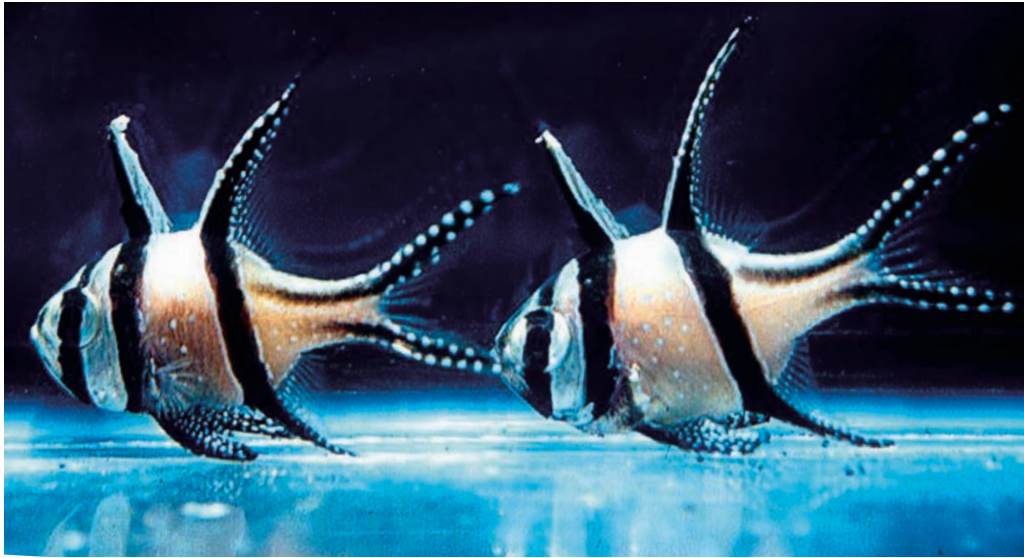


Manual for the Production of the Banggai Cardinalfish, *Pterapogon kauderni*, in Hawai'i



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Rain Garden Ornamentals is the first company in the world to be certified under the Mariculture and Aquaculture Management (MAM) Standard for fish production by the Marine Advisory Council (MAC). The procedures described herein are consistent with the MAM standards, although their use does not automatically bestow MAC certification.



Cover photo by Clyde Tamaru

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I. INTRODUCTION

As seen on the front cover, the Banggai cardinalfish, *Pterapogon kauderni*, is extremely attractive in appearance, very hardy in captivity, and exhibits an unusual mode of reproduction in that the males incubate their female partner's eggs in their mouth (Tullock, 1999; Vagelli, 1999). They make outstanding tank companions with all fish, coral and other marine ornamentals. For these reasons, they have become very popular in the marine ornamental trade (Michael, 1996). The remarkable rise in popularity of this species since 1995 has begun to raise concerns amongst the various stakeholders in the aquarium industry as currently between 50,000 and 118,000 individuals per month are collected from the wild and make their way to overseas markets (Vagelli and Edmann, 2002; Lunn and Moreau, 2004). What is cause for concern is that this species is endemic to the Banggai Archipelago in Indonesia with a geographic range which is estimated to be less than 10,000 km² (Fig. 1).

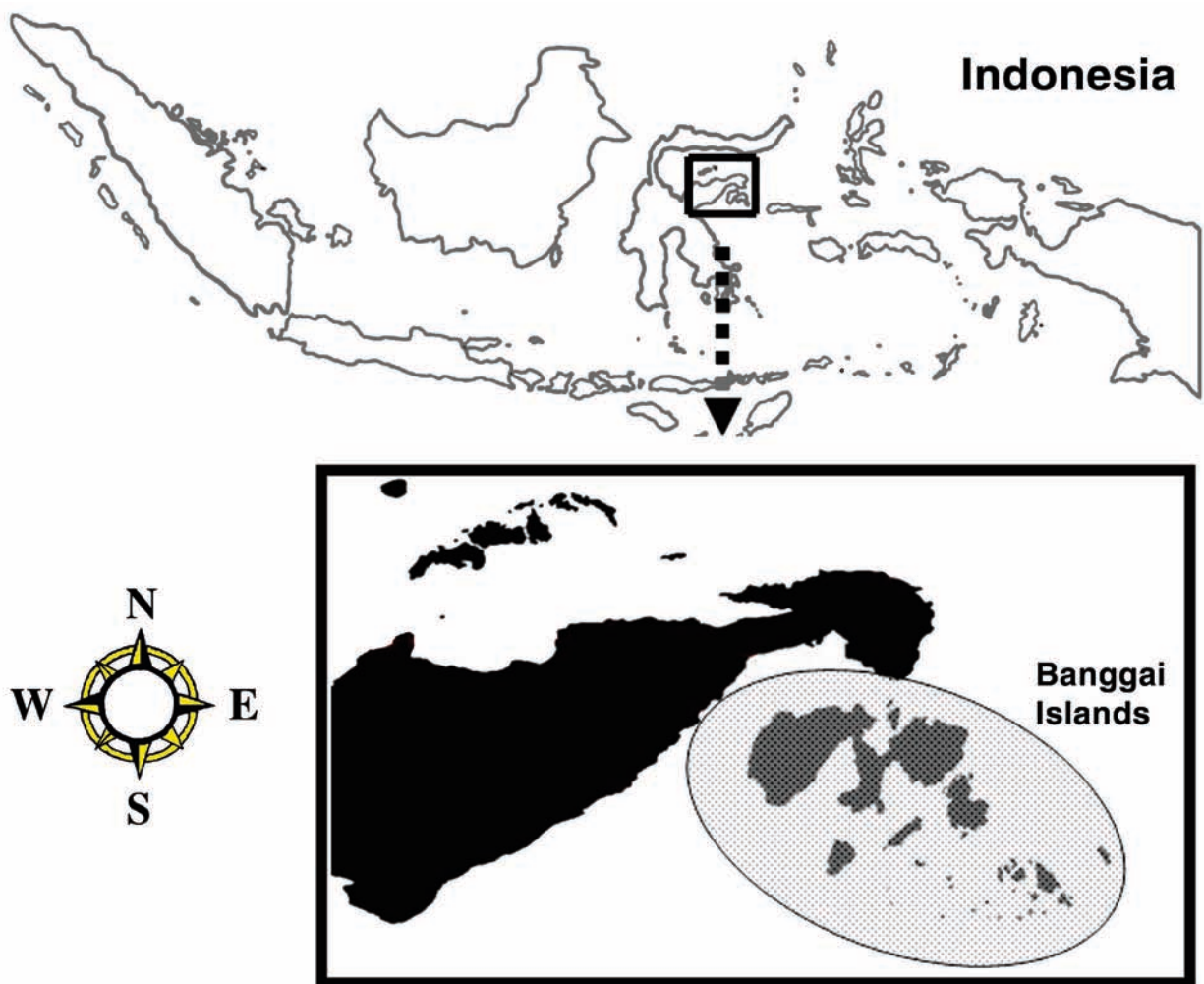


Figure 1. Geographic distribution of the Banggai cardinalfish. The stippled area represents the limited natural geographic range.

