altivelis</i> **	Comet
<i>Chrysiptera cyanea</i> **	Orange-tail Damsel
<i>Gobiodon citrinus</i>	Citron goby
<i>Gobiodon okinawae</i> *	Okinawa goby
<i>Gobiosoma evelynae</i>	Sharknosed goby
<i>Gobiosoma genie</i>	Genie's cleaning goby
<i>Gobiosoma macrodon</i>	Tiger goby
<i>Gobiosoma oceanops</i>	Neon goby
<i>Gramma loreto</i> *	Royal gramma
<i>Gramma melacara</i> *	Black cap basslet
<i>Ophistognathus aurifons</i> *	Yellowheaded jawfish
<i>Premnas biaculeatus</i>	Maroon clownfish
<i>Pseudochromis dutoiti</i>	Neon dottyback
<i>Pseudochromis flavivertex</i>	Sunrise dottyback
<i>Pseudochromis fridmani</i>	Orchid dottyback
<i>Pseudochromis paccagnellae</i> *	Royal dottyback or Bicolor dottyback
<i>Pseudochromis porphyreus</i> *	Magenta dottyback
<i>Pseudochromis springeri</i> *	Springer's dottyback
<i>Pterosynchitropus splendidus</i> **	Mandarinfish

* Occasionally available.

** Not likely to be commercially available
in the near future.

In addition to the species listed above, C-Quest has produced one hybrid variety of clownfish that has resulted from five accidental pairings of *Amphiprion ocellaris* and *A. percula*. The hybrids were reared without problems and are called "Percularis" clownfish. C-Quest has also developed or cultured several color varieties or strains of some of the clownfish species. A bright orange-red *A. percula* and a fast developing dark strain called Early black *A. percula* have been developed. A strain of *A. clarkii* called White-backed *A. clarkii* contains a localized albino area which resembles a white saddle on the fish's back. Another strain called Heart-sleeved tomato clownfish is an *A. frenatus* with a white dot at the base of one or both pectoral fins. C-Quest also produces both white-striped (normal coloration) and blue-striped Cinnamon clownfish (*A. melanopus*), and white-striped (normal coloration) and yellow-striped Maroon clownfish (*Premnas biaculeatus*).

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C-Quest is now the largest commercial hatchery for marine aquarium fish in the world. The company is able to sell everything it produces, and is in the process of developing an arrangement to market its fish through a large chain of pet stores in California called Pets Mart (M. Moe, per. comm., 1996). Pets Mart plans to market only tank-raised specimens of marine organisms, instead of wild-caught animals. C-Quest has about 50 wholesale customers and sells about 10,000 fish per month. Dottybacks (*Pseudochromis* sp.) account for only about 150 fish per month, although the company has been getting \$15 to \$20 per fish (W. Addison, per. comm., 1996). However, wild-caught dottybacks have started becoming more available as new Red Sea collecting stations have recently opened, and the price of these wild-caught dottybacks is much lower than the tank-raised fish. As a result, C-Quest is beginning to have some difficulty in maintaining its price for the dottybacks. The exception is *Pseudochromis dutoiti*, which has not been collected in large numbers from the wild at this time (W. Addison, per. comm., 1996). The owner of C-Quest, Bill Addison, is planning to go to South Africa in the near future to investigate the possibility of a joint venture with an investor there who claims to have successfully cultured Clown triggerfish, Emperor angelfish, Moorish idols, and Powder blue tangs.

Reef Propagations, founded by Joe Lichtenbert in April 1990, is a small (400 square foot) basement operation located in the Chicago, IL area. The company began producing their first salable fish in October 1990, and by the end of 1995 had sold about 50,000 fish for more than \$150,000. About one third of these fish were purchased from C-Quest and resold by Reef Propagations (J. Lichtenbert, per. comm., 1996). Sales per week averaged 70 fish for the first two years and rose steadily to over 250 per week (Hoff, In Prep.). Production has recently declined due to technical problems. Total cash outlay at the beginning of 1995 was about \$53,000 of which about \$10,000 was allocated for research and development (Hoff, In Prep.). This operation now has a positive operating revenue and is operated by 1.5 people with a 45 hour work week.

A company in Utah called Desert Fisheries is using marine geothermal water to raise several species of marine aquarium fish. The company was started in early 1994 and is commercially producing several species of clownfish, including *Amphiprion frenatus*, *A. melanopus*, *A. ocellaris*, and *A. percula*. Desert Fisheries has also successfully reared small quantities of the dottybacks *Pseudochromis flavivertex* and *P. fridmani*. At the research stage, the company is spawning the goby *Amblygobius phalaena*, although larval rearing attempts have not been successful at this time. Other species that are targeted for research are the Comet and some angelfish species.

Information obtained from an advertisement in an aquarium magazine indicated that a tropical marine fish hatchery in London, probably Tropical Marine Centre Ltd., has bred the following species in commercial quantities using the artificial sea salt Tropic Marin: *Amblygobius phalaena*, *Amphiprion allardi*, *A. clarkii*, *A. frenatus*, *A. melanopus*, *A. ocellaris*, *A. percula*, *Gobiodon okinawae*, *Gobiosoma evelynae*, *G. oceanops*, *G. puncticulatus*, *Hippocampus comes*, *H. fuscus*, *H. hystrix*, and *Lysmata debelius* (Fire Shrimp).

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Red Sea Fish pHarm Ltd. is a manufacturer of artificial sea salt (Coral Reef Sea Salt) and other aquarium products in Israel. This company began an intensive Research and Development program to develop new methods for artificial propagation of marine ornamental fishes in 1994, and has successfully cultured the following species: *Amphiprion bicinctus*, *A. clarkii*, *A. frenatus*, *A. ocellaris*, *Chrysiptera parasema*, *Ophistognathus aurifrons*, *Callopleysiops altivelis*, *Pseudochromis fridmani*, *P. flavivertex*, and *Pterosynchiropus splendidus* (Brons, 1995). Red Sea Fish pHarm was the first company to breed two species of fish (i.e., *Pseudochromis fridmani* and *P. flavivertex*) that are indigenous only to the Red Sea. The aquariums and larval tanks at the experimental hatchery are arranged in a closed system equipped with biological filters and protein skimmers. Although the facility is located near the Red Sea, artificial seawater is used in the culture systems. The hatchery has separate facilities for the culture of microalgae and zooplankton. The main goal of the research program is to develop the knowledge and expertise to establish a commercial hatchery producing a wide variety of coral reef fishes. However, the company is also using this research project to promote their artificial sea salt. Red Sea Fish pHarm is presently looking at the economic potential of a commercial hatchery facility (M. Moe, per. comm., 1996). The company believes that all the species they have raised have commercial potential, except for *Chrysiptera parasema* and *Pterosynchiropus splendidus*. The former species does not have a good market value and the latter species has a very low fecundity which limits production (Brons, 1995). The hatchery operation is still at the research and development stage, although the company has reportedly marketed some fish into Europe (S. Brown, per. comm., 1996).

SeaPhiz (formerly EcoActivity, Inc.) is a relatively new company specializing in the manufacture and operation of specialized home aquarium systems (model ecosystems) the company calls Ecotariums and wildlife mariculture systems called ecoCulture facilities. The culture systems are recirculating hatchery systems using algae scrubber technology developed at the Smithsonian Institute. SeaPhiz currently has one ecoCulture system operating in Cleveland, Tennessee and another in Terahaute, Indiana. Species currently cultured in these hatcheries include *Amphiprion clarkii*, a black strain of *A. percula*, *Gobiosoma oceanops*, *Gramma loreto*, *Gramma melacara*, *Hippocampus* sp., *Pseudochromis dutoiti*, *P. flavivertex*, and *P. fridmani*. At the present time, these facilities are just beginning to get up to commercial scale production, with one or two active breeding pairs of each species and a production of about 200 to 250 marketable fish per month (J. Walch, per. comm., 1996). The Arizona-based company is also experimenting with the culture of *Valenciennesa strigata*. In addition to the culture of marine aquarium fish, the company is also culturing live coral, live rock, and live sand.

Aqualife Research Corp., located in Walker's Cay, Bahamas, was the oldest company producing marine ornamentals. Until the recent expansion of the C-Quest, Inc. hatchery, it was also the largest marine ornamental fish hatchery in the world. Aqualife Research Corp. closed its hatchery operation in January 1996, after operating on Walker's Cay for the last 12 years. The fish inventory and equipment were sent to Harbor Branch Oceanographic Institute in Fort Pierce, Florida, where a new investor (Dick Williams from

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Palm Beach, FL) is beginning to investigate commercial marine aquarium fish culture (G. Waugh, per. comm., 1996).

Aqualife's 25,000 ft² hatchery and grow-out facility included a broodstock room containing over 250 broodstock tanks for clownfish and neon gobies (the goby broodstock were kept in a separate room that was air conditioned) and a larval room containing over 40 large tanks (the number of larval rearing tanks was later doubled as production was expanded) and several smaller glass aquariums for larval culture. The live foods area contained an algae laboratory for maintaining small algae cultures, several tanks for hatching brine shrimp, four large transparent cylinders for outdoor algae culture, six 5-ton tanks for growing algae and rotifers, and several large outdoor tanks for brine shrimp. The grow-out area contained over two hundred 300 gallon tanks. Aqualife used saltwater wells which reportedly pumped more than a million gallons of sea water through the hatchery daily. Over 20,000 gallons of microalgae were produced each month. The facility was designed to be capable of producing more than a million fish each year, but the actual production never came close to that amount.

During its peak production in 1989, Aqualife produced and sold over 20,000 fish per month of the following species: *Amphiprion clarkii*, *A. frenatus*, *A. melanopus*, *A. ocellaris*, *A. percula*, *A. perideraion*, *A. polymnus*, *A. sandaracinos*, *Gobiosoma oceanops*, and *Premnas biaculeatus* (both Gold-stripe and regular Maroon clownfish). Aqualife's customers included 42 marine aquarium fish wholesalers in the United States, one in Canada, one in England, and one in West Germany. In 1994, Aqualife was producing about 10,000 fish per month.

In the early days of Aqualife (1972-1983), several other species were successfully cultured that never reached commercial production or were commercially produced for only a short time: *Equetus acuminatus*, *E. lanceolatus*, *E. punctatus*, *Gobiosoma evelynae*, *G. multifasciatum*, *Gramma loreto*, *G. melacara*, *Hypoplectrus gemma*, *H. unicolor*, *Microspathodon chrysurus*, *Opistognathus aurifrons*, *Pomacanthus arcuatus*, and *P. paru*. Aqualife also produced two hybrids: *Equetus acuminatus* x *E. lanceolatus*, and *Pomacanthus arcuatus* x *P. paru*.

Dynasty Marine Associates operated a small hatchery near Marathon, Florida from 1983 to 1995. While it operated, the hatchery produced several species of clownfish, the Neon goby, and seahorses, as a supplement to the main part of the company's business, which is the collection of marine fish, invertebrates, and live rock for the aquarium trade. Although the company no longer raises fish in its own facility, it buys some tank-raised fish from C-Quest, Inc. to include in the inventory of wild-caught fish that Dynasty sells to wholesalers. Species commercially cultured at Dynasty included *Amphiprion clarkii*, *A. frenatus*, *A. melanopus*, *A. ocellaris*, *A. perideraion*, *Gobiosoma xanthiprora*, *G. oceanops*, *Hippocampus* sp., and *Premnas biaculeatus*. Other species reared at an experimental level included *Amphiprion bicinctus*, *Amphiprion chrysopterus*, *Apogon compressus*, *Bodianus rufus*, *Chrysiptera cyanea*, *Equetus acuminatus*, *Equetus lanceolatus*, *E. punctatus*, *Gramma loreto*, *Microspathodon chrysurus*, and

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Opistognathus aurifrons. Dynasty had also produced hybrids on an experimental basis, including *Equetus acuminatus* x *E. punctatus* and *E. lanceolatus* x *E. punctatus*.

At one time, the broodstock included about 60 pairs of clownfish and 48 pairs of Neon gobies. Seahorses were raised from wild-caught fish carrying eggs. About two thirds of the broodstock pairs spawned on a regular basis. Each broodstock tank had its own individual filtration system, but the larval and juvenile grow-out systems were connected in a semi-recirculating system, which included a 30,000 gallon reservoir tank where seawater pumped directly from the ocean was sterilized with chlorine and dechlorinated before use in the system. A header tank, rapid sand filter, biofilters, and protein skimmer completed the filtration system. The larval tanks were located inside the hatchery building and the grow-out tanks and live foods production tanks were located in a greenhouse outside the hatchery. The grow-out period was approximately four months.

Aquaculture Development A/S (ADAS) in Denmark, was also producing clownfish commercially, but is no longer in operation. The company commercially produced ten species of clownfish, using synthetic seawater (Instant Ocean) and recirculating systems. The main species produced were *Amphiprion frenatus*, *A. ocellaris*, *A. sebae*, and *Premnas biaculeatus*. The company began supplying the aquarium markets in Scandinavia, Great Britain, Germany, and the Netherlands with tank-raised anemonefish in 1990, and in November, 1991, the first Danish-produced clownfish were introduced to the American market (Torben, 1992).

The hatchery facility at ADAS consisted of a broodstock system, a larval rearing system, a juvenile grow-out system and a live food production area. The broodstock system contained about 16,000 liters of water that was circulated through sand filters and two UV sterilizers before returning to the 98 spawning tanks. Each 125 liter spawning tank contained an undergravel filter for biological filtration. The larval rearing system contained about 1000 liters of water in four tanks. The grow-out system had a capacity of 52,000 liters and was separate from but similar in design to the broodstock filter system. The live food production system was probably the most sophisticated part of the facility, with two species of microalgae, *Rhodomonas* and *Isochrysis*, grown in two 600 liter photoreactors. Algae production was fully automated, with a maximum daily production of 400 liters of algae. Rotifers were produced in seven tanks, each with a volume of 800 liters. The company developed its own dry food for grow-out of the juvenile fish (Torben, 1992).