Certain aspects of the reproductive life history of this fish such as unusually low fecundity, lack of a pelagic larval phase, and strong site attachment are thought to be the primary reasons for this extremely limited geographic range (Allen, 2000). Even with the use of so-called “nondestructive” collection techniques, a clearly negative relationship is already demonstrated between fishing pressure on the density of this fish, on group sizes, and even some collateral impacts such as a decrease in density of the long-spined sea urchin, *Diadema setosum*, that the Banggai cardinalfish associates with in the wild (Kolm and Berglund, 2003).

The mounting body of evidence indicates that the natural populations of this species are clearly under threat due to the demands of the aquarium industry. In light of that information, the species has already been informally proposed for placement on the threatened species list (Allen, 2000). However, simply regulating the curtailment of the collection of wild-caught individuals is not an appropriate solution as the market demand for the species has not diminished, as socioeconomic issues would also need to be addressed. The collectors of the Banggai cardinalfish, and for that matter the majority of marine ornamentals originating from Southeast Asia, are typically small-scale fishermen working either alone or in groups to supply a wholesale distribution chain (Green, 2003; Olivier, 2003). While these individuals are usually paid very low prices for their products (in Indonesia US\$0.10/individual for *Amphiprion percula* and US\$0.03/individual for *Pterapogon kauderni*) they are also among the poorest of the poor income wise in Indonesia (Hemdal, 1984; Olivier, 2003; Lunn and Moreau, 2004). Collection of marine ornamentals for market provides a valuable source of income for this group of people.

With an estimated 99% of the marine ornamental products originating from wild-caught sources (Olivier, 2003), it is no wonder that the aquarium industry has attracted much controversy over the sustainability of the industry. While it is still very difficult to accurately weigh the costs and benefits of the marine ornamental industry, remedial actions can already be taken. One of these is to develop a means of artificially propagating a target species ultimately resulting in decreased dependence on wild-caught specimens. Artificial propagation would have the added benefit of providing more employment opportunities for coastal fishermen. Relevant production technology for *Pterapogon kauderni* forms the basis for this report.

This manual is not intended to be a repository of all that is known about the Banggai cardinalfish and its culture, but rather an attempt to document a process currently being practiced to commercially produce this species as part of a diversified tropical ornamental fish producing enterprise. It is hoped that the information will be useful to others to create or improve production capacity of this species with the long-term goal of relieving the dependence on wild-caught individuals. The production process described herein is economically viable in Hawai'i and requires little or no sophisticated equipment. It is a semi-intensive process which is forgiving in its day-to-day management and takes advantage of food stuffs produced on site. The technology may also be readily adopted by small family farms in coastal areas throughout the tropics.

II. SYSTEMATICS

The Banggai cardinalfish belongs to the phylum Chordata which also includes various groups of the subphylum, Vertebrata: fishes, amphibians, reptiles, birds and mammals (Fig. 2). Because this species has an internal skeleton that is ossified, a swim bladder, and has bony scales and gill slits covered by an operculum, it is placed in the class of fishes known as Osteichthyes. Within that class there is one order that exceeds all others in diversity, the order Perciformes, or perch-like fishes to which this species belongs. The perciform fishes are believed to include 148 families in approximately 1,500 genera, encompassing almost 10,000 species of fishes. The cardinalfishes of the family Apogonidae are characterized as possessing two separate dorsal fins. The first dorsal fin contains 6 to 8 spines and 8 to 14 soft rays in the second dorsal fin. There are two spines in the anal fin and the number of soft rays range between 8 and 18 (Nelson, 1994). Males are mouth-brooders with most species being nocturnal, feeding on zooplankton and small benthic invertebrates. This family is thought to be one of the largest families of fishes with about 27 genera and 250 species. The largest genus, *Apogon* (which means without a barb) accounts for the highest number of species in the family. There is, however, only one species, *kauderni*, found in the genus *Pterapogon* and that is the Banggai cardinalfish.

- Phylum: Chordata
 - Subphylum: Vertebrata
 - Class: Osteichthyes
 - Order: Perciformes
 - Family: Apogonidae
 - Genus:
Pterapogon
 - Species:
kauderni



Clyde Tamaru

Figure 2. Systematic relationship of the Banggai cardinalfish.

This species (also known as Banggai cardinals) was first described in 1920 from collections of fishes originating in waters surrounding Sulawesi in Indonesia (Koumans, 1933). It had remained unknown to western science when it first began to appear in 1995 and since then it has exploded into the marine ornamental trade (Michael, 1996).

III. GETTING STARTED

The information presented in this manual primarily includes the observations and experiences of the authors and represents insights obtained from a firsthand perspective. However, the procedures described have also been supplemented with data on the reproduction and early life history of this species that has largely been obtained from various attempts to produce this species in captivity both by hobbyists and researchers (Marini, 1996; Vagelli, 1999).

A. Establishing Broodstock

In acquiring Banggai cardinals for broodstock, it should be made a priority that captive-raised fish should be sought. From experience, it is clear that the cultured individuals easily adapt to a new facility and in the long run were found to be less expensive than wild-caught stocks. For example, in acquiring broodstock for the prototype system at the Rain Garden Ornamentals facilities, three captive-bred pairs were initially purchased. One of the six fish died after several months, but the others went on to be very productive. Wild fish were also purchased for broodstock purposes and on one occasion, 48 adults were purchased from a mail-order dealer. Of these 48 wild adults, only one lived long enough to reproduce in captivity and most died within a month. The causes of the mortalities are unknown but they could be attributed to stress and disease brought on by capture, handling and shipping, or possibly the after-effects of collection practices in Indonesia. If a disease agent was involved, it did not affect the captive-bred fish contained within the same system which was often separated from the wild fish only by a tank partition. Thus, there are economic as well as environmental reasons to seek out captive-bred and raised Banggai cardinals for reproduction.

B. Establishing Mating Pairs

The hatchery process begins by establishing mating pairs that can be reared in separate containers from which their reproductive outputs can be easily monitored and collected. However, developing compatible reproducing pairs can be a challenge as the species is reported to be lacking in any secondary sexual dimorphism and exhibits a 1M:1F sex ratio (Vagelli and Volpedo, 2004). Differentiating male and female Banggai cardinalfish is difficult with large adults and nearly impossible with small adults just reaching reproductive age. From our experiences, one characteristic that has been useful in the identification process is the squared-off jaw line of the male that gives it a “bull dog” appearance. However, there seems to be both skeletal and muscular aspects of this characteristic and the male’s jaw line and masculine appearance can change somewhat depending on his mood and current social interactions. It is easiest to discern male and female individuals in a pair after spawning has taken place because of the enlarged buccal cavity of the male as he incubates the spawned eggs in his mouth (Fig. 3). At this time, the male can be definitively identified as he will be the one who does

not eat and appears to have a mouth full of marbles. It should also be noted that when holding eggs/offspring, the male also becomes reclusive and withdraws to the back of the holding tank. Although this is of little use when trying to separate the sexes before they have spawned, once they have spawned, the individual's markings or spotting pattern will allow it to be identified and sexed, albeit after the fact.

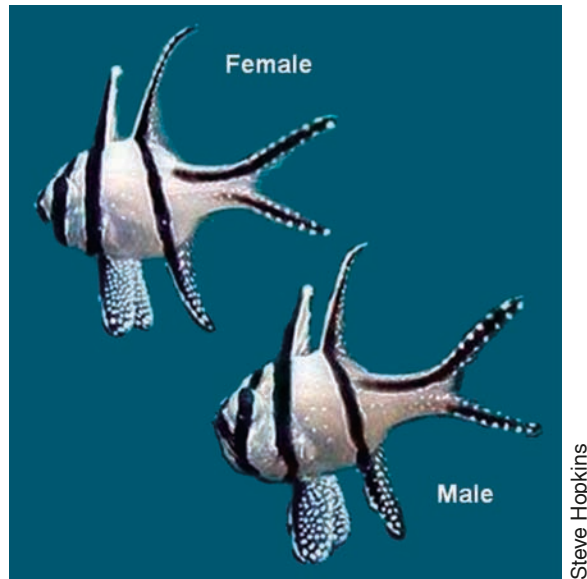


Figure 3. Photograph of female and male Banggai cardinalfishes.

It has been reported that pair bonding takes from several days to two weeks prior to spawning (Vagelli, 1999; Kolm and Berglund, 2004). It is not known whether the pair bond is strong in the wild but it appears to be transient in captivity and a female may be willing to mate with several different males (Vagelli, 1999). Nonetheless, finding compatible mates takes time and careful attention. Several approaches have been tried with varying degrees of success and are described below.

Approach #1: A group of sub-adults (four to six individuals) are placed together in a communal tank where they develop pair bonds as they mature. It has been reported that Banggai cardinals mature at 10 to 14 months of age in the wild (Marini, 1999). However, in developing broodstock from captive-bred offspring, we have found that Banggai can begin pairing off at four to five months of age and breed at six to seven months of age. In the wild, mating pairs establish spawning territories several meters away from the main group and rigorously defend them (Kolm and Berglund, 2004). Within the confines of the communal tank, aggressive interactions will erupt and must be monitored very carefully as the sub-adults mature. It is important to note that any Banggai cardinal seen with dark coloration and cowering in a corner must be removed immediately or it will soon die as a result of social stress and because the rival fish will

prevent it from feeding. Ideally, the communal tank would provide approximately one square meter of space per pair to minimize aggression and simplify the identification of mated pairs. As in the wild, a mated pair will often establish a small territory where they usually “hang out” although that territory can move around a bit. Catching a mating pair from a communal tank is the next challenge and it can be a rather difficult process. Swiping at them with a net will only result in losing track of which two are the pair. Swiping with a net will also excite and disturb the entire group, resulting in unnecessary stress. What has been found to be an effective method is to identify the fish in the late afternoon and return to the tank just after dark with a flashlight. The pair will usually be sleeping in the same general area, although the individuals that make up the pair may sleep a foot or two apart. If you hold the light on the fish, you can usually catch them with a 30 to 40-cm in diameter black net.

Approach #2: Another approach to developing broodstock pairs is to house individual fish in isolation and then selectively place two fish together while closely monitoring them to see if they are compatible. This is often the only option when potential broodstock are acquired as large adults. It should be mentioned that a group of large adults placed together will result in much social aggression and unnecessary mortalities. These aggressive interactions seem to begin as soon as the wild fish are placed in communal tanks or shipping containers. The aggressive behavior only intensifies as the fish become accustomed to their new surroundings.

In contrast, it is often comical to observe two identical-looking fish that are placed together for the first time. If one is male and the other female, when the male realizes that the other is a female he will often puff up his mouth and behave like he is carrying eggs. This behavior only lasts a few seconds and then he deflates his buccal cavity and the pair appears the same once again. The ability to inflate the buccal cavity apparently is entirely muscle control as there are no reported structural differences in their morphology (Vagelli and Volpedo, 2004). If two males are placed together, confrontation and aggression will usually be noticed within a few minutes. To a lesser extent, there may be aggression between large females and even between a male and female which, for some reason, are not compatible. Whenever any kind of aggressive behavior is detected, the two individuals must be separated and another individual introduced until compatible pairs have been identified and placed in appropriate broodstock tanks.

C. Signs to Watch For

When first introduced, a male and female will usually decide within an hour whether or not if they are compatible. They may start off by interacting as described above, only to suddenly have one fish dart after another and perhaps nip at the fins. If one fish darts with a simultaneous retreat by the other fish, they are unlikely to be compatible and the retreating fish is likely to be intimidated to the point of death over the coming days or weeks. With an incompatible pair, one fish (usually the larger of the two) will maintain normal coloration and move freely about the tank while the other fish will have a dark

coloration and be forced to cower in a corner. There may, or may not, be noticeable fin nipping, but the weaker, subservient fish will die irrespective of physical damage and for that reason it will need to be removed from the situation. In contrast, when first introduced, a compatible pair will tend to sit together for several hours.

D. Spawning Tanks

Once a compatible pair is found, they readily spawn in captivity as reported by both hobbyist and researcher alike (Marini 1996; Vagelli, 1999). Mating pairs are maintained in separate broodstock tanks. Obviously this avoids having to separate mating pairs again, avoids the territorial interactions, and eases the monitoring of behavior and reproductive outputs.

Reproducing pairs have been maintained in cages and glass aquaria with equal success. It is thought that the fish probably like the cages, but it makes observation of the male holding eggs difficult while in a cage as they can only be viewed from above. The smallest volume of the cage or tank that has been tried for holding mating pairs is 15-gal (60-L) or a 2-ft³ cage (approximately 60-L). Alternatively, a 29-gal (110-L) aquarium with a partition can be used to house two pairs. A 20-gal (76-L) aquarium works for a single mating pair and a 55-gal (210-L) aquarium with two partitions can be used to house three mating pairs. The partitions are made of plastic egg crate material and a shade cloth covering is used to maintain water movement between the partitions but not to have fish see their neighbors on the other side of the partition. Without the shade cloth, the males will be preoccupied with making sure the male next door is not trying to invade his area. Details about the construction and costs of the holding facilities are provided in a later section.

Each cage or tank is equipped with an air stone for continuous aeration. Aquaria are supplied with a constant supply of saltwater trickling in from a reservoir that consists of a nearby saltwater pond or large tank. The dilution factor and the macrophyte and microbial community which develops in the reservoir insures that water quality parameters such as ammonia, nitrite and nitrate do not deviate from zero. Using a pond or large tank as a reservoir has one disadvantage in that salinities may vary depending on the amount of rainfall. Salinities have been observed to fluctuate from 18 to 40 ‰ without changes in reproductive performance. Water temperature in Hawai'i ranges 20 to 24°C from November through April and between 25 and 28°C during the rest of the year. A summary of the annual variation in both environmental parameters is provided in the section on feeding and growth.