Instant Ocean Hatcheries, founded and managed by Frank Hoff, was in operation from 1974 to 1984, producing clownfishes and neon gobies commercially at an inland hatchery in central Florida. Annual production of juveniles reached over 250,000 using Instant Ocean artificial sea salts in a closed culture system. Species produced included: *Amphiprion akallopisos*, *A. clarkii*, *A. ephippium*, *A. frenatus*, *A. ocellaris*, *A. melanopus*, *A. percula*, *A. polymnus*, *Gobiosoma oceanops*, and *Premnas biaculeatus*. In addition, Instant Ocean produced three hybrids: *A. frenatus* x *A. ephippium*, *A. frenatus* x *A. melanopus*, and *A. ephippium* x *A. melanopus*. The hatchery was an 11,200 ft² split level building that contained a 10,000 gallon larval rearing system, a 12,000 gallon

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broodstock system and a 24,000 gallon grow-out system. The larval rearing system contained 80 forty gallon aquariums. The broodstock system contained 180 plywood tanks of 29 or 40 gallons. The grow-out system contained 384 forty gallon plywood tanks. The aquariums in each system were connected to reservoirs and filters. The facility also contained a 10,000 gallon experimental system used for growing algae, invertebrates and extra pairs of subadult fish. There were ten additional 4,000 gallon reservoirs and six 10,000 gallon outdoor recycling ponds. Approximately 70% of the water used in the hatchery was recycled (Hoff, In Prep).

Some research has been conducted by a few universities, public aquariums, and private research institutions on the culture of marine aquarium species. The University of Hawaii, in a research program under the direction of Dr. Chris Brown, was successful in rearing two species of damselfish, *Dascyllus albisela* and *Dascyllus aruanus*, to the juvenile stage, with one of the rearing trials resulting in over 40% survival. Larval feeding during the first two weeks post-hatch was accomplished by maintaining a continuous indoor culture of algae (*Tetraselmis* sp.) and rotifers in the larval tanks, supplemented by progressively larger wild zooplankton (Danilowicz and Brown, 1992). Dr. Brown is presently working on developing methods for captive spawning of *Centropyge fisheri*, *C. loriculus*, and *C. potteri* (C. Brown, per. comm., 1996).

Harbor Branch Oceanographic Institution (HBOI), Fort Pierce, FL began an ornamental marine fish and invertebrate culture program in 1993. To carry out this research, the Institute equipped a 450 ft² glass greenhouse with three independent seawater systems, each containing four 40-gallon aquaria, a wet/dry biofilter and foam fractionator, a GAC contactor, and UV sterilization. The facility also houses translucent fiberglass columns for phytoplankton and marine zooplankton production, and four 3000-liter recirculating larviculture systems. HBOI has successfully reared larvae of the Banded coral shrimp, *Stenopus hispidus*, through metamorphosis after 102 days and the Peppermint shrimp, *Lysmata wurdemanni*, after 35 days. In addition, the Sergeant major, *Abudefduf saxatilis* was successfully cultured. The Institute is also conducting research on the following species:

Species	Common Name
<i>Lysmata grabhami</i>	Scarlet lady
<i>Periclimenes pedersoni</i>	Pederson's cleaner shrimp or Caribbean anemone shrimp
<i>P. yucatanicus</i>	Clown anemone shrimp
<i>Stenopus scutellatus</i>	Golden banded coral shrimp

The University of Texas recently began a research program to investigate the culture of marine ornamental fish and shrimp. Under the direction of Dr. Joan Holt, the following species were spawned in the laboratory: *Bodianus pulchellus*, *Centropyge argi*, *Equetus umbrosus*, *Halichoeres caudalis*, *H. maculipinna*, *Lysmata wurdemanni*, *Serranus tigrinus*, and *Thalassoma bifasciatum*. A large tank (i.e., 1000 gallon raceway) was required to get adult *B. pulchellus* to spawn. However, larval rearing has not been successful for any of the fish species, except *B. pulchellus* and *E. umbrosus*, due to a lack

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of a suitable first food for the tiny larvae. Rotifers are too large as a first food for these species. Wild plankton was also used without success for some of the fish species. However, the laboratory has been more successful in culturing the Peppermint shrimp *Lysmata wurdemanni* through the larval and juvenile stages. The Peppermint shrimp can be reared through the larval stage in 30 to 50 days, depending on water temperature and other factors. High water quality and the use of enriched *Artemia* are important for successfully culturing this species (J. Holt, per. comm., 1996).

The University of Wales began a research program in 1993 to rear several species of cleaner shrimp. Dr. David Fletcher, principal investigator, has succeeded in rearing *Lysmata amboinensis*, *L. debelius* and *Stenopus hispidus*, with the cultured juveniles now producing larvae in the laboratory (D. Fletcher, per. comm., 1994). He is now developing techniques for the mass culture of the two *Lysmata* species, and refinements in rearing techniques has reduced the larval period for these species from 13 or 14 weeks to seven weeks (D. Fletcher, per. comm., 1996). Of the three species listed above, *S. hispidus* is the most difficult to culture through to the juvenile stage. Rotifers are not required as the first feed for this species, and Dr. Fletcher has had success with a variety of live and inert diets. In particular, a nematode species enriched with marine lipids and pigments was used to supply the essential nutrient profile for these crustaceans (D. Fletcher, per. comm., 1994).

Dr. Fletcher is also investigating nutritional requirements of clownfish, including vitamin C requirements in *Amphiprion clarkii* (Fletcher and Wilson, 1996). He is consulting with a private company to develop new diets for clownfish and other marine aquarium fish. He is also consulting with another private company to develop commercial rearing methods for other species of marine aquarium fish, including tangs. The company has been successful in rearing the Catalina goby, *Lythrypnus dalli*, and may be the first to culture this species through the larval stage. They have also been very successful in rearing a variety of seahorse species, producing a second generation in just 7 to 11 weeks. Improved nutrition has been an important factor in the success with seahorses. This group is also developing new live feeds for species with very small larvae (i.e., larvae that cannot consume rotifers at first-feeding) and new technology for induced breeding (D. Fletcher, per. comm., 1996). Emphasis is also being placed on improving broodstock and larval nutrition to achieve greater success with marine aquarium species that have been difficult or impossible to culture in the past.

Various public aquariums have in the past or are presently conducting research on the culture of marine aquarium species. The Waikiki Aquarium, affiliated with the University of Hawaii, conducted small-scale research on culturing marine aquarium shrimp. Under the direction of Syd Kraul, the Aquarium successfully developed rearing methods for the Harlequin shrimp, *Hymenocera picta*. Using enriched *Artemia*, the Aquarium was able to rear thousands of juveniles. The Aquarium is not presently involved in research on the culture of marine ornamental fish or shrimp, but Mr. Kraul is planning to resume commercial culture of the Harlequin shrimp later this year (S. Kraul, per. comm., 1996). The Berlin Aquarium has been culturing several species of clownfish and selling them to raise funds for various projects at the Aquarium (C. Brown, per. comm., 1996). The

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Aquarium is also conducting research with other species of marine aquarium fish. The Baltimore Aquarium is successfully culturing the Highhat, *Equetus acuminatus* and the Neon goby, *Gobiosoma oceanops*. The Tulsa Zoo has cultured the Atlantic spadefish, *Chaetodipterus faber*, and spawned several other species.

Several species of marine aquarium fish and shrimp are now being cultured on a small scale by marine aquarium hobbyists, and many of the articles on rearing marine ornamental species in the aquarium hobby literature were written by individual hobbyists (see Appendix 2). Martin Moe, the first person to commercially culture marine aquarium fish and the founder of Aqualife Research Corp., is currently culturing the Orchid dottyback, *Pseudochromis fridmani*, in an experimental culture system at his home. He reported that the adults began spawning before they were a year old, and produce a spawn of about 1000 eggs every 5 or 6 days. The larvae can eat rotifers when they first begin feeding, and can begin eating *Artemia* nauplii within a few days (M. Moe, per. comm., 1996).

Objective 2. Literature review

A survey of the aquarium hobby literature was conducted for information on spawning and rearing marine aquarium species of fish and shrimp. Information on culture efforts by marine aquarium hobbyists has recently been collected and organized into a quarterly newsletter called "The Breeder's Registry." In addition to articles in the newsletter, the editors have also formed a computer database of spawning and rearing reports for marine aquarium fish and invertebrates. Other aquarium literature containing reports of culturing marine aquarium species include the newsletter "SeaScope" and various aquarium magazines. A review of the scientific literature was also conducted for information on culturing marine aquarium species and other warmwater marine fish and shrimp. The results of these reviews are listed in Appendixes 2, 3 and 4.

Objective 3. Micronesian Species List:

Species	Common Name
<i>Acanthurus leucosternon</i> +	Powder-blue tang
<i>Amblygobius phalaena</i> *	Banded goby, Brown-barred goby, Dragon goby
<i>Amblygobius rainfordi</i> *	Rainford's goby
<i>Amphiprion bicinctus</i> *	Two-banded clownfish or Red Sea clownfish
<i>Amphiprion chrysopterus</i> *	Orange-fin or Blue-stripe clownfish
<i>Amphiprion clarkii</i> *	Clark's clownfish
<i>Amphiprion ephippium</i> *	Red saddleback clownfish
<i>Amphiprion melanopus</i> *	Cinnamon clownfish
<i>Amphiprion perideraion</i> *	Pink skunk clownfish
<i>Balistoides conspicillum</i> +	Clown triggerfish
<i>Callopleziops altivelis</i> *	Comet or Marine betta
<i>Cryptocentrus cinctus</i> *	Yellow prawn goby
<i>Gobiodon citrinus</i> *	Citron goby
<i>Gobiodon okinawae</i> *	Okinawa goby

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<i>Hoplolatilus starcki</i>	Purple-headed sand tilefish
<i>Meiacanthus atrodorsalis</i>	Lyretail blenny
<i>Meiacanthus grammistes</i> *	Striped blenny
<i>Nemateleotris decora</i> **	Decorated dartfish
<i>Nemateleotris magnifica</i>	Fire dartfish or Fire goby
<i>Platax orbicularis</i> +	Circular spadefish or Round batfish
<i>Platax pinnatus</i>	Pinnate spadefish or Redfined batfish
<i>Platax teira</i>	Longfin spadefish or Teira batfish
<i>Pomacanthus imperator</i> +	Emperor angelfish
<i>Pseudochromis porphyreus</i> *	Magenta dottyback
<i>Ptereleotris zebra</i> **	Zebra goby
<i>Siganus (Lo) vulpinus</i>	Foxface rabbitfish
<i>Signigobius biocellatus</i> +	Signal goby or Two-spot goby
<i>Valenciennesa puellaris</i>	Maiden goby
<i>Valenciennesa strigata</i> **	Blue-Streak Goby
<i>Zanclus cornutus</i> +	Moorish idol

* Successfully cultured in captivity.

** Spawning in captivity.

+ Reports of this species being successfully cultured need to be verified.

Objective 4. Culture Methods

Two mated pairs of the Cinnamon clownfish, *Amphiprion melanopus* were obtained by cannulating several individuals and examining the samples to determine the sex of the fish (Figures 5 and 6). Mature eggs or sperm were found in these fish. The sex of some of the fish sampled could not be determined by cannulation. Three pairs of *A. melanopus* spawned during this study, for a total of 21 spawns (Table 1). However, the majority of the spawns were produced by one pair. A second pair was lost early in the study when one of the fish jumped out of the tank. A third unidentified pair in a tank containing several clownfish spawned one time, but was later accidentally disturbed during routine maintenance and did not spawn again. No pair formation was observed and no spawnings occurred in the 10-ton tank containing the large group of *A. melanopus* during the 3 to 4 months that the fish occupied the tank. Also, no spawning occurred in the other clownfish species over a several month period.

Spawn size increased during the study period from less than 100 to nearly 500 eggs per spawn. Some spawns were not used for larval rearing trials. Clownfish eggs hatched in 8 to 9 days, depending on water temperature (ambient water temperatures during the project ranged from 27^o to 29^o C). The eggs elongated and became darker as the embryos developed. The eyes developed and the yolk sacks became smaller shortly before hatching (Figure 7). Newly-hatched larvae were 4 to 5 mm in length. Larval survival in rearing trials with *Amphiprion melanopus* ranged from less than 8% to over 90% (Table 2). Larval mortality occurred primarily during the first few days after hatching.

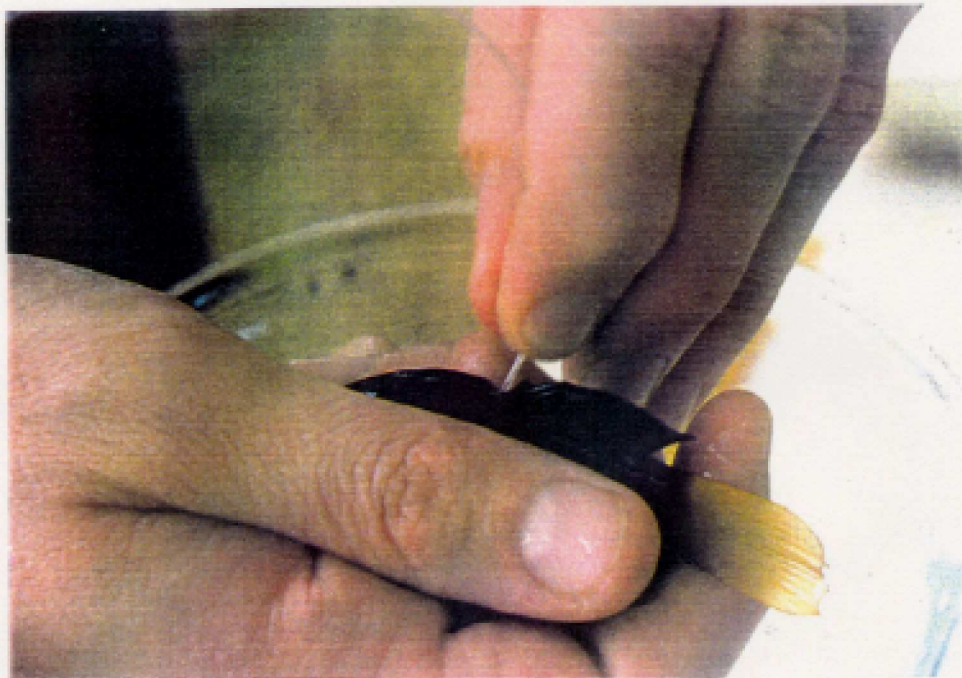


Figure 5. Cannulating Cinnamon clownfish to determine sex and maturation.