IV. REPRODUCTION

A. Spawning

Once adults are large enough to pair off they readily reproduce in captivity (Marini, 1998, 1999; Vagelli, 1999). Under tropical conditions, they will reproduce year-round as long as they receive a suitable diet and that topic will be covered in a later section. The actual spawning has reportedly taken place at night (Marini, 1999) and also during the daytime hours, e.g., 10:00-15:00 hr (Vagelli, 1999). As with other apogonids, the spawning event is preceded by several hours of elaborate courtship behaviors that have been described elsewhere (Kuwamura, 1983; 1985; Vagelli, 1999). Following the courtship activities actual spawning of *P. kauderni* that were observed and videotaped occurred between 13:00-15:00 hr (Vagelli, 1999). As described by Vagelli (1999) egg release and transfer was completed when both individuals were in very close proximity with one another (within one to two cm) and within a few centimeters above the bottom of the tank. At the time of spawning the male was usually situated ahead of the female and when approximately three-quarters of the egg mass was protruding from her body the male would turn and quickly gulp the clutch of eggs and pull them from the female. Although it has yet to be observed, it is assumed that sperm release must take place before the eggs were taken by the male as the egg mass when removed from the male's mouth a couple of hours after the spawning event are fertilized. The transfer of eggs from female to male occurs very rapidly, taking place within one to two seconds. It is reported that the removal of the eggs from the female requires a bit of pulling and not all of the eggs end up in the male's mouth, so it is common to see several eggs fall to the bottom of the tank. Between 10 and 20 eggs are reportedly lost during the egg transfer process. A male Banggai cardinalfish holding eggs in his mouth is easily distinguished from others by the distended (puffed-out) jaw line and the fact that he suddenly becomes reclusive and refuses to eat when offered food.

B. Fecundity

The size of the egg mass varies with the size (and probably condition) of the female. One egg mass which was removed from the male and examined was found to consist of 40 eggs (Fig. 4) averaging 3.0-mm in diameter which is consistent with previous reports (Vagelli, 1999; Vagelli and Volpedo, 2004). Each egg had a strong fibrous attachment to a stringy matrix. These attachments keep the egg mass intact and the entire mass can be teased or rolled about under water without dislodging any eggs. It is not known whether these attachments persist until hatching, but it helps to explain how the male can keep the eggs in his mouth while simultaneously providing them with enough movement to insure they are properly aerated.



Figure 4. Photograph of egg mass removed from a male *Banggai cardinalfish*.

C. Larval Development

Larval developmental stages leading up to hatching have been thoroughly described by Vagelli (1999) and will not be presented here. In summary, development of the larvae takes place over the course of 19 to 20 days at which time hatching occurs. However, the larvae are not immediately released into the water column after hatching but remain in the male's mouth for an additional six to ten more days. If hatched larvae are expelled prematurely, they are not capable of swimming and generally exhibit slow growth if they survive. Premature juveniles have never been observed in the wild (Vagelli and Erdmann, 2002). Normal release of the juveniles occurs at night over the course of one to three days. One must remain vigilant during these crucial days, as after the last offspring is released, the male may attempt to eat the juveniles. The mating pair may spawn again in five days or up to several weeks and the whole process is repeated.

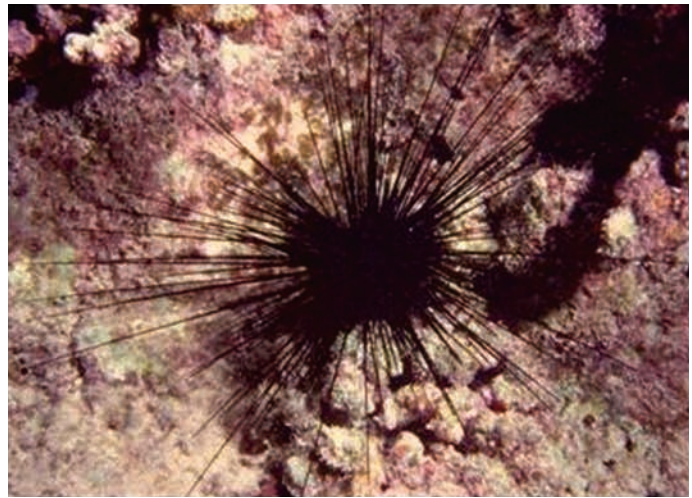
D. Release of Juveniles

Upon release from the male's mouth, the juveniles are approximately eight-mm in standard length (SL) with most of their yolk already utilized. The number of offspring per release has ranged from one to 50 individuals and the number of offspring is positively correlated with broodstock size. Just after release from the male the offspring will immediately seek shelter. In the wild, the preferred shelter appears to be among the spines of a sea urchin belonging to the genus *Diadema*. There are several reports of using a facsimile of a sea urchin in aquaria to attract the offspring and protect them from predation by larger fish (Marini, 1999). Artificial "urchins" can be made using 1-1/2-inch

or 2-inch PVC pipe caps and solid plastic wire. Fifty to seventy holes are drilled in the top of a cap and a six to eight-inch piece of plastic wire is inserted into each hole and secured with a dab of hot glue inside the cap. A comparison of an artificial sea urchin and one that occurs in Hawai'i is presented in Fig. 5. Alternatively, plastic weed-whacker line can replace the plastic wire. It has been found that the weed-whacker line is a little more flexible and the "spines" stick out better. When using this type of material, cut the line twice as long as the desired length of a spine. Thread the line down one hole, and back up another hole. In that way there is no need for glue to hold it in place. One disadvantage and this is only from an aesthetic point of view is that the weed-whacker line currently is not produced in a black color. At any rate, this does not seem to bother the juveniles with regard to using the artificial urchin as a shelter.



Artificial Sea Urchin



***Diadema* sp.**

Clyde Tamaru/John Hoover

Figure 5. Comparison of an artificial sea urchin and a long spined urchin found in Hawaiian waters.

E. Aborted Spawns

It has also been observed that the male may sometimes swallow or spit out the eggs or hatched offspring. There seems to be a relationship between failure to hold the offspring full-term and whether or not the male has had ample time to regain his strength and nutritional status during the time between release of juveniles and the next spawning event. If the male is well fed and/or is physically separated from the female for some additional time between the release of offspring and the next spawn, he is more likely to carry the next spawn to full term. This is especially important to consider when cooler water temperatures are encountered as experienced during the cooler months in Hawai'i. During this time period it is expected that offspring will develop more slowly and the incidence of males aborting a clutch of eggs increases unless appropriate conditioning of the male takes place.

V. BROODSTOCK NUTRITION AND FEEDING

A. Feeding Ecology

Gut contents of Banggai cardinals from the wild indicate that this species is a planktivore-carnivore with the size range of their prey items ranging from 0.1-mm (calanoid copepods) to 14-mm (megalop larvae and small fish) as reported by Vagelli and Erdmann (2002). It is clear from the observations made in captivity that they are fairly proficient hunters and juveniles and adults eagerly eat small fish, amphipods and other moderate-size crustaceans throughout the daylight hours. With some attention and persistence they can be weaned onto artificial or non-living prey in captivity. However, there are always concerns about reduced rates of reproduction when broodstock are fed non-living food and this area still needs to be investigated. In addition, when dealing with a large number of broodstock/mating pairs and the mass production of juveniles for market, it may be easier to provide the live prey than try to spend the additional time providing the individual attention necessary to wean each fish to prepared foods.

B. Feeder Guppies

In the prototype operation at Rain Garden Ornamentals, it was decided early on to produce live feed for adults and large juveniles. After exploring several options, work focused on mass production of the guppy (*Poecilia reticulata*) fry and small guppy juveniles. The guppy is probably one of the easiest fishes to breed in captivity and is also the number one freshwater ornamental fish species imported into the United States accounting for approximately 25% of the freshwater ornamentals fishes (Chapman et al., 1997). Guppies that are traded are also known as fancy guppies and are the result of generations of inbreeding, resulting in a myriad of color varieties and fin types. Another class of guppies is known as feeder guppies and the name “feeder” should already indicate the purpose of this type of guppy. These guppies resemble the wild-type in coloration and fin structure and most common/feeders are bred for one specific purpose which is to be fed to larger fish or other animals. A proximate analysis conducted on feeder guppies by the University of Hawai‘i Agricultural Diagnostic Service Center reveals that on a dry weight basis the ash, crude protein and crude fat are 16%, 62% and 14%, respectively. The Internet provides a host of Web sites showing reptiles (e.g., snakes, turtles), amphibians (Mexican salamander, frogs) freshwater fishes (e.g., Jack Dempsey, African cichlids) and a variety of marine ornamentals (e.g., seahorses, clownfish, snowflake eel, lionfish, and octopus) for which feeder guppies have been successfully used as a food source. While guppies are often thought to be a freshwater fish they can survive for two to four hours after direct transfer to full strength sea water. This gives the Banggai cardinalfish more than enough time to consume the guppies before they succumb to the high salinity. In addition, because most uneaten guppies eventually will die, they do not readily become established in the Banggai cardinalfish rearing system. It has been observed that at salinities less than 25 ‰ a few individual guppies may survive and become established. This has occurred in a few of the smaller

ponds at Rain Garden Ornamentals and has even progressed to the point where they spawn and develop reproducing populations.

C. Essential Fatty Acids

Reportedly one essential nutritional component necessary for captive Banggai cardinalfish are diets that have the appropriate essential fatty acids that have been reported to be important for other marine species (Marini, 1999; Vagelli, 2004). Two essential fatty acids in particular, eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA), have also been shown to be important in the growth, survival and stress resistance in a number of marine teleosts (Ako et al., 1994; Tamaru et al., 1993). Because of concern about the fatty acid profile of the freshwater guppy *versus* other marine food items, a short study was conducted to determine if there is any benefit in feeding guppies with a commercial diet high in marine lipids prior to feeding them to the Banggai cardinalfishes. Fatty acid analysis of three groups of guppies: 1) unfed controls, 2) guppies fed Silver Cup™ salmon starter for three days and guppies fed the salmon starter for seven days were conducted at the Department of Molecular Biosciences and Bioengineering. The results (Table 1) indicate that regardless of the feeding treatments, all guppies had similar total and essential fatty acid levels. This suggests that the feeding schemes did not boost fatty acid profiles of the treated guppies and also indicates that the species may possess the appropriate enzymes that allow it to synthesize its own essential fatty acids from precursors obtained in their diet as reported in other fish species (Tamaru et al., 1993). Overall, the fatty acid profile of the guppy appears to be appropriate with respect to its fatty acid profile for marine fish in general. This is not surprising as guppies are thought to have evolved in a brackish water environment.

Table 1. Analysis of fatty acids (mg/100 mg body weight) in guppies fed salmon starter for 3 days, seven days and for unfed controls.

FA	Unfed Control	Fed 3 Days	Fed 7 Days
14:0	0.34	0.52	0.54
16:0	2.03	1.65	2.32
16:1 n7	0.66	0.67	0.87
18:0	0.85	0.62	0.66
18:1 n9	1.41	1.12	1.94
18:2 n6	0.79	0.69	0.80
18:3 n3	0.43	0.50	0.30
18:4 n3	0.07	0.13	0.13
20:1 n9	0.07	0.36	0.58
20:4 n6	0.97	0.90	0.53
20:5 n3	0.28	0.40	0.42
22:6 n3	1.72	1.27	2.01
Total	10.09	9.22	12.05

D. Feeder Guppy Production

Year-round mass production of guppy fry is fairly easy in Hawai'i. In the prototype system, a 50-m² pond lined with a polyethylene sheet was used (Fig. 6). If soils are not conducive to pond construction, large liner tanks (a plywood hoop with plastic liner) would be equally effective. The guppy broodstock were a combination of wild-type guppies and a strain of yellow-snakeskin guppies which were known to be especially prolific. The pond had three air stones and was filled with freshwater from a spring but receives little to no water exchange. The pond received enough filtered sunlight to maintain a bloom of phytoplankton and a secchi disk visibility of about 50-cm. The guppy pond was fed daily with five to 10-g of a tropical fish flake food. Since this particular pond is under trees, there are additional nutrient inputs from leaf litter.

As needed, a small dip net was used to capture schools of guppy fry from along the edges of the pond. Because the pond is larger than needed, more sophisticated means of capturing and size grading fry (Tamaru et al., 2001) was not necessary. The contents of the dip net were placed in a small basket of plastic mesh with four-mm openings. The basket was partially submerged in a plastic tray with three to five-cm of water. The fry and small juveniles swam through the mesh and were poured into a bucket for later



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Figure 6. Polyethylene-lined production pond for mass culture of feeder guppies.

feeding to the Banggai cardinalfishes. The larger guppies were retained in the mesh basket and returned to the pond. The fry and small juveniles that were separated had an average weight of 10-mg and ranged between five to 12-mm SL.

In the prototype system, about 300 to 400 guppies were harvested daily but the production capacity of the pond is thought to be considerably higher. A rule of thumb for production output of livebearers is one fry per female per day (Tamaru et al., 2001). Therefore with 1,000 females stocked into the pond a production output of approximately 30,000 guppy fry can be produced within a month. The Banggai broodstock and large juveniles in grow-out were fed guppies once or twice daily. A large adult will eat about ten fry per day while large juveniles may eat two to three per day and it is estimated that the production pond could support approximately 1,000 Banggai fry per month with very little operating costs. Excess guppy fry were sold as feeder fish (\$0.02-0.03 per fish) to make up feed costs and the adult population may have to be thinned if overcrowding begins to adversely impact reproduction.

It should be mentioned that in the prototype system that houses the fishes, the Banggai adults eat some gammarid amphipods which they find naturally occurring in the tanks and cages due to the use of a reservoir pond. The diet of juveniles in grow-out is also supplemented with brine shrimp nauplii and naturally occurring rotifers, copepods and other mesoplankton.

VI. BROODSTOCK SYSTEM

A. Types of Holding Facilities

Two types of broodstock holding containers were evaluated and these are glass aquaria and cages. The aquaria were either 29-gal (110-L) that can accommodate one or two pairs of broodstock or 55-gal (208-L) that can hold two to three mating pairs. Pairs are separated by a center partition as seen in Fig. 7. Recently, we have started covering the partitions with black polyethylene shade cloth so that a male does not become preoccupied by another male on the other side of the partition. The cost for the glass aquaria is approximately between US\$0.35 to US\$0.40/L, excluding tank stand/supports.



Figure 7. Glass aquarium modified for holding Banggai cardinalfish mating pairs.