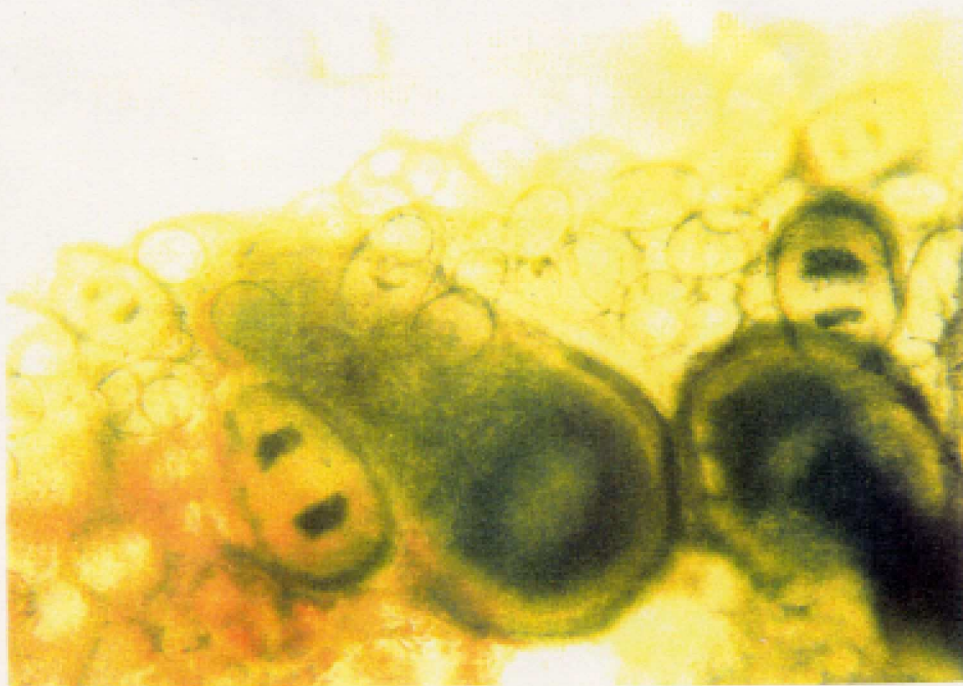


Figure 6. Clownfish oocytes in various stages of development.

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Amphiprion melanopus
Cinnamon Clownfish

Pair T-7	Pair T-8	Pair T-17
Hatch Date	Hatch Date	Hatch Date
12/23/94	12/16/94	04/20/95
01/10/95	01/22/95	
01/31/95	02/01/95	
02/25/95	02/13/95	
05/03/95		
05/12/95*		
06/01/95		
07/26/95		
10/14/95		
10/25/96		
11/06/95		
11/18/95		
11/30/95		
12/31/95		
02/04/96		
02/27/96		

* Note: Egg mass died before hatch

Table 1. Spawning frequency of the Cinnamon clownfish, *Amphiprion melanopus*.



Figure 7. Clownfish egg mass showing eye development in embryos.

Table 2. Aquarium Species Larval Rearing Trials: Cinnamon Clownfish (*Amphiprion melanopus*)

Date Stocked	Date Terminated	No. Stocked	% Survival	Results/Comments
01/22/95	03/06/95	120	25.0	Larvae siphoned from broodstock tank in PM. Found 23 dead next AM.
02/01/95	03/06/95	249	21.3	Larvae siphoned from broodstock tank in PM. About 25% appeared be in shock after transfer. Found >15 dead next AM.
02/26/95	04/17/95	255	33.7	Air off for about 5 hours on day 2. Moved outside at 49 days.
05/04/95	07/30/95	40	67.5	Very small spawn. Larvae siphoned from broodstock tank next AM.
07/27/95	08/20/95	320	94.4	Egg mass stocked in larval tank. Few hatched the first night and remainder hatched the second night. Excellent run with high survival and growth.
10/14/95	10/15/95	150	30.0	Standard use of regular and enriched rotifers and enriched artemia.
10/25/95		0		Less than 50% hatch. High mortality the next day of larvae that did hatch.
11/06/95		0		Larvae hatched in broodstock tank. Larvae not stocked.
11/18/95	12/20/95	170?	< 8.0	Larvae hatched in broodstock tank. Larvae not stocked.
11/30/95	12/28/95	450?	>95.0	First night some larvae hatched in broodstock tank. Moved about 50 larvae to the larval tank. Most of the eggs hatched the following night in the larval tank, but high initial mortality. Only 13 survivors at end of larval run.
12/31/95	01/16/96	60	33.0	Large spawn hatched in larval tank over 2 days. About 50 eggs never hatched. Very good survival of hatched larvae from beginning. Fed rotifers for first two weeks and Artemia beginning on day 8. Harvested 442 juveniles after one month.
				Poor hatch and early and late mortalities in the larval cycle. Normal feeds and water management. Average to poor result.

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Juvenile mortality was less than 5% during the study period. No serious outbreaks of disease were observed during this time. Clownfish reared in the study reached a marketable size at about 3 to 4 cm total length (Figure 8). Growth of juveniles in the first half of the study was very slow, with only about 20% reaching a marketable size after six months. Faster growth was obtained for a later batch of juveniles, with over 50% reaching a marketable size after four months. About 100 clownfish were marketed to a local pet store and another 55 fish were marketed on a trial basis through a local fish collector/exporter to wholesalers in Hawaii and Los Angeles. The incidence of deformities was very low (< 5%) in most of the batches of clownfish raised at the GADTC. However, a higher incidence of deformities (> 35%) occurred in juveniles from a couple of the latter spawns.

The pair of Blue-streak goby broodstock began spawning about one month after being purchased from a local pet store. Initial spawns were noted when larvae were discovered in the broodstock tank at night. In the beginning of the study, the pipe used as a spawning site was not examined for the presence of eggs for fear of disturbing the spawning behavior of the adults. However, it was later determined that the spawning pipe could be examined on a regular basis without disrupting spawning and parental behavior. During spawning, an egg mass was attached to the inside of one of the pipes (Figure 9). Artificial incubation of later stage eggs was attempted, by putting the pipe containing the eggs into a large beaker, with an airstone placed to provide bubbles flowing over the eggs. Another method of artificial incubation was to place the egg mass in a screened open container submerged near the surface of the larval tank with an airstone placed next to the eggs. The eggs would stay at the bottom of the screened container and the newly hatched larvae would be carried over the edge of the container and into the larval tank. Hatch rates of approximately 50% to 90% were obtained when artificial incubation was attempted.

Development of an egg mass was observed under the microscope. Newly laid eggs were yellow and spherical in shape, forming a large mass or cluster that extended out from the substrate. As the eggs developed, light yellow larvae could be seen in a clear oval shaped egg sack. The egg sacks were attached to a strand at the tail end of the larvae and the egg mass was similar in form to a grape cluster. The larvae were initially straight, but as the larvae grew, the tail began to curve back toward the head to form a "U" shape (Figure 10). Larvae were well-developed at hatching, with pigmented eyes and what appeared to be a fully formed gut. Only a small amount of yolk remained at hatching. The newly hatched larvae were less than 2 mm in length.

Fecundity data was not collected during the study, although some egg masses were preserved for later examination. However, it appeared that egg masses contained about 2,000 to 5,000 eggs. The incubation period was 4 to 5 days, with eggs hatching early in the evening. The interval between spawns (i.e., hatching dates) was variable, ranging from 9 to 24 days (Table 3). One spawn may have gone undetected early in the study. Spawning occurred eleven times from August 1994 to April 1995 for an average of about one spawn every 17 days. Two gaps in spawning may have been due to missed observations and were not calculated into the average spawning interval. Shortly after the



Figure 8. Juvenile Cinnamon clownfish raised at the GADTC.



Figure 9. Blue-streak goby egg mass beginning to hatch.

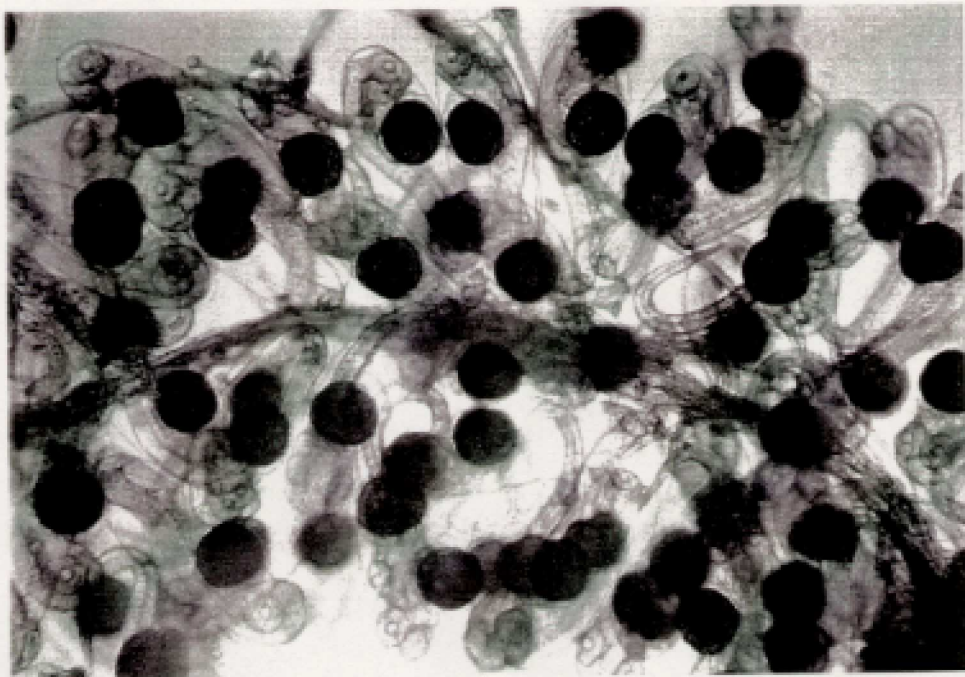


Figure 10. Goby egg mass showing eye development in embryos.

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Ptereleotris zebra
Zebra Goby

Valenciennea strigatus
Blue-streak Goby

Spawn Date	Spawn Interval *	Hatch Date	Spawn Interval
11/06/94	0	08/17/94	0
11/19/94	13	08/27/94	9
12/02/94	13	09/07/94	11
12/17/94	15	09/24/94	17
01/15/95	?	10/24/94	?
01/26/95	11	11/09/94	16
02/04/95	9	11/28/94	18
02/14/95	10	12/18/94	19
02/25/95	11	01/11/95	24
03/10/95	13	02/04/95	24
03/21/95	11	04/04/95	?
03/31/95	10		
04/15/95	15	Average	17.25
04/25/95	10		
05/05/95	10		
05/16/95	11		
05/27/95	11		
06/06/95	10		
06/15/95	9		
06/24/95	9		
07/04/95	10		
07/12/95	8		
07/25/95	13		
08/06/95	12		
08/16/95	10		
09/15/95	?		
09/29/95	14		
10/13/95	14		
10/27/95	14		
11/09/95	13		
11/19/95	10		
12/02/95	13		
12/19/95	17		
01/17/96	?		
01/27/96	10		
02/08/96	12		
02/19/96	11		
03/01/96	11		
03/13/96	12	Average	11.57

* Note: Spawn intervals are days between spawns. Intervals with a "?" indicate an unknown interval due to a possible missed spawning observation.

Table 3. Spawning frequency of gobies at the GADTC.

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last recorded spawning in April of 1995, the pair were moved to another smaller tank with some other fish because the tank they were in was needed for another project, and other suitable tanks were not available at the time. The pair did not spawn after being moved and eventually died. A few rearing trials were conducted with Blue-streak goby larvae, feeding them s-type rotifers. However, none of the larvae survived more than a few days.

Spawning of the zebra goby broodstock pair occurred on a regular basis throughout the study, beginning about two months after the pair was acquired. Spawns were found early in the morning, and hatching occurs immediately after dark on the fourth night. The egg mass was attached to the underside of the clam shell and appeared to be about the same size as the Blue-streak goby egg masses (Figure 11). Embryonic development appeared to be similar to that of the Blue-streak goby. The eggs could be successfully hatched in the larval tank by removing the shell containing the eggs from the broodstock tank shortly before hatching and placing it in the larval tank. However, if the eggs were taken from the parents too soon, hatching was usually unsuccessful, even when the eggs were well aerated. A total of 39 spawns were produced in a 16 month period for an average of about one spawn every 11.5 days (Table 3).

Several rearing trials were conducted with the zebra goby larvae. SS-type rotifers were fed to the larvae and attempts were also made to feed them oyster larvae and an artificial diet. However, the larvae did not survive past the first few days when fed ss-rotifers or artificial diets and oyster larvae could not be obtained in sufficient quantities when required to feed the goby larvae.

The three pairs of Banded coral shrimp began spawning about one to two months after being purchased from local collectors. Five additional pairs were obtained later in the study and also began spawning. Banded coral shrimp spawned (i.e., larvae hatched) at intervals of 13 to 23 days (Table 4). Some larger periods of time between spawns is believed to be due to missing observations of hatching larvae. Eggs usually began hatching about 3 to 4 hours after sunset and hatching was completed within an hour. Spawn size usually ranged from 1000 to 3000 larvae, but spawn size may reach as high as 5000 larvae. Limited data from one pair suggests that fecundity did not decline with time in captivity.

Several larval rearing trials were conducted with the Banded coral shrimp (Table 5). In the beginning of most of the trials, rotifers were maintained in the larval tank at a density of 10 to 20 rotifers/ml and the green algae *Nannochloropsis oculata* was added daily to maintain a concentration of about 500,000 to 1,000,000 cells/ml in the larval tank. High larval survival was usually obtained through the first three weeks. Then the diet was gradually changed from rotifers to *Artemia* nauplii and survival began to decline. In the best rearing trial, some larvae survived until day 51. Contamination of larval tanks with growths of hydroids and small free-swimming jellyfish medusae sometimes occurred, especially after larvae were reared in the same tank for a few weeks. Total mortality of the Banded coral shrimp larvae usually occurred within a few days after their appearance in large numbers.