The aquaria are located outside under light shade. Each tank is equipped with an air stone and provided constant aeration but is otherwise bare with no filtration or substrate. Water is continuously exchanged between the aquaria and a small seawater reservoir pond. The low biomass and feeding rate is such that no filtration apparatus is required for the pond as long as circulation is maintained. Maintenance of the aquaria is limited to occasional scraping of the front glass and monthly siphoning of detritus from the tank bottom. If it is not practical to re-circulate water between the aquaria and a reservoir as in the prototype being described then traditional filtration systems (e.g., under-gravel filters and external biofilters) can be employed.



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Figure 8. Floating cages for holding broodstock.

Floating cages in a seawater pond was also evaluated for holding broodstock and for grow-out (Fig. 8). It should be mentioned that the cages could just as easily be floated in a large tank. The low biomass and feeding rate is such that no filtration apparatus is needed for the pond. The cages are made of a polypropylene mesh with 1.5-mm openings. Each cage is 60-cm x 40-cm x 40-cm deep with a volume of about 100-L. To construct the cage, the mesh is seamed with a hot glue gun. The mesh is attached to a 40-cm x 60-cm frame of 1-inch PVC pipe and elbows which holds the cage shape and provides flotation. Each cage has an air stone and houses one pair of broodstock. The cost of the cage fabrication materials was \$9 each or about \$0.09/L.

B. Cage Construction

To construct the cage, the 48-inch (about 120-cm) wide material was cut into 16-inch (about 40-cm) wide panels. An 82-inch (about 208-cm) long x 16-inch (about 40-cm) wide panel formed the four sides of the cage and a 24-inch (about 60-cm) long x 16-inch (about 40-cm) wide panel forms the bottom. A hot glue gun is used to seam the side panel to the bottom. A section of the side panel was overlapped on the bottom panel about three-eighths inch and hot glue was worked into the mesh with the tip of the gun. After the bottom panel was secured to the side panel, the side panel was trimmed to

(it was purposely cut too long initially) and the two ends of the side panel were seamed together to form a corner.

The advantage of an aquarium is the glass front which allows a side view of the broodstock. The side view makes it easy to determine when the male is holding eggs/offspring. A cage only provides a top view. However, with experience, males holding eggs/offspring can be differentiated by their reclusive attitude, and confirmed by their refusal to eat. The advantage of the cages is their low initial cost. Even when the costs of materials for a seawater pond or large liner tank are included, the cage system has a lower material cost. However, the cages and the pond or large tank to house them have fabrication costs while the aquaria may be set up relatively quickly. No differences in production or ease of maintenance have been found, so the choice of aquarium or cage will depend on circumstances specific to each operation.

VII. GROWOUT SYSTEM

A. Rain Garden's Prototypes

Reports in the literature describe the grow-out of Banggai cardinalfish offspring in intensively managed indoor aquaria (Vagelli, 1999, Marini, 1999). However, the techniques described are somewhat labor intensive and involve feeding enriched brine shrimp every few hours (Marini, 1999). Two less-intensive approaches which provide excellent results (e.g., high survival, good growth) have been field-tested and these are pond culture and cage culture and are described in the following sections.

B. Extensive Pond Culture

A group of offspring were released directly into a small seawater pond measuring about 130-square ft (12-m²) with a 3,500-gal (13-m³) capacity. Salinity varied seasonally from 25 to 35 ‰. This was a clearwater pond with well-established flora and fauna and there were no feed inputs or attempts to promote zooplankton production. All of the offspring survived and grew to market size in 125 days. While this rate of growth and survival cannot be surpassed, it is important to note that the fish density was only 1.3 fish/m³ which is a prime example of how effective an extensive culture system can be to produce this species. The advantages of the extensive culture systems, high survival and growth along with the minimal input into the rearing process, however, is overshadowed by its low productivity. This approach was not pursued long enough to determine pond carrying capacity or the effect of zooplankton management because in addition to the low output, it became clear that harvesting fish for market would be difficult.

In a second trial, a group of 40 small juveniles were released into the same 13-m³ seawater pond and left to grow to adult size for use as future broodstock. Within five months, the fish had started reproducing in the pond. As needed, broodstock were captured and transferred to aquaria. Market-size fish (offspring of the fish which were

originally stocked) were occasionally removed and sold. After twelve months, all fish were removed in order to use the pond for other tasks. Of the 40 juveniles stocked, 38 matured and were removed as adults. A total of 66 juveniles were also removed. This is not a high rate of reproduction and many offspring were probably consumed by the adults. The maximum number of fish which were in the pond at any one time is not known. However, this trial indicates that production in open ponds may be on the order of four to eight fish per m³ without supplemental feed and relying totally on natural forage.

C. Cage Culture

From experience, the cage production system seems to be a more manageable and viable approach to commercial grow-out of this species (Fig. 8) and cages that are 35-L and 100-L in volume have been used with equal success. Groups of up to 15 newly-released offspring are stocked into the 35-L cages and up to 50 offspring can be stocked into the 100-L cages. Offspring from separate broodstock pairs can be mixed if the age difference is only a few days. It would not be advisable to mix offspring of vastly different sizes as the smaller fish will find it difficult to compete for food. The 100-L cages (60-cm x 40-cm x 40-cm deep) are the same as those used for holding the broodstock as discussed previously. The dimensions of the 35-L cages are 20-cm x 30-cm x 40-cm deep and they are constructed from the same materials as the larger cages. All of the cages are floated in a small pond or large tank with each cage being equipped with an air stone and continuous aeration. For a pond, it is convenient to build a wooden walkway and have several cages suspended on both sides of the walkway (Fig. 8) taking full advantage of the available space available. A PVC pipe air manifold is attached to the side of the walkway where an airline can be dropped into each of the cages that are lined up alongside the walkway. In a large tank or steep-sided pond with easy access from the side, the cages can be positioned around the perimeter.

D. Pond Preparation

To provide for the necessary food organisms the pond or large tank should be managed for zooplankton production. This involves controlling zooplankton predators (except for the Banggai cardinalfish) and by providing nutrients into the pond to stimulate phytoplankton growth that in turn supports the zooplankton populations. The phytoplankton bloom also helps in controlling excessive macrophyte production. To accomplish this, the pond is first filled with raw seawater that is passed through a 100-micron mesh filter bag. This will allow an inoculum of various types of zooplankton and phytoplankton while excluding some of the larger forms. The pond is then fertilized to stimulate a phytoplankton/zooplankton bloom. Inorganic (chemical) fertilizers such as soluble Miracle-Gro™ develop phytoplankton blooms faster, but organic materials such as dry fish feed formulation or cereal meal can stimulate zooplankton blooms faster. As a rule of thumb, add enough inorganic fertilizer to provide about one-mg/L of total nitrogen

and phosphate. Repeat the application weekly until the secchi disk visibility approaches 60-cm. It is easy to over-shoot the mark with chemical fertilizers and the green water may become too dense for comfort or macrophytes may proliferate at the expense of phytoplankton.

Organic fertilizers stimulate phytoplankton blooms more slowly because the material must first be broken down by microbes and the nutrients mineralized before they are available for phytoplankton growth. However, as the organic material is breaking down, it is available to stimulate the growth of many types of zooplankton. There is an endless array of organic materials which can be used including various types of agricultural wastes. We have found that because it is but a minor cost it is safest to use pulverized fish food. A daily application of about 0.5 to 1.0-g/m² of water surface of a commercial fish food diet that is 25-35% in crude protein provides the desired results. Feed additions should be stopped as the secchi disk visibility approaches 60-cm, and resumed if the water begins to clear. A small amount of macrophyte is not detrimental, but floating mats which exclude light, need to be removed.