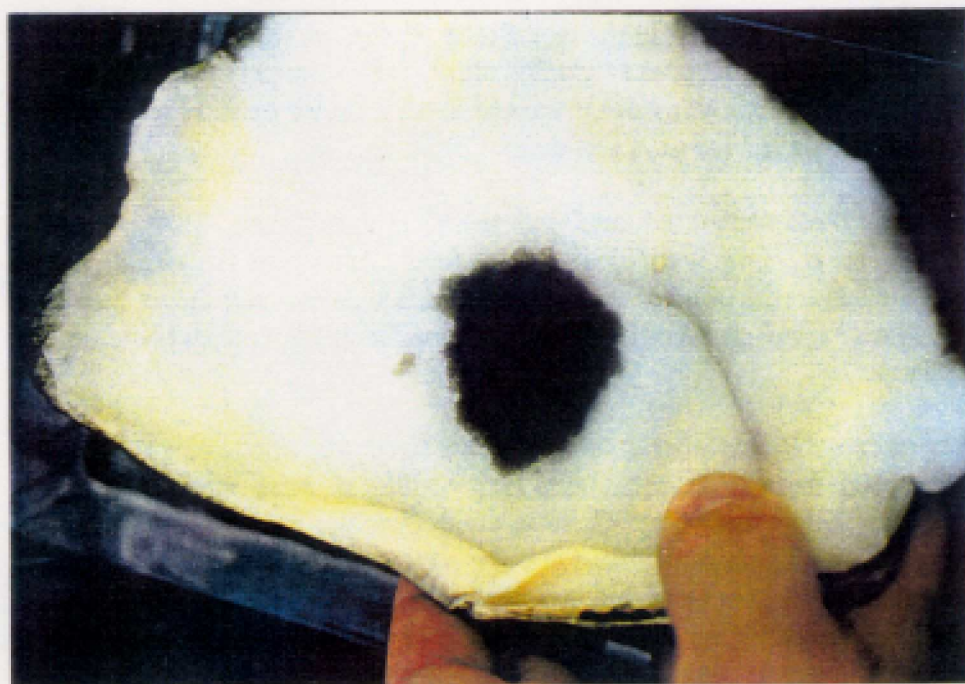


Figure 11. Zebra goby egg mass shortly before hatching.

Table 5. Aquarium Species Larval Rearing Trials: Banded Coral Shrimp (*Stenopus hispidus*)

06/04/95	07/17/95	1,500	0	Survival to day 42. Tank became too dirty and hydroids ate all the artemia.
07/05/95	07/19/95	1,500	0	Overfed and overrun with artemia. Also larvae hatched in broodstock tank. Low survival early on.
07/21/95	09/11/95	1,000	0	Larvae stocked from two broodstock tanks. Began 70% daily water exchange and flushing at day 7. Fed combination of enriched rotifers (first two weeks) and newly hatched and 24 hour enriched artemia. Larvae healthy and growing up to last two days of run, when tank became heavily infested with jellyfish medusae and hydroids. Survival to day 51.
08/23/95	09/04/95	3,279	0	Stocked over six days, and too large a tank for too few larvae. Stocking rate was 1.6 per liter. Too hard to manage.
09/02/95	09/10/95	1,150	0	Larvae were able to escape through screen system when lowering water level each day. Too few larvae left to keep running tank.
10/04/95	11/04/95	3,473	0	Tried to move animals when tank dirty, but too slow to move them and medusae appeared on day 21 and 30. Dead by day 31.
10/20/95	11/23/95	2,867	0	Also tried to move this tank when dirty, but too slow to move them and medusae appeared about day 19. The few that were moved slowly disappeared by day 35.
11/21/95	12/10/95	3,400	0	Eliminated contamination by filtering all water going into tank, rinsing Artemia before feeding, and stocking larvae hatched in filtered water. Fed Artemia from beginning of run. Survival high for first two days, but sharp decline during the next week.
11/23/95	12/10/95	1,715	0	Same result as previous run.

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Stenopus hispidus
Banded Coral Shrimp

BCS #1	Spawn Interval	BCS #2	Spawn Interval	BCS #3	Spawn Interval
Hatch Date	(Days)	Hatch Date	(Days)	Hatch Date	(Days)
08/04/94	0	07/19/94	0	08/25/94	0
08/27/94	23	08/04/94	16	09/09/94	15
10/13/94	?	08/22/94	18	10/10/94	?
10/29/94	16	09/08/94	17	10/24/94	14
11/14/94	16	09/27/94	19	11/09/94	16
11/29/94	15	10/28/94	?	11/26/94	17
12/15/94	16	11/18/94	21	12/16/94	20
01/07/95	23	12/09/94	21	01/07/95	22
01/23/95	16	02/05/95	?	01/22/95	15
02/11/95	19	02/21/95	16	02/09/95	18
02/28/95	17	03/09/95	16	02/27/95	18
04/29/95	?	03/25/95	16	03/12/95	13
05/17/95	18	06/19/95	?	04/14/95	?
06/03/95	17	08/07/95	?	05/02/95	18
07/31/95	?	08/23/95	?	05/18/95	16
09/24/95	?			06/05/95	18
10/10/95	16	Average	17.8	07/04/95	?
10/29/95	19			07/20/95	16
11/23/95	?			08/07/95	18
				08/22/95	15
Average	17.8			09/24/95	?
				10/11/95	17
				11/13/95	?
				Average	16.8

* Note: Spawn intervals with a "?" indicate an unknown interval due to a possible missed spawning observation.

Table 4. Spawning frequency of the Banded coral shrimp, *Stenopus hispidus*.

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A few early attempts to spawn newly collected sea urchins were relatively successful in producing fertilized eggs and larvae. However, the use of sea urchin larvae for larval rearing of gobies was later abandoned because it became very difficult to induce spawning in the sea urchins held at the hatchery. Difficulties were also encountered in spawning oysters.

Objective 5. Regulatory Constraints

In 1987 Germany banned the importation and sale of butterflyfish, angelfish, and Moorish idols. However, determined German hobbyists can still drive across the border to Holland and obtain these species (R. Meyers, per. comm., 1996). Nature conservation laws in Israel forbid collection of reef specimens entirely, and the export of wild caught Red Sea fishes is restricted in most other countries on the Red Sea (Brons, 1996). Hawaii has banned the importation of scorpionfishes, eels, and sharks (R. Pyle, per. comm., 1996). Florida has size limits and bag limits for marine angelfish collected from its waters. The Commonwealth of the Northern Mariana Islands banned aquarium fish collection in 1992. The Republic of Palau banned the collection and export of tropical marine aquarium fish in May 1993 (Baker, 1993). This ban was the result of The Nature Conservancy efforts to protect the coral reef habitats of this island group. The U.S. Territory of Guam is preparing to ban the export of wild-caught marine aquarium species from its reefs and lagoons. Local aquarium stores and private citizens would still be allowed to collect with government-issued licenses after the regulations are adopted, but would not be permitted to export their catches (Baker, 1993; G. Davis, Division of Aquatic and Wildlife Resources, per. comm., 1996). Captive-bred aquarium specimens could still be exported under this proposed legislation, provided the mariculture operation receives the certification. In various countries (including the Territory of Guam), selected areas have been designated as marine parks or reserves, where the collection of animals is prohibited.

Objective 6. Mariculture certification of captive-bred marine aquarium fish and shrimp cultured on Guam would consist of an official certificate of origin from a mariculture facility approved by the Division of Aquatic and Wildlife Resources (G. Davis, per. comm., 1996).

Objective 7. Cash flow projections were developed for a small clownfish hatchery and grow-out operation. Table 6 provides start-up costs for the facility. Tables 7 and 8 provide cash flow projections at 16% and 4% interest rates, respectively. Assumptions used in the analyses are provided in Table 9. The most important factors affecting profitability are consistent production of marketable fish, market price and sales volume. Feed costs are not a major factor affecting economic return, although the unit cost of feed for marine ornamental fish is much higher than commercial diets for foodfish.

Aquarium Project Economics TABLE 6. LISTS OF START-UP COSTS

Hatchery and Growout Combined Operation
(Clownfish)

List Costs of Facility	List Costs of Supplies & Equip. for Start-Up	Hatchery Production Assumptions	
Site Preparation/Well	\$10,000	No. Pairs of Broodstock	15
Tanks	\$10,600	Active Spawning Pairs	10
Plumbing	\$9,000	No. Spawns per Month Stocked per Pair	2.3
Pumps	\$2,100	No. Larvae per Spawn	750
Blowers/Aerators	\$1,400	Survival of Larvae to Transfer to Growout	50%
Generator	\$2,500	No. Days to Transfer/Sale	30
Other Electrical Costs	\$2,000	Transfer/Sale Price	\$0.20
Fence/Lights	\$2,500	Cost of Feed per Month	\$575.00
Pick-up Truck	\$10,000	Revenue per Batch	\$75.00
Misc.	\$4,010	Total Gross Revenue per Month	\$1,725.00
Total	\$54,110		
		Grow-out Production Assumptions	
		No. Juveniles per Month Stocked	8625
		% Survival of Juveniles to Market Size	70%
		% Saleable	70%
		No. Days to Sale	120
		Sale Price	\$2.25
		Cost of Feed per Month	\$542
		Cost of Fry per Month	\$1,725.00
		Total Gross Revenue per Month	\$9,509.06

TABLE 7.

**CASH FLOW PROJECTIONS FOR MARINE AQUARIUM FISH PRODUCTION
COMBINED HATCHERY AND GROW-OUT OPERATION**

ITEM/YEAR	1ST QTR	2ND QTR	3RD QTR	4TH QTR	YEAR 1	YEAR 2	YEAR 3	YEAR 4	YEAR 5
INCOME									
1 BALANCE FORWARD	\$0	\$49,485	\$17,296	\$6,988	\$0	\$4,731	\$17,039	\$29,348	\$41,656
2 CAPITAL	\$25,000				\$25,000				
3 LOAN	\$90,000				\$90,000				
4 SALES		\$9,509	\$19,018		\$28,527	\$114,109	\$114,109	\$114,109	\$114,109
5 OTHER									
6 TOTAL INFLOW	\$115,000	\$49,485	\$26,806	\$26,006	\$143,527	\$118,840	\$131,148	\$143,456	\$155,765
CAPITAL COSTS									
7 SITE PREPARATION/WELL	\$10,000				\$10,000				
8 TANKS/PLUMBING	\$7,600	\$12,000			\$19,600				
9 PUMPS	\$2,100				\$2,100				
10 BLOWERS/AERATORS	\$1,400				\$1,400				
11 FENCE/LIGHTS	\$2,500				\$2,500				
12 PICK-UP TRUCK	\$10,000				\$10,000				
13 STORAGE SHED	\$1,600				\$1,600				
14 GENERATOR AND ELECTRICAL	\$4,500				\$4,500				
15 LAB EQUIPMENT	\$1,700				\$1,700				
16 MISCELLANEOUS	\$3,140	\$1,200			\$4,340				
17 TOTAL CAPITAL COSTS	\$44,840	\$13,200			\$57,740				
OPERATIONAL COSTS									
18 BROODSTOCK	\$600				\$600	\$600	\$600	\$600	\$600
19 FEEDS	\$3,626		\$1,117	\$2,234	\$6,977	\$13,403	\$13,403	\$13,403	\$13,403
20 MEDICATIONS	\$500		\$0	\$0	\$500	\$500	\$500	\$500	\$500
21 EQUIPMENT & SUPPLIES	\$3,250		\$0	\$0	\$3,250	\$3,250	\$3,250	\$3,250	\$3,250
22 ELECTRICITY	\$1,500	\$1,500	\$1,500	\$1,500	\$6,000	\$6,000	\$6,000	\$6,000	\$6,000
23 MANAGEMENT/LABOR	\$9,000	\$9,000	\$9,000	\$9,000	\$36,000	\$42,000	\$42,000	\$42,000	\$42,000
24 AUDIT/LEGAL/INSURANCE	\$600	\$600	\$600	\$600	\$2,400	\$2,400	\$2,400	\$2,400	\$2,400
25 LEASE	\$1,500	\$1,500	\$1,500	\$1,500	\$6,000	\$6,000	\$6,000	\$6,000	\$6,000
26 MAINTENANCE	\$0	\$577	\$577	\$577	\$1,732	\$2,310	\$2,310	\$2,310	\$2,310
27 CONTINGENCY (5%)	\$999	\$689	\$715	\$771	\$3,173	\$3,823	\$3,823	\$3,823	\$3,823
28 LOAN PAYMENTS	\$0	\$4,523	\$4,523	\$4,523	\$13,569	\$18,091	\$18,091	\$18,091	\$18,091
29 TAXES	\$0	\$0	\$285	\$571	\$856	\$3,423	\$3,423	\$3,423	\$3,423
30 TOTAL OPER. COSTS	\$20,975	\$18,989	\$19,817	\$21,275	\$81,056	\$101,801	\$101,801	\$101,801	\$101,801
31 TOTAL OUTFLOWS	\$65,515	\$32,169	\$19,817	\$21,275	\$138,796	\$101,801	\$101,801	\$101,801	\$101,801
32 NET CASH BALANCE	\$49,485	\$17,296	\$6,988	\$4,731	\$4,731	\$17,039	\$29,348	\$41,656	\$53,964

104.48% NOTE: COMPARATIVE CALCULATION FOR 10 YEAR PROJECTIONS WITH 8% LOAN
 60.92% NOTE: CALCULATED ON 10 YEAR PROJECTIONS WITH 16% LOAN
 104.48% NOTE: COMPARATIVE CALCULATION FOR 10 YEAR PROJECTIONS WITH 16% LOAN

TABLE 8.

**CASH FLOW PROJECTIONS FOR MARINE AQUARIUM FISH PRODUCTION
COMBINED HATCHERY AND GROW-OUT OPERATION (4% INTEREST RATE)**

ITEM/YEAR	1ST QTR	2ND QTR	3RD QTR	4TH QTR	YEAR 1	YEAR 2	YEAR 3	YEAR 4	YEAR 5
INCOME									
1 BALANCE FORWARD	\$0	\$49,485	\$19,086	\$10,567	\$0	\$10,099	\$29,564	\$49,029	\$68,494
2 CAPITAL	\$25,000				\$25,000				
3 LOAN	\$90,000				\$90,000				
4 SALES		\$9,509	\$19,016		\$28,527	\$114,109	\$114,109	\$114,109	\$114,109
5 OTHER									
6 TOTAL INFLOW	\$115,000	\$49,485	\$28,595	\$29,565	\$143,527	\$124,208	\$143,673	\$163,138	\$162,603
CAPITAL COSTS									
7 SITE PREPARATION/WELL	\$10,000				\$10,000				
8 TANKS/PLUMBING	\$7,600	\$12,000			\$19,600				
9 PUMPS	\$2,100				\$2,100				
10 BLOWERS/AERATORS	\$1,400				\$1,400				
11 FENCE/LIGHTS	\$2,500				\$2,500				
12 PICK-UP TRUCK	\$10,000				\$10,000				
13 STORAGE SHED	\$1,600				\$1,600				
14 GENERATOR AND ELECTRICAL	\$4,500				\$4,500				
15 LAB EQUIPMENT	\$1,700				\$1,700				
16 MISCELLANEOUS	\$3,140	\$1,200			\$4,340				
17 TOTAL CAPITAL COSTS	\$44,540	\$13,200			\$67,740				
OPERATIONAL COSTS									
18 BROODSTOCK	\$600				\$600	\$600	\$600	\$600	\$600
19 FEEDS	\$3,626		\$1,117	\$2,234	\$6,977	\$13,403	\$13,403	\$13,403	\$13,403
20 MEDICATIONS	\$500		\$0	\$0	\$500	\$500	\$500	\$500	\$500
21 EQUIPMENT & SUPPLIES	\$3,250		\$0	\$0	\$3,250	\$3,250	\$3,250	\$3,250	\$3,250
22 ELECTRICITY	\$1,500	\$1,500	\$1,500	\$1,500	\$6,000	\$6,000	\$6,000	\$6,000	\$6,000
23 MANAGEMENT/LABOR	\$9,000	\$9,000	\$9,000	\$9,000	\$36,000	\$42,000	\$42,000	\$42,000	\$42,000
24 AUDIT/LEGAL/INSURANCE	\$600	\$600	\$600	\$600	\$2,400	\$2,400	\$2,400	\$2,400	\$2,400
25 LEASE	\$1,500	\$1,500	\$1,500	\$1,500	\$6,000	\$6,000	\$6,000	\$6,000	\$6,000
26 MAINTENANCE	\$0	\$577	\$577	\$577	\$1,732	\$2,310	\$2,310	\$2,310	\$2,310
27 CONTINGENCY (5%)	\$999	\$689	\$715	\$771	\$3,175	\$3,823	\$3,823	\$3,823	\$3,823
28 LOAN PAYMENTS	\$0	\$2,734	\$2,734	\$2,734	\$8,201	\$10,934	\$10,934	\$10,934	\$10,934
29 TAXES	\$0	\$0	\$285	\$571	\$856	\$3,423	\$3,423	\$3,423	\$3,423
30 TOTAL OPER. COSTS	\$20,975	\$17,200	\$18,028	\$19,486	\$75,686	\$94,644	\$94,644	\$94,644	\$94,644
31 TOTAL OUTFLOWS	\$65,516	\$30,400	\$18,028	\$19,486	\$133,428	\$94,644	\$94,644	\$94,644	\$94,644
32 NET CASH BALANCE	\$49,485	\$19,086	\$10,567	\$10,099	\$10,099	\$29,564	\$49,029	\$68,494	\$67,959

LOAN CALCULATION
IRR CALCULATION
130.68% NOTE: CALCULATED ON 10 YEAR PROJECTIONS WITH 4% LOAN
\$911.21 NOTES: PRINCIPAL=90,000, RATE=4%, TERM=10YRS

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Table 9. Cash Flow Assumptions

INCOME

- 1) Balance Forward: Cash balance brought forward from previous year.
- 2) Capital: Investment by owner.
- 3) Loan: Amount borrowed from bank or other financial institution. Assume loan of \$90,000 for 10 years at 16%.
- 4) Sales: Total income from sale of aquarium species.
- 5) Other: Other sources of income such as interest earnings.
- 6) Total Inflow: The sum of items 1 through 5 above.

CAPITAL COSTS

- 7) Site Preparation/Well: Well cost of \$3,000 plus \$40/ft. for a 100 ft well. \$3,000 for site clearing and leveling.
- 8) Tanks/Plumbing: Broodstock tanks/aquariums; 15pcs. @ \$100 ea. Hatchery tanks; 8pcs. @ \$200 ea. Growout tanks; 25pcs. @ \$300 ea. Plus general PVC plumbing for air and water at \$9,000 lump sum estimate.
- 9) Pumps: 2 submersible pumps for well at \$800 each and 2-3 small submersible pumps for total cost of \$500.
- 10) Blowers/Aerators: 2 blowers at \$700 each.
- 11) Fence/Lights: Perimeter fence and outdoor security lighting lump sum estimate of \$2,500.
- 12) Pick-up Truck: Purchase used truck for \$10,000.
- 13) Storage Sheds: 2 sheds at \$800 each. One shed for storage and one for office/lab.
- 14) Generator and Electrical: one generator at \$2500 and \$2000 electrical work.
- 15) Lab Equipment: One microscope at \$1500 and \$200 of laboratory glassware and supplies.
- 16) Miscellaneous: 10% of cost of capital costs (except for pick-up truck).
- 17) Total Capital Costs: Sum of items 7 through 16 above.

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Table 9. Cash Flow Assumptions (continued)

OPERATIONAL COSTS

- 18) Broodstock: 20 pairs clownfish with anemones at \$30 per clownfish pair plus anemone.
- 19) Feeds: 1st Qtr: \$1000 for Artemia and \$2,626 for Inert feeds for hatchery based on start-up assumptions. 2nd Qtr: No feed purchase required. 3rd Qtr: Cost of feed for broodstock and grow-out for one month. 4th Qtr: Cost of feed for broodstock and grow-out for two months. Yearly thereafter \$13,403 for 12 months of hatchery and growout feeds based on assumptions in "Growout Production Assumptions" section.
- 20) Medications: Lump-sum estimate of \$500 per year.
- 21) Equipment & Supplies: Cost of algae nutrients, hosing/tubing/netting/screens, glassware, and buckets/nets. \$3,250 per year lump sum estimate.
- 22) Electricity: Estimate of \$500 per month.
- 23) Management/labor: Salaries/benefits for owner/family at \$3000/month for first year and \$3500/month each year thereafter.
- 24) Audit/Legal/Insurance: Lump sum estimate of \$200 per month.
- 25) Lease: \$500/month for 1/2 acre site.
- 26) Maintenance: Estimated at 4% per year of total capital costs (no cost for 1st quarter).
- 27) Contingency: 5% of operating costs each quarter or year.
- 28) Loan Payments: No payment for first quarter, then monthly payments based on a 10 year loan of \$90,000 at 16% and 4% interest for tables 7 and 8, respectively.
- 29) Taxes: Estimate based on 3% of sales costs.
- 30) Total Operating Costs: Sum of items 18 through 29 above.
- 31) Total Outflows: Total Operating Costs plus Total Capital Costs.
- 32) Net Cash Balance: Total Inflows minus Total Outflows.

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DISCUSSION OF RESULTS:

Objective 1. C-Quest struggled in the beginning of its operation with several problems, including permitting delays, disputes with government agencies, personnel problems, difficulty in getting quality broodstock, disease and water quality problems. The company has survived the difficulties of start-up and is now becoming successful from a biological perspective, culturing large numbers of clownfish as well as several other high-value species (e.g., *Pseudochromis* sp. and the Comet). C-Quest is not yet profitable, but it is expected to hit the break-even point and begin making a profit before the end of the year (W. Addison, per. comm., 1996). The company has put emphasis on research and development of new species, and this should continue to pay off in the future and increase profitability of the company. C-Quest could benefit from more effective marketing, and higher prices could probably be obtained for their fish, making the company more profitable.

Reef Propagations is a family owned and operated business, and was started by a marine aquarium hobbyist without biological training or experience in aquaculture. Reef Propagations has the advantage of being very close to a large market, and has been able to get relatively high prices for the clownfish it produces (probably as a result of savings on shipping costs to the customer). The fish produced have good coloration and the percentage of saleable fish is nearly 100 percent. The company has also benefited by buying some other species of clownfish from another hatchery (C-Quest) and reselling them at a profit, thereby increasing product diversity and sales volume. The capital costs for replicating the culture facility (building already provided) is less than \$10,000. Although this operation is successful financially, the company has had some technical problems that have limited production and profitability in the last two years. These problems will probably be resolved in the near future. The ultimate goal of this small facility is 450 fish per week with gross sales of 70,000 per year and anticipated operating expenses of about \$10,000 to \$15,000.

Desert Fisheries has achieved initial success using an unlikely source of saltwater (i.e., marine geothermal water in Utah), for their hatchery operation. The water is not the same as natural seawater in chemical composition, being low in sulphates and magnesium and having a low pH. Ammonia is also present at about 0.2%. The temperature of the incoming water is 90° F. Various water treatments were tried at one time, but the untreated water was found to provide better results than the treated water (L. Nelson, per. comm., 1996). Aquarium Systems and one or more other business partners backed out of the business because they didn't think the hatchery would be successful using the marine geothermal water. Desert Fisheries has good results culturing some species of clownfish using their water, but they have had difficulty breeding other species (*Amphiprion clarkii*, *A. perideraion* and *Premnas biaculeatus*), and this may be related to the different water chemistry of their saltwater. However, the company is producing several hundred marketable fish per week and obtaining a high market price.

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SeaPhiz believes that their technology has the potential for culturing 100 to 125 species of marine ornamental fish. The algae scrubber technology used in these culture systems is reported to be very efficient at removing ammonia and other waste products, with no water changes being required in the last 1.5 years of operation (J. Walch, per. comm., 1996). The company is not selling their hatchery system technology at this time, but has decided to test the facilities they have in operation first to evaluate the potential for commercial culture of marine ornamental fish and invertebrates.

After more than ten years of operation, Dynasty Marine Associates phased out their hatchery operation because the company could not make a profit raising clownfish in the Florida Keys. This was due mainly to the high cost of labor and electricity. Air conditioning was required in the hatchery in the heat of the summer, and heating was required in the colder winter months (F. Young, per. comm., 1996).

Aqualife was never profitable and finally closed down as a result of cost-cutting measures by the parent company, Precision Valve Corporation. One of the main problems affecting profitability at Aqualife was poor water quality. The saltwater wells that supplied that hatchery became contaminated by petroleum from the nearby power plant. Other wells were later drilled farther away from the power plant, but the water from these wells contained a high concentration of iron. Aqualife had plans for setting up an intake pipe to pump water directly from the ocean, but the hatchery operation was closed down before the project was completed (G. Waugh, per. comm., 1996).

When the Instant Ocean hatchery operation shut down, the company was selling clownfish at about \$1.65 per fish, with a production cost estimated to be about \$2.25 per fish. Several things contributed to the high production cost. Personnel costs were high due to a large number of employees and a staff top-heavy with management personnel. Salaries were 53.9% of the monthly operating expenses, and the hatchery operations could have been conducted with half the number of personnel that were on staff (Hoff, In Prep.). The hatchery used artificial sea salts in a recirculating system, but the filtration system used (sub-gravel filters for biological filtration) was not very efficient compared to the filtration technology available today (F. Hoff, pers. comm., 1996). Another problem was that juvenile survival in grow-out was about twice the amount of those fish that were considered marketable (i.e., overall survival in grow-out was 43.6%, but 23% of the survivors were discarded as unsalable). The production of a large number of fish that were unmarketable greatly increased the cost of production.

Objective 2. Literature on marine ornamental culture is becoming more abundant, although little scientific research has been done in the past. Most of the available information on spawning and rearing marine aquarium species comes from aquarium hobby publications, although some research institutions have recently begun research programs on marine ornamentals. Advances in rearing techniques for other marine species can be applied to the culture of marine aquarium fish and shrimp.

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Objective 3. Several species of marine aquarium fish on the Micronesian Species List have been reared commercially or on an experimental level. The Banded goby or Dragon goby, *Amblygobius phalaena*, has been reported to be successfully cultured by a commercial hatchery in England. Desert Fisheries is also attempting to culture this species, but the small larvae will not eat rotifers and the company has yet to get the larvae to survive to the juvenile stage (L. Nelson, per. comm., 1996). A related species, Rainford's goby, has been cultured in small quantities by C-Quest, although the larvae are also difficult to rear in large numbers because the tiny larvae cannot eat rotifers during the first 4 to 5 days. Ciliates and copepod nauplii were successfully used to get a few larvae through to the juvenile stage (W. Addison, per. comm., 1996). These two species have potential for commercial culture in Micronesia, although they will be more difficult to culture than many other species.

All the clownfish species have been cultured on a commercial or semi-commercial basis by several companies or research institutions, although *Amphiprion chrysopterus* is reported to be especially difficult to get to spawn in captivity and this species is probably not being reared commercially at the present time (Young, 1991A). Due to its limited availability to the aquarium hobby and striking appearance, this species has a higher market value than most other clownfish species. Probably the only exception is *A. latezonatus*, which is even less common in the aquarium trade and has a wholesale value of about \$25 per fish.

The Comet has been successfully cultured by at least three companies, and commands a high price in the aquarium market (Wassink and Brons, 1990; Brons, 1995). It is reported to be difficult to induce to spawn, but the larvae are relatively easy to rear (Wilkerson, 1996). The Citron and Okinawa gobies have been commercially cultured by C-Quest (Gutierrez, 1996; Wilkerson, 1996). The Yellow prawn goby has also been reared in captivity and will probably become available commercially in the future. The Purple-headed sand tilefish, *Hoplolatilus starcki*, is uncommon in the aquarium trade and no attempts to culture this species have been reported, but the small size, stunning appearance and high market value of this species make it worth investigating. The Striped blenny has been reared in captivity, and a related species, the Lyretail blenny *Meiacanthus atrodorsalis*, could probably also be reared, making these species good candidates for commercial culture in Micronesia. The dartfish (*Nemateleotris* sp.) are small colorful fish that are popular for marine aquariums. At least one species of dartfish (*N. decora*) has spawned in aquariums, although the eggs are reported to be only about half the size of clownfish eggs (Schiller, 1990). No attempts to rear the larvae have been reported.