Despite attempts to exclude pest and predators, they will eventually gain entry and proliferate if raw seawater is used. Particularly annoying are glass anemones, hydroids which bud off medusa, and also some types of tunicates. When this occurs, the tank or pond must be completely drained, dried or disinfected with 200-mg/L of chlorine bleach, and the process started over again. Even in the absence of pests or predators, the zooplankton population may occasionally crash. Thus, it is a very good practice to have two tanks or ponds operating simultaneously so that water and zooplankton may be moved from one to the other or while one may be completely drained for rejuvenation.

E. Grow-out and Feeding

Newly released offspring are able to eat relatively large prey like rotifers and brine shrimp nauplii (Manini, 1999; Vagelli, 1999). Once released from the male, the offspring actively seek their prey and are proficient hunters. In six weeks they grow to about 15-mm SL. In four to five months they will be ready for market. Although already widely understood, it should be re-emphasized that growth rate is largely influenced by water temperature and it should be remembered that Hawai'i has a subtropical climate. There is a distinct winter season where grow-out may take several weeks longer than during the summer months. A summary of the temporal changes in daylength, average monthly water temperatures observed at the Windward Community College Aquaculture Complex and rainfall measured at the Honolulu International Airport over the course of a year is provided in Fig. 9. Clearly these changes in environmental factors present challenges that need to be taken into consideration as it is imperative that both the right kinds and adequate numbers of feed items be provided to assure maximum survival and growth of the Banggai cardinalfishes.

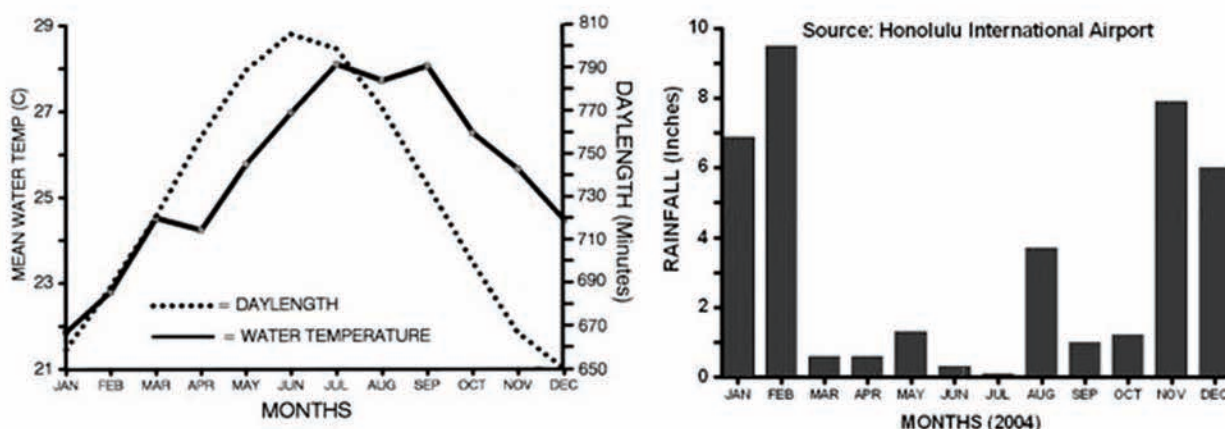


Figure 9. Temporal changes in water temperature, daylength and rainfall.

It should be noted that the mesh openings in the grow-out cages are 1.5-mm. From experience we have found that even when the mesh is partially fouled by algae and detritus, there is sufficient free flow of water into and out of the cage caused by the water movement generated by the aeration from the air stone in the cage. Zooplankton (Fig. 10) also flow into the cage and will be quickly consumed by the young Banggai juveniles. With good pond management and barring any catastrophes such as stormy weather there will always be sufficient zooplankton to support good nutrition and rapid growth. In many cases, no other sources of feed are necessary until the Banggai individuals are “large” juveniles. From that point on and until they reach market size they are provided a diet of live feeder guppy fry.



Figure 10. Photomicrographs of *Artemia nauplii* (top left corner) and other wild zooplankton.

F. Supplementing with *Artemia*

To insure there is sufficient bulk feed, the wild zooplankton should be supplemented with newly-hatched brine shrimp. Again from experience we have found that about 0.25-g of brine shrimp cysts must be hatched for each cage each day. The estimated cost is about US\$2.50 per cage over a 130-day grow-out period. But, just as the zooplankton can flow into the cage, live brine shrimp can flow out so it should be added very slowly to insure that as much as possible is eaten before it is lost. This is accomplished using a drip feeder that can easily be constructed out of simple supplies (Fig. 11).

Items that are needed for constructing a drip feeder are:

- 30-gal trash can or 55-gal drum
- small power-head pump
- electrical source
- 100- μ m mesh Nytex™ screening
- silicone sealant
- associated plumbing supplies such as PVC pipe and fittings
- one-eighth inch black polyethylene tubing sold for micro-drip irrigation systems

The system works on the basis of the power head pump supplying pond/tank water to the main feed tank. A drip tube continuously siphons water from the feed tank to each cage supplying the *Artemia* in the process. The flow rate is typically 4 to 5-L/hr depending upon the length of the tubing leading to the cages. Excess flow into the main feed tank is drained out of the feed tank through an outlet port which is screened with 100-micron mesh. Newly hatched brine shrimp are poured into the feed tank and are slowly siphoned to the cages. No enriching of the brine shrimp is needed as the essential fatty acids are provided by natural zooplankton generated by the pond. The use of brine shrimp in this case is more as a supplement rather than the vector for these essential nutrients.

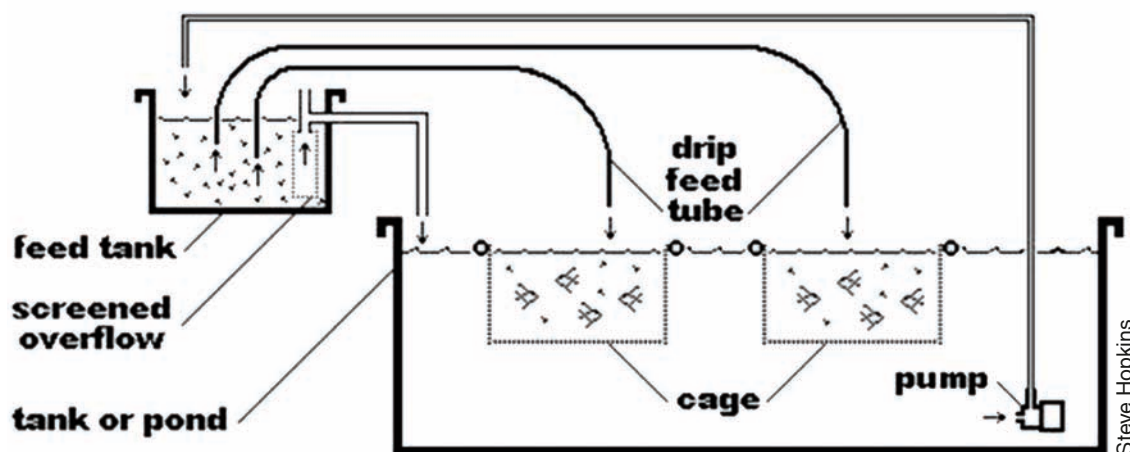


Figure 11. Schematic of single-stage drip feeder for brine shrimp delivery.