The Round batfish *Platax orbicularis* is reported to be under cultivation in Taiwan (Liao et al., 1995). A related species, the Atlantic spadefish, has also been cultured in captivity, and captive spawning and larval rearing are not difficult (Walker, 1991). No published information was found on spawning or larval rearing of the other two species of Indo-Pacific batfish. However, these latter species (*P. pinnatus* and *P. tetra*) are likely to have similar requirements for cultivation and have a much higher market value. No published information was found on spawning or larval rearing of the Magenta dottyback. However, this species and three other *Pseudochromis* species have been commercially reared by C-

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Quest, Inc. (W. Addison, per. comm., 1996). Adult *P. fridmani* begin spawning before they are a year old, and produce a spawn of about 1000 eggs every 5 or 6 days. The larvae can eat rotifers when they first begin feeding, and can begin eating *Artemia* nauplii within a few days (M. Moe, per. comm., 1996). However, larval survival appears to be better when wild plankton is provided, and this may indicate nutritional problems that need to be addressed. No reports of culturing the Foxface (*Siganus vulpinus*) were found, but other rabbitfish of the genus *Siganus* have been successfully cultured on an experimental or commercial basis, and it is likely that the Foxface could also be cultured.

The Zebra goby (*Ptereleotris zebra*) spawns on a consistent basis at the GADTC, but larval rearing to date has not been successful, due to the extremely small size of the larvae and the difficulty in providing a suitable food. The Blue-streak goby (*Valenciennea strigata*) also spawned on a regular basis at the GADTC, but the larvae of this species is similar in size to the Zebra goby larvae, and they also have not been successfully reared. These gobies are popular aquarium species and have a good market value. It is believed that success with both of these species, as well as the Maiden goby (*V. puellaris*) and many other more highly valued marine aquarium fish, could be achieved if oyster larvae or other similar sized organisms could be supplied on a consistent basis. Until this happens, species with very small larvae are not recommended for commercial culture.

Many other species of commercially valuable marine aquarium fish from other parts of the world could be cultured in Micronesia if their importation for mariculture is permitted. These include some of the clownfish species (e.g., *Amphiprion frenatus*, *A. latezonatus*, *A. ocellaris*, *A. percula*, *A. polymnus*, and *Premnas biaculeatus*), most of the dottybacks (i.e., *Pseudochromis dutoiti*, *P. flavivertex*, *P. fridmani*, *P. paccagnellae*, and *P. springeri*) and several other species that have been reared in captivity (Appendix 4).

Several species of marine ornamental shrimp have been successfully reared in captivity, and a few species (e.g., *Lysmata debelius* and *Hymenocera picta*) appear to have good commercial potential. Although the larval period of these species are relatively long (i.e., 35 to 50 days), it is manageable and can probably be decreased somewhat with improved rearing methods. A potential problem with commercializing ornamental shrimp culture may be in producing large enough quantities of juveniles (M. Moe, per. comm., 1996).

Objective 4. Broodstock pairing was a problem area throughout the study. None of the Cinnamon clownfish (*Amphiprion melanopus*) were obtained as mated pairs, but were collected as groups of unpaired fish. Cannulation to determine the sex of the fish was partially successful in the beginning, resulting in two mated pairs. However, the sex of many of the fish could not be determined by cannulation, and these fish may not have had mature eggs or sperm at the time. The fastest and most reliable method for obtaining mated pairs of aquarium fish for broodstock is to collect naturally mated pairs from the wild. With clownfish, it would also be helpful to collect the anemone that the pair was occupying. Anemones are not required for breeding most clownfish, but are desirable if available for stimulating more natural behavior and facilitating pair formation. Some species of clownfish prefer a particular type of anemone. Mated pairs can usually be

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obtained by placing together one large clownfish (i.e., a female) with a small clownfish (immature or male fish), or by placing together two immature fish. However, these latter methods may take several months to obtain mature pairs. Another alternative to collecting wild caught clownfish pairs or trying to pair up individual fish, is to purchase pairs of tank-raised clownfish. A U.S. company called The Breeder's Connection, a division of Aquatic Technologies, offers medium to large pairs of tank-raised clownfish of the following species: *Amphiprion bicinctus*, *A. clarkii*, *A. ephippium*, *A. frenatus*, *A. ocellaris*, *A. percula*, *A. sandaracinos*, and *Premnas biaculeatus*. Although this option may seem expensive or impractical for a hatchery in Micronesia, it may be a good source of broodstock of clownfish species that are not found locally and not frequently imported, such as *A. bicinctus* and *A. ephippium*.

Broodstock tanks of about 140 to 160 liters should be adequate for conditioning and spawning most species of clownfish. Using tanks that are larger than required adds to the initial cost of construction and makes for less efficient use of resources. An exception is *Amphiprion chrysopterus*, which is reported to require a larger tank for spawning (Young, 1991A). Broodstock of this clownfish species should probably be kept in tanks of at least 400 liters. Larger or more active species of marine aquarium fish may also require tanks that are 400 liters or larger in size to stimulate spawning. Water depth may be as important as volume for some species, and deeper tanks may be desirable for species that exhibit a large spawning ascent during courtship.

Strong lighting is required to maintain healthy anemones. Relatively strong lighting (15,000 to 50,000 lux) is also desirable for broodstock and grow-out culture, and artificial lighting, preferably from full spectrum light bulbs, may be used to supplement natural sunlight if needed to reach the desired light intensity. Commercial hatcheries, such as C-Quest, and formerly Aqualife, use translucent fiberglass roofs that allow about 50% of incoming sunlight to enter the hatchery facility, which may result in hatchery light levels as high as 50,000 lux on a bright day in the tropics. Spawning occurred in the Blue-streak goby and Banded coral shrimp at maximum light levels of only about 1,000 to 1,500 lux. However, higher light levels may have resulted in better maturation and more frequent spawning. Natural sunlight has been reported to aid in maintaining good coloration in marine aquarium fish (McLarney, 1985). However, high light intensities occurring under direct sunlight (e.g., about 75,000 to over 100,000 lux) will stimulate rapid growth of filamentous algae and add to the maintenance. Clownfish held in tanks with excessive filamentous algae may develop more subdued or washed out coloration. Larvae of at least some species of marine fish are reported to be sensitive to strong lighting (Wassink and Brons, 1990), and reduced light levels (1,000 to 5,000 lux) should be adequate, and in some cases required for obtaining optimal results in larviculture.

Good nutrition is required for broodstock to promote frequent spawning, large spawn size, and healthy eggs and larvae. Marine aquarium species have high nutritional requirements because of the high frequency of spawning, and a high quality balanced diet is needed in addition to a diet of fresh or frozen and live seafoods to achieve optimal results (D. Fletcher, per. comm., 1996). However, a commercial diet that meets all the

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nutritional requirements for breeding marine aquarium species does not appear to be readily available at this time.

The low level of spawning in the clownfish held at the GADTC may be due primarily to ineffective conditioning of broodstock as a result of deficiencies in broodstock nutrition. There may have been insufficient quantities of vitamins or other micronutrients and essential fatty acids in the frozen diet fed to the broodstock. The frozen diet used in this project was relatively low in lipids, and a higher level of fat may be required for optimizing broodstock conditioning and juvenile grow-out (H. Ako, per. comm., 1996). Live foods, especially when enriched with fatty acids and other nutrients, are an excellent food for conditioning fish for spawning. However, they were often not available as a supplement for feeding the broodstock during this study. Also, the size and quantity of feedings may not have been adequate for maturation and spawning. Due to restraints in staffing and other resources, the broodstock were usually fed only twice daily with the frozen diet or dry feed. Commercial clownfish hatcheries typically provide four feedings daily to broodstock, including at least one feeding with live food.

Broodstock should be fed four times daily, using a high quality frozen diet and enriched live adult *Artemia* or other enriched live food. The frozen diet should have high levels of highly unsaturated fatty acids (HUFA) and particularly DHA (docosahexaenoic acid), carotenoids (astaxanthin), and vitamins (especially vitamin C in a stabilized form). A good source of carotenoid pigments are two species of krill (*Euphausia superba* and *E. pacifica*). Astaxanthin and other carotenoid pigments can also be obtained commercially from at least one pharmaceutical manufacturer. The addition of spirulina may also provide pigments in the diet. Deficiencies in HUFA, especially DHA, have been shown to result in lower stress resistance and lower egg quality. Carotenoids are important for enhancing coloration and may have nutritional benefits as well, possibly as antioxidants.

Clownfish juveniles may be moved from the larval tanks to nursery or grow-out tanks after about three weeks, but it is probably better to wait until day 30 before moving them. Very young juveniles are more susceptible to handling stress, especially if they have any nutritional deficiencies, and losses could occur during the transfer process. Relatively high levels of dietary DHA have been shown to reduce stress-related mortality in marine fish postlarvae (Ako et al., 1994). Adequate DHA levels can be supplied to larval clownfish through the use of commercial enrichment diets to enrich rotifers and *Artemia* with this HUFA. One feed manufacturer has developed a diet for mahimahi that may give good results for grow-out of marine aquarium fish and another company has an *Artemia* flake diet enriched with HUFAs that has worked very well for improving broodstock performance of freshwater ornamental fish (H. Ako, per. comm., 1996). A frozen *Artemia* product, enriched with HUFAs, is also available commercially and may be a good supplement or ingredient in broodstock and grow-out diets for marine aquarium species. Lipid levels should be above 15% and perhaps as high as 25-30% to optimize juvenile growth and broodstock egg production. The grow-out diet should also be fortified with vitamins (especially with high levels of vitamin C) and high levels of astaxanthin (e.g., 100 ppm or higher of astaxanthin).

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Grow-out of juvenile clownfish was not optimized in this study, although some improvement was made later in the project. Market size fish should be produced within approximately 3 to 4 months of grow-out under good conditions. Slow growth in some batches of clownfish probably resulted from inadequate feeding levels and nutrition. Fish in grow-out were only fed twice each day, whereas in most commercial hatcheries the grow-out fish are usually fed four times per day. The method of feeding the frozen diet was also not very efficient (i.e., larger pieces of food often fell to the bottom and were not eaten by the fish), primarily due to the lack of proper equipment to grind the food to a small uniform size. As a result, the amount of food consumed at each feeding was probably less than required for good growth. The nutritional value of the grow-out diet was not optimized in this study and should be improved. However, some improvements were made in feeding the juveniles during the course of the study, and better growth was obtained. Fish should be size-sorted at frequent intervals during the grow-out period to prevent inhibition of growth in the smaller fish. Deformed fish should also be culled out as soon as possible during the grow-out period (i.e., as soon as the fish are large enough for deformities to be detected). Early culling will decrease overall costs of production and provide more space and faster growth in the remaining fish.

The higher incidence of deformities in juvenile clownfish from a couple of the latter spawns reared may have been due to a nutritional deficiency in the broodstock diet, because vitamins were often omitted from the frozen broodstock diet during the last several months of the grant. Fletcher and Wilson (1996) found that vitamin C deficiencies in the diet of *Amphiprion clarkii* broodstock resulted in significant increases (36% to 83%) in the incidence and severity of certain deformities. In addition, the 24 hour post-hatch larval survival was 10% to 15% lower in these fish.

However, deformities may have also occurred due to problems in larval rearing. Instant Ocean Hatcheries found that individual tanks of fish frequently had 50% to 80% salable fish and 80% to 98% survival, while other tanks had 10% to 20% salable (Hoff, In Prep.). However, since both types of results often came from fish from the same spawns, it appears that the problem occurred due to rearing conditions in the individual tanks (probably a combination of water quality and feeding/nutrition problems). Enrichment or booster diets that contain vitamins can be used to increase the vitamin level in rotifers and *Artemia* fed to the larvae, and this may also aid in reducing deformities and increasing overall health of the larvae. The time around metamorphosis appears to be a particularly sensitive time, where suboptimal conditions can lead to increased incidence of deformities (F. Hoff, per. comm., 1996). The low incidence of deformities in fish from most of the spawns cultured at the GADTC suggests that problems of deformities can be kept at a very low level if time and resources can be put into optimizing diets and rearing conditions.

Coloration was good during the first half of the study, when fresh shrimp was available for use in the clownfish diet. The shrimp was believed to provide carotenoid pigments in the diet. However, shrimp was not available later in the study and this probably contributed to

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the reduction in bright coloration observed towards the end of the project. This coloration problem can easily be corrected if sufficient quantities of carotenoid pigments are put back into the diet.

The Zebra goby pair spawned regularly throughout the study, indicating that the broodstock diet and maintenance were adequate for promoting spawning. However, there were a couple periods of time when the interval between spawns was extended to about twice the normal interval. This may have been due to inadequate broodstock nutrition during certain periods, or it may have been a natural period of reproductive inactivity.

The Blue-streak goby pair spawned regularly before being moved to another smaller tank. This disturbance probably disrupted their spawning behavior. One of the fish also became noticeably thinner after the move, and may not have been getting enough food to maintain it in spawning condition. It is also possible that spawning may be seasonal in this species, although most reef fish appear to have extended or year around spawning, especially in locations near the equator.

A variety of shapes and sizes of tanks can be used for larval rearing, including aquariums. However, round fiberglass or plastic tanks with a depth of thirty inches or more are standard for rearing larval marine fish, and probably are the most efficient design. The sides of the tank should be a dark color to provide a better visual contrast of food organisms against the background, and white bottoms allow for easier observation of the larvae by the larval culturist. Tanks of 300 liters or larger are preferable to smaller tanks for commercial culture situations.

The ss-strain of rotifer is now being mass-cultured on a routine basis at the GADTC and has been successfully used as a first food for larval rearing of *Amphiprion melanopus* and the early stages of *Stenopus hispidus*. The ss-rotifer is smaller than the s-strain of rotifer and should have better potential for rearing small larvae of some marine aquarium fish. However, larval rearing of the Blue-streak goby and Zebra goby remains unsuccessful, due to the lack of a suitable-sized live food during the first few days of feeding. The larvae of both species of goby are apparently too small to eat ss-type rotifers at first-feeding, and complete mortality always occurred after a few days, probably due to starvation. The larvae of many other species of marine aquarium fish have been reported to be too small to eat rotifers at first-feeding, and there have also been reports of the larvae of some species (e.g., *Pomacanthus* sp.) refusing to eat rotifers even though the larvae are large enough to eat them (Young, 1994A and 1994B). Sieving the smallest rotifers out of the GADTC cultures was not attempted, but may give better results than using the entire size range of the rotifer cultures.

Sea urchin larvae were not obtained on a dependable basis, and adequate quantities were not available to accurately evaluate its potential for rearing goby larvae and other small fish larvae. It is not known whether problems in spawning the sea urchins later in the study was due to nutritional deficiencies in the sea urchins held at the GADTC, or if spawning in this species is seasonal. Also, the sea urchin larvae grow fairly rapidly and need to be fed soon after hatching (Figures 12 and 13). Fertilized sea urchin eggs may also have potential

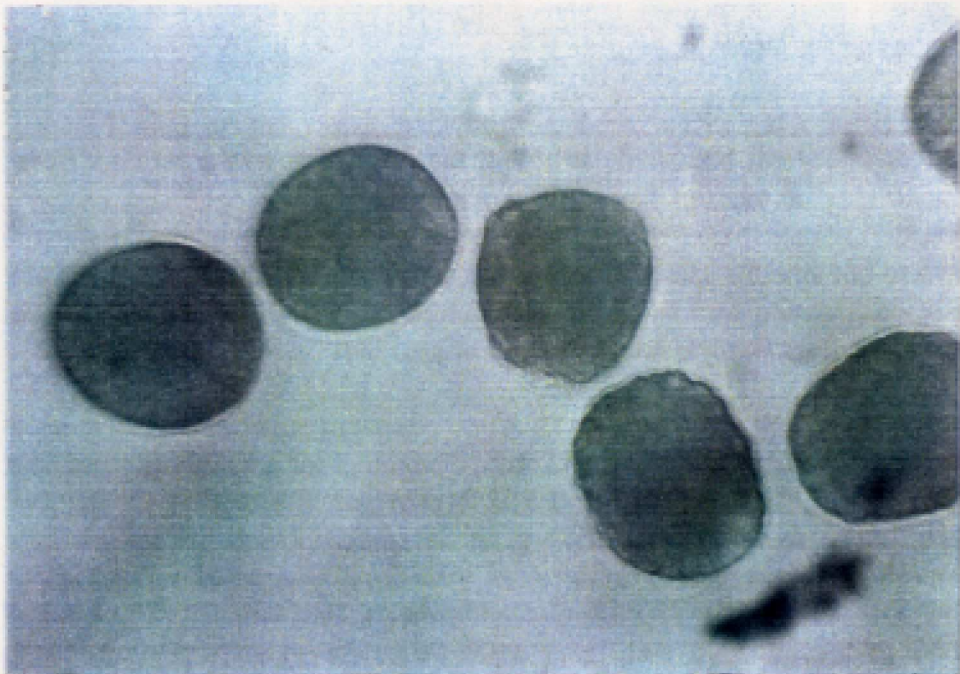


Figure 12. Swimming stage of the sea urchin (*Echinometra mathaei*) at 9 hours after fertilization. Size is about 60-70 microns.

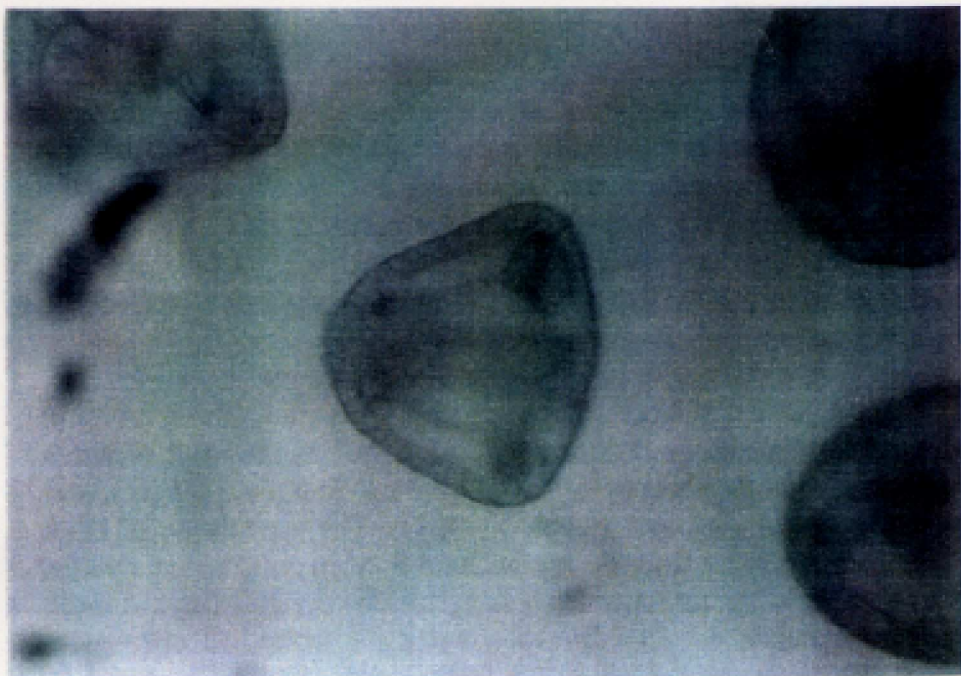


Figure 13. Early Pluteus stage sea urchin larvae at 28 hours after fertilization. Size is about 100-120 microns.

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as a live food, but some method of suspending the eggs in the water column is needed that will not damage the larvae.

Similar problems were encountered in producing oyster larvae. Induced spawning was only successful on two occasions, although the procedure was attempted over 20 times. Attempts to dissect out the gonads and manually mix the eggs and sperm were also unsuccessful, because there was very little eggs or sperm in the gonads, indicating that the oysters were not ripe for spawning. The species of oysters found on Guam are reported to have some spawning activity throughout the year, suggesting that a nutritional problem prevented them from being in spawning condition.

Oyster and sea urchin larvae may still have good potential as a live food for marine aquarium fish larvae, but methods need to be developed for conditioning and spawning the adults in captivity. Braley (1982 and 1984) provides information on the reproductive biology of these oysters. The major peak in spawning activity in the larger oyster *Crassostrea echinata*, occurred in March and the development of another was observed in late November. The smaller species, *Saccostrea cucullata*, was found to have a low level of continuous reproduction with three main peaks throughout the year, in November-December, March-April, and late June. The difficulty in spawning these oysters, especially *C. echinata*, may be due to natural seasonal reproductive periodicity. Oyster trocophores are reported to be a good first food for successful culture of some species with small larvae (e.g., Yellowtail snapper), but the larvae of some other species will not eat them (J. Holt, per. comm., 1996). Alternate live foods should be examined for culturing species with very small first-feeding larvae. Other possible live foods include cultured copepods, nematodes, and ciliates.

Fecundity in the Banded coral shrimp did not decline with time in captivity, indicating that broodstock maintenance and nutrition were adequate for spawn production. However, improvements could be made in the broodstock diet and feeding to maintain maximum output of quality larvae. The spawning data shows that the Banded coral shrimp are able to spawn every two to three weeks throughout the year. A few gaps in the spawning record for two of the pairs is more likely due to unreported or unobserved hatches rather than actual disruptions in their spawning rhythms.

Although only a limited number of rearing trials were attempted, larval rearing of the Banded coral shrimp improved during the study, and in the most successful trial some larvae appeared healthy and probably would have survived much longer if the tank had not become contaminated with hydroids and jellyfish. The appearance of large numbers of hydroids and small jellyfish medusae in the larval Banded coral shrimp tanks shortly before complete mortality of the larvae strongly suggests that the hydroids and medusae were the cause of the mortality, possibly due to stinging the larvae. However, the larvae were bigger than the medusae and no larvae were observed to be captured by them. It is unlikely that the hydroids and medusae fouled the water or ate all the *Artemia*, since the tanks were on a continuous flow-through of seawater at the time and there was an abundance of *Artemia* in the tank. The hydroids and jellyfish medusae have occasionally

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been observed in *Macrobrachium* larval rearing tanks at the GADTC, and may come in with the seawater from the saltwater wells. Contamination of the larval tanks with hydroids and jellyfish was successfully prevented in the last couple of larval rearing trials by filtering the water with a five micron filter bag. However, only *Artemia* was used as the larval food in these trials to avoid possible contamination from the rotifer cultures, and the larval shrimp experienced relatively large mortalities in the first two weeks and the trial was discontinued. Better survival of Banded coral shrimp larvae early in trials where rotifers were fed suggests that the rotifers were more nutritious than *Artemia* nauplii for the larvae or that the larvae were having difficulty capturing and consuming enough of the *Artemia*.

No special environmental cues are required to initiate metamorphosis in Banded coral shrimp (D. Jones, per. comm., 1995). However, the larval period is very long compared to most fish or shrimp larvae, and laboratories that have successfully reared Banded coral shrimp larvae through to the juvenile stage report metamorphosis occurring after more than 100 days (D. Fletcher, per. comm., 1996; L. Creswell, per. comm., 1996). The extremely long larval period of the Banded coral shrimp makes the commercialization of this species more difficult compared to other more valuable aquarium shrimp with shorter larval periods. In addition to the added time and expense of raising species with long larval periods, there is a higher probability of problems developing before the larvae reach the juvenile stage (L. Creswell, per. comm., 1996). The larvae also appear to be very sensitive to nutritional deficiencies (D. Fletcher, per. comm., 1994).

Objective 5. There appears to be a growing trend towards tighter restrictions on the harvest and import/export of marine tropical fish and invertebrates. This trend should ultimately work to the advantage of the commercial hatcheries, with the reduced availability of wild-caught fish making the tank-raised fish more competitive in the market. It is quite possible that at some time in the future, the harvest and sale of wild-caught reef fish and invertebrates will be completely banned for those species that are being cultured in captivity. Pyle (1993) provides additional information on the status of the marine aquarium industry in the Pacific region.

Objective 6. Mariculture certification does not appear to pose a problem for the growth of the industry. Government certification of approved hatchery facilities, with periodic inspections could insure that marine aquarium species are captive-bred.

Objective 7. A clownfish hatchery and grow-out facility can be profitable if a reliable production of marketable juveniles can be achieved and all marketable fish produced can be sold at a minimum price of \$2.25 each. The business could be more profitable if several species of clownfish were raised, especially less common species, to achieve more diversity and a slightly higher average price. Higher prices for market size clownfish are possible, and this would result in a higher level of profitability. Reef Propagations is obtaining between \$3.25 and \$4.00 per fish (J. Lichtenbert, per. comm., 1996), and Desert Fisheries is obtaining \$6.00 per fish (L. Nelson, per. comm., 1996). A hatchery for marine aquarium fish would also be more profitable if some production of other higher value

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species (e.g., *Pseudochromis* sp. or the Comet) was added to supplement clownfish production and achieve a higher average price for fish produced (perhaps two or three active pairs of each of the other species). Inactive broodstock should be replaced quickly with active spawning clownfish or other species. Additional low cost, low maintenance holding tanks could be added to keep new broodstock in reserve. Examples of species that could be cultured in a commercial hatchery in Micronesia and their estimated farm gate value are listed below:

Micronesian Species	Farm Value *
<i>Amphiprion bicinctus</i>	\$ 3.75
<i>A. chrysopterus</i>	\$ 4.00
<i>A. clarkii</i>	\$ 2.25
<i>A. ephippium</i>	\$ 3.75
<i>A. melanopus</i>	\$ 2.25
<i>A. perideraion</i>	\$ 2.50
<i>Pseudochromis porphyreus</i>	\$10.00
Imported Species	Farm Value *
<i>Amphiprion ocellaris</i>	\$ 2.50
<i>A. frenatus</i>	\$ 2.50
<i>Calloplestiops altivelis</i>	\$10.00
<i>Premnas biaculeatus</i>	\$ 3.50
<i>Pseudochromis dutoiti</i>	\$15.00
<i>P. flavivertex</i>	\$10.00
<i>P. fridmani</i>	\$10.00
<i>Hymenocera picta</i>	\$ 9.00
<i>Lysmata debelius</i>	\$10.00
<i>Lysmata grabhami</i>	\$ 7.00

* Estimated or potential value to producer based upon information obtained from commercial producers or wholesale price lists. When prices varied between producers, the lowest price normally obtained was used for these estimates.

Some of the higher-priced species are harder to produce (e.g., *Amphiprion chrysopterus*, *Calloplestiops altivelis*, and *Lysmata debelius*) or can only be marketed in small quantities (e.g., *Pseudochromis* sp. and *Hymenocera picta*). However, the production of a small quantity of these higher-priced species in addition to the more easily cultured and lower-priced species can improve the economic return and the larger product diversity will facilitate marketing efforts.

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RECOMMENDATIONS FOR CULTURE OF MARINE AQUARIUM SPECIES:

- Collect naturally mated pairs from the wild when possible.
- For clownfish broodstock, try to collect the anemone the pair was occupying or at least another anemone of the same species.
- If mated pairs of clownfish cannot be collected, place together one large clownfish (i.e., a female) with a small clownfish (immature or male fish) less than 4 cm in length.
- Use broodstock tanks (aquariums) of 120 to 160 liters for clownfish broodstock. For *Amphiprion chrysopterus*, use a tank of at least 400 liters.
- Use strong lighting (i.e., light intensity of 15,000 to 50,000 lux) for broodstock and grow-out. Use natural sunlight or full-spectrum bulbs in artificial lighting.
- Use lower light levels (i.e., light intensity < 5,000 lux) for larval culture.
- Feed broodstock four times daily, using a high quality frozen diet and enriched live adult *Artemia* or other enriched live food. A high quality dry diet formulated for marine fish culture may be substituted for one feeding, if available.
- Frozen diets for broodstock and grow-out should have lipid levels above 15% and high levels of HUFAs (particularly DHA), carotenoids (primarily astaxanthin at 100 ppm or higher), and vitamins (especially vitamin C and vitamin E).
- Use round tanks with depth of thirty inches or more for larval rearing. The tanks should have dark sides and a white bottom.
- Feed larval clownfish enriched rotifers for the first ten days, and *Artemia* nauplii from day 7 to day 15. Enriched 24-hour *Artemia* and small particle size artificial diets should be fed beginning at about day 15. This feeding schedule may need to be adjusted based upon growth rate of the larvae.
- Use live foods (e.g., rotifers and *Artemia*) that are enriched with HUFAs and vitamins whenever possible in larval culture.
- Use microalgae during the rotifer feeding stage of larval culture. Species that can be used include *Chaetoceros gracilis*, *Isochrysis* sp., *Nannochloropsis oculata*, *Rhodomonas* sp. and *Tetraselmis* sp.
- Use daily water exchange or continuous flow-through of seawater after the larvae are feeding on *Artemia*.
- Move clownfish juveniles from larval tanks to nursery or grow-out tanks at 25 to 30 days. Move a few fish and watch for handling stress problems before moving all the fish in a tank.
- Stock larval tanks at no more than 5 fish per liter.
- Stock grow-out tanks at no more than one fish per liter.
- Feed fish four times per day in grow-out, with two feedings of a high quality frozen diet and two feedings of a high quality dry diet (flake or crumble) formulated for marine fish.
- Young juvenile clownfish should receive a supplement of live enriched *Artemia* for the first two to three weeks after they are transferred to the nursery or grow-out tanks.
- Sort juveniles for size at frequent intervals during grow-out.
Examine juveniles at an early age for deformities and continue checking them throughout the grow-out period. This can be done when size-sorting the fish.

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Deformed fish should be culled out as soon as possible and not offered for sale (except possibly as feeder fish).

- Off-color fish should be fed diets with higher levels of carotenoids for 4 to 6 weeks before they reach market size (3 to 4 mm) to improve coloration.
- Keep excessive filamentous algae growth out of all rearing tanks.
- Use live foods (e.g., rotifers, *Artemia*, nematodes) enriched with HUFAs, vitamins and carotenoid pigments for larval culture of marine aquarium shrimp.

CONCLUSIONS:

Advances have recently been made in the culture of marine aquarium fish and invertebrates, as reflected in the literature and the increasing diversity of species under commercial cultivation. Although some major marine aquarium fish hatcheries have closed operations in the last few years, the remaining commercial and research facilities are improving their production capabilities and economic potential. Increased diversity of species and production of more highly-valued species are positive trends occurring in this industry.

Several marine aquarium species occurring in Micronesia have been identified as prime candidates for aquaculture production. Some of these, such as the clownfish species, some of the gobies, the Comet, and the Magenta dottyback, have been successfully cultured in other parts of the world. Other valuable Micronesian species may prove to be conducive to captive rearing with additional research.

The Cinnamon clownfish *Amphiprion melanopus*, was successfully cultured in this project and demonstrates the feasibility of marine aquarium fish culture in Micronesia. Spawning was achieved for two species of goby, although the tiny larvae were not successfully reared. A live food that is significantly smaller than the ss-strain of rotifer will be required in large quantities to rear these species and other valuable marine aquarium fish that have very small first-feeding larvae. Problems in obtaining mated pairs of clownfish can be solved through increased efforts at collecting mated pairs from the wild or by collecting small juveniles and pairing them with other juveniles or adult fish (although these latter methods will take more time).

Better quality prepared diets and enriched live feeds, as well as improved feeding methods can be used to properly condition the broodstock pairs to achieve better spawning performance, as well as improved larval quality and survival through first-feeding stages. New live feeds for species with very small first-feeding larvae are currently being developed by a private company in the United Kingdom, increasing the potential for larval culture of many reef fish (D. Fletcher, per. comm., 1996).

Good growth was obtained with one batch of clownfish juveniles, and market size (3 to 4 cm) fish can be obtained after three to four months if attention is given to optimizing nutrition and feeding. Marketability, in terms of coloration and appearance (i.e., lack of

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deformities) was high with most batches of clownfish juveniles reared in the study, and high quality fish can again be obtained by optimizing nutrition during all phases of production.

Regular spawning of captive broodstock of the Banded coral shrimp occurred during much of the study and consistent production of newly hatched larvae should not be a problem for their culture. However, larval rearing of this species is a long and difficult process, and the potential for their eventual commercial production is poor. Other species of marine aquarium shrimp, such as *Lysmata debelius* and *Hymenocera picta* are much more promising candidates for commercial culture.

Regulatory constraints for the import/export of marine aquarium species will increase in near future, but this trend towards restricting the collection of wild-caught animals favor should work in the favor of captive breeding programs. Mariculture certification of cultured aquarium fish can be done without much difficulty through certification and monitoring of government-approved culture facilities.

Competition in the aquarium market with cheaper wild-caught fish and shrimp presently makes commercial culture of marine aquarium fish and shrimp more difficult economically. However, the commercial culture of various marine ornamental species could be profitable in Micronesia if the business is set up properly and well managed. The future for commercial marine aquarium fish culture looks even brighter, as more species come under culture and the regulatory climate restricts the collection of wild-caught reef fish.

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APPENDIX I. FROZEN DIET FOR MARINE AQUARIUM FISH

GEL MIX RECIPE (MODIFIED WAIKIKI AQUARIUM RECIPE)

1.00 kg fish (marine fish preferred: smelt, reef fish, etc...) *
1.00 kg squid *
0.50 kg trout chow
0.35 kg shrimp ***
0.85 kg vegetables
50 g yeast
3.8 g Vitamin C

3.8 kg subtotal
3.8 L water
7.6 kg TOTAL (Before Gelatin)

760 g Knox Gelatin (10%)

* Other seafood items can be substituted for part of the fish or squid, including the following: clams, oysters, scallops, marine bloodworms, crab meat, and fish eggs.

** Combination of two or more of the following: zucchini, spinach, carrots, green peas, or romaine lettuce.

*** Use whole, shell-on shrimp. Using only shrimp heads or a too high of a percentage of shrimp will cause the mix not to gel with 10% knox gelatin.

DIRECTIONS:

Measure out 1750 ml of water to be used in the grinding process. The remainder of the water (2050 ml) can be set to heat for dissolving the gelatin. Chop all ingredients into small pieces and grind in the blender with sufficient water. Add ingredients in about four equal parts with the addition of water each time and using a spatula to push solid pieces toward the blades.

Put gelatin in a large mixing bowl, add the boiling water (2050 ml), and mix with the electric mixer, scraping the sides and bottom often. After thoroughly dissolved, allow the mixture to cool to lukewarm, mixing occasionally to hasten cooling and keep gelatin from lumping. When cooled, add ground ingredients to dissolved gelatin and mix. Next add the Vitamin C and the yeast -- but only when the ingredients are lukewarm or cooler -- and continue mixing.

Pour this mixture into 4 to 5 trays and allow to set in the freezer for 12 to 15 minutes. Remove trays before the mix freezes and cut into squares. Package the squares in the zip lock bags and store in the freezer.

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APPENDIX II Aquarium Hobby Literature on the Culture of Marine Aquarium Fish and Shrimp

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APPENDIX V. List of Marine Aquarium Fishes Reared in Captivity

Species	Common Name
<i>Abudefduf abdominalis</i>	Maomao damselfish
<i>Abudefduf caeruleus</i>	Electric blue damselfish
<i>Abudefduf cyaneus</i>	Blue devil damselfish
<i>Abudefduf saxatilis</i>	Sargeant major
<i>Acanthostracion quadricornis</i>	Cowfish
<i>Acanthurus leucosternon</i> *	Powder-blue tang
<i>Amblygobius phalaena</i>	Banded goby, Brown-banded goby, Dragon goby
<i>Amblygobius rainfordi</i>	Rainford's goby
<i>Amphiprion akallopisos</i>	Skunk clownfish
<i>Amphiprion akandynos</i>	Barrier Reef clownfish
<i>Amphiprion allardi</i>	Allard's Anemonefish
<i>Amphiprion bicinctus</i>	Two-banded clownfish or Red Sea clownfish
<i>Amphiprion chrysopterus</i>	Orange-fin clownfish or Blue-stripe clownfish
<i>Amphiprion clarkii</i>	Clarkii clownfish
<i>Amphiprion ephippium</i>	Red saddleback clownfish
<i>Amphiprion frenatus</i>	Tomato clownfish
<i>Amphiprion latezonatus</i>	Wide-band clownfish or Lord Howe clownfish
<i>Amphiprion melanopus</i>	Cinnamon clownfish
<i>Amphiprion nigripes</i>	Black-finned clownfish
<i>Amphiprion ocellaris</i>	Common clownfish
<i>Amphiprion percula</i>	Percula clownfish
<i>Amphiprion perideraion</i>	Pink skunk clownfish
<i>Amphiprion polymnus</i>	Saddleback clownfish
<i>Amphiprion rubrocinctus</i>	Australian clownfish
<i>Amphiprion sandaracinos</i>	Orange skunk clownfish
<i>Amphiprion sebae</i>	Seba's clownfish
<i>Amphiprion tricinctus</i>	Three-band clownfish
<i>Anisotremus virginicus</i>	Porkfish
<i>Apogon sp.</i>	Cardinalfish
<i>Apogon compressus</i>	Blue-eyed cardinalfish or Split-banded cardinalfish
<i>Balistoides conspicillum</i> *	Clown triggerfish
<i>Bodianus pulchellus</i>	Spotfin Hogfish
<i>Bodianus rufus</i>	Spanish hogfish
<i>Calloplelesops altivelis</i>	Comet (Marine betta)
<i>Chaetodipterus faber</i>	Atlantic spadefish
<i>Chrysiptera cyanea</i>	Orange-tail damsel
<i>Chrysiptera parasema</i>	Gold-tail demoiselle
<i>Cryptocentrus cinctus</i>	Yellow prawn goby
<i>Dascyllus albisela</i>	Threespot damselfish
<i>Dascyllus aruanus</i>	Whitetailed damselfish
<i>Diodon holacanthus</i>	Balloonfish or porcupinefish
<i>Equetus acuminatus</i>	High-hat

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APPENDIX V. List of Marine Aquarium Fishes Reared in Captivity (Continued)

<i>Equetus lanceolatus</i>	Jacknife fish
<i>Equetus punctatus</i>	Spotted drum
<i>Equetus umbrosus</i>	Cubbyu
<i>Glyphidodontops hemicyaneus</i>	Yellow-tailed damselfish
<i>Gobiodon citrinus</i>	Citron goby or Clown goby
<i>Gobiodon okinawae</i>	Okinawa goby or Lemon or Yellow clown goby
<i>Gobiosoma evelynae</i>	Sharknosed goby
<i>Gobiosoma genie</i>	Genie's cleaning goby
<i>Gobiosoma macrodon</i>	Tiger goby
<i>Gobiosoma multifasciatum</i>	Greenbanded goby
<i>Gobiosoma oceanops</i>	Neon goby
<i>Gobiosoma puncticulatus</i>	Redhead goby
<i>Gobiosoma xanthiprora</i>	Yellowstripe cleaning goby
<i>Gramma loreto</i>	Royal gramma
<i>Gramma melacara</i>	Black cap basslet
<i>Hippocampus comes</i>	Seahorse
<i>Hippocampus erectus</i>	Northern seahorse
<i>Hippocampus fuscus</i>	Seahorse
<i>Hippocampus hystrix</i>	Thorny seahorse
<i>Hippocampus kuda</i>	Oceanic seahorse
<i>Hippocampus novaehollandiae</i>	Australian seahorse
<i>Hippocampus</i> sp.	Lattice seahorse
<i>Hippocampus zosterae</i>	Dwarf seahorse
<i>Hypoplectrus gemma</i>	Blue hamlet
<i>Hypoplectrus unicolor</i>	Butter hamlet
<i>Hypsypops rubicundus</i>	Garibaldi
<i>Liopropoma rubre</i>	Peppermint basslet
<i>Lythrypnus dalli</i>	Catalina goby
<i>Meiacanthus grammistes</i>	Striped blenny
<i>Meiacanthus nigrolineatus</i>	Black-lined blenny
<i>Microspathodon chrysurus</i>	Jewelfish or Yellow-tailed damselfish
<i>Opistognathus aurifrons</i>	Yellowhead jawfish
<i>Pholidichthys leucotaenia</i>	Convict blenny
<i>Phycodurus eques</i>	Leafy seadragon
<i>Phyllopteryx taeniolatus</i>	Common seadragon
<i>Platax orbicularis</i> *	Circular spadefish or Round batfish
<i>Pomacanthus arcuatus</i>	Grey angelfish
<i>Pomacanthus imperator</i> *	Emperor angelfish
<i>Pomacanthus paru</i>	French angelfish
<i>Pomacentrus caeruleus</i>	Yellow-bellied blue damsel
<i>Premnas biaculeatus</i>	Maroon clownfish
<i>Pseudochromis dutoiti</i>	Neon dottyback
<i>Pseudochromis flavivertex</i>	Sunrise dottyback

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APPENDIX V. List of Marine Aquarium Fishes Reared in Captivity (Continued)

<i>Pseudochromis fridmani</i>	Orchid dottyback
<i>Pseudochromis paccagnellae</i>	Royal dottyback or Bicolor dottyback
<i>Pseudochromis porphyreus</i>	Magenta dottyback
<i>Pseudochromis springeri</i>	Springer's dottyback
<i>Pterosynchiropus splendidus</i>	Mandarinfish
<i>Serranus subligarius</i>	Behed sandfish
<i>Siganus canaliculatus</i>	White-spotted spinefoot (rabbitfish)
<i>Signigobius biocellatus</i> *	Signal goby or Two-spot goby
<i>Zanclus cornutus</i> *	Moorish idol

* Reports of this species being successfully cultured need to be verified.

APPENDIX VI. List of Marine Aquarium Shrimp Reared in Captivity

Scientific Name	Common Name
<i>Lysmata amboinensis</i>	Scarlet cleaner or White-striped cleaner shrimp
<i>Lysmata californica</i>	Catalina cleaner shrimp
<i>Lysmata debelius</i>	Blood or Fire shrimp or Cardinal cleaner shrimp
<i>Lysmata rathbunae</i>	Peppermint shrimp
<i>Lysmata seticaudata</i>	Monaco cleaner shrimp
<i>Lysmata wurdemanni</i>	Peppermint shrimp or Caribbean cleaner shrimp
<i>Hymenocera picta</i>	Harlequin shrimp or Clown shrimp
<i>Rhynchocinetes uritai</i>	Rosy dancing shrimp
<i>Stenopus hispidus</i>	Banded coral shrimp

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