With ten cages being fed from a 55-gal drum, the rate at which brine shrimp are delivered to each cage will drop from 3,000 individuals/minute to one individual/minute over the course of about 16 hours using the single-stage feeder. The delivery rate will be much more uniform if a two-stage feeder is used (Fig. 12). With the two-stage feeder, a second feeder tank needs to be used. The brine shrimp are poured into the first stage tank and are transferred to the second stage tank with a large-bore siphon before being drained to the cage through the drip tubing. With a two-stage system, the brine shrimp are delivered at a more uniform rate with a maximum of about 100 individuals/minute/cage. After sixteen hours the delivery rate is still over five individuals/minute/cage and does not drop below one individual/minute/cage over the course of 21 hours.

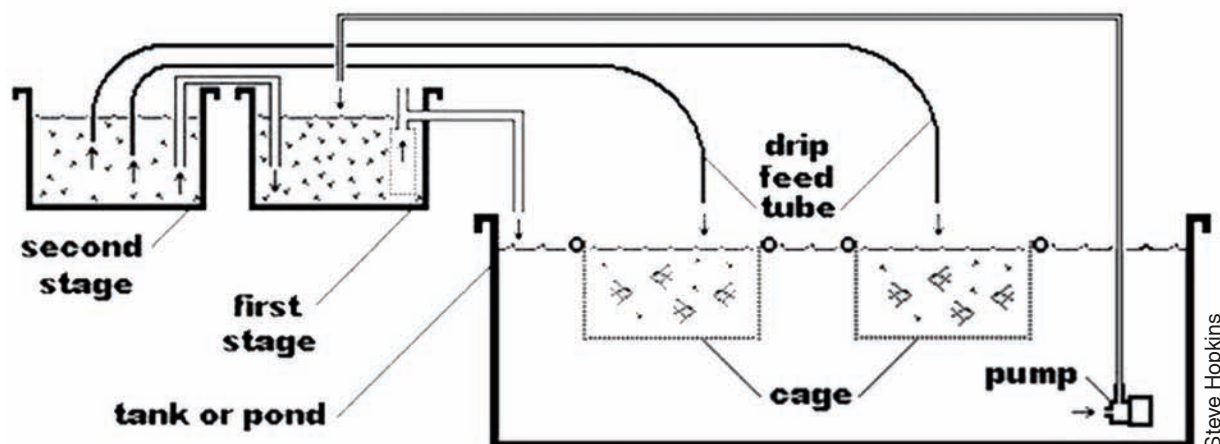


Figure 12. Schematic of a two-stage brine shrimp feeder.

In intensive indoor aquarium culture, the enrichment of brine shrimp and other prey is essential as they lack the essential fatty acids required by the Banggai cardinalfish juveniles (Manini, 1999; Vagelli, 2004). In the cage culture system, Banggai cardinalfish offspring are free to prey on naturally occurring zooplankton and therefore enrichment of brine shrimp is not necessary. A summary of only the essential and total fatty acids in newly hatched *Artemia* and copepods both cultured and from the wild are provided in Table 2. While *Artemia* nauplii have a fairly good total fatty acid content they are either low or do not possess 20:5 n3 and 22:6 n3. In contrast copepods either from the wild or under culture possess significantly higher levels of these two essential fatty acids.

Table 2. Comparison of essential fatty acids from brine shrimp and copepods. Values are expressed in terms of mg/100 mg dry weight.

Essential Fatty Acid	Newly Hatched <i>Artemia</i> Nauplii	Wild Copepods Kaneohe Bay	<i>Euterpina Acutifrons</i> Fed <i>Chaetoceros</i> sp.	Wild Copepods Fed <i>Isochrysis</i> sp.
18:2 n6	0.49	0.08	0.51	0.89
18:3 n3	1.94	0.07	1.11	1.24
20:4 n6	0.17	0.07	0.39	0.21
20:5 n3	0.36	0.49	1.64	1.24
22:6 n3	nd	2.08	1.90	2.73
Total	6.55	4.68	13.3	15.1

nd = not detectable

With the use of the feeder system described the brine shrimp that are provided, to some extent, will enrich themselves on the phytoplankton rich water that is placed in the drip feeder tank(s). This is possible because the enrichment process for brine shrimp and rotifers reaches its full capacity within the first hour of exposure and there is ample time for their nutritional value to be boosted while they are in the feeder tanks waiting to be siphoned into the cages. It is proposed that the mixture of brine shrimp and assorted other zooplankton that the prototype system provides presents a more varied and complete diet. Therefore, it is not surprising that offspring survival and growth in the outdoor cage system is as high as it has been.

One of the main advantages of the cage system is that it requires very little maintenance and labor. Cages are normally only cleaned between crops or roughly 130 days. Likewise, the brine shrimp-feeder tanks and drip tubes are cleaned every two to three weeks. A pond may need to be drained, disinfected and refilled every three to four months. It appears that this production system may only work in Hawai'i and other tropical or nearly-tropical environments where rearing temperatures are fairly stable. Sunlight drives the system as it produces the phytoplankton upon which the zooplankton grazes. Being outdoors, the seawater tanks or ponds are vulnerable to dilution by heavy rainfall in Hawai'i, especially during the months from November through March. Banggai cardinalfishes seem to be somewhat euryhaline although the lower limits of salinity tolerance have not been determined. However, there does not seem to be any adverse impact on reproduction, growth or survival when salinities decline from 35 ‰ (full strength sea water) to 25 ‰.

VIII. PRODUCTION, MARKETING AND ECONOMICS

A. Production Outputs

Using the cage grow-out system, survival has averaged 88% from time of release of juveniles from the male to market size which varies between 125 and 130 days. In some cases, all of the offspring survive to market-size. Prior to this work, the best source of production data on the Banggai cardinalfish was from Marini (1998), who raised Banggai offspring in indoor aquaria on enriched brine shrimp nauplii and weaned them to adult brine shrimp and enriched ghost shrimp (*Palaemonetes sp.*). Marini reported survival rates averaging 66% at 100 days. Survival from intensive larval rearing experiments under laboratory conditions that assessed the impacts of using enriched versus non-enriched *Artemia* reported average survival of 95% over a 116-day grow-out period (Vagelli, 2004). Clearly the prototype system developed at Rain Garden Ornamentals is consistent with laboratory scale outputs and is felt to be competitive in commercially producing this species. Its greatest asset is that it minimizes the day-to-day labor costs and incorporates the advantages of the extensive culture system (e.g., natural productivity to produce food).

B. Time to Market

The best time to market hatchery-produced fish is just before they begin to pair off and become aggressive. Once a few males in the group become aggressive, they cannot be maintained in communal tanks, either at the grow-out facility, the wholesaler facility, or the retail outlet. At this stage fish must be individually bagged for shipping and most pet stores are not set up to effectively house each fish separately. Thus it is in everyone's best interest to market individuals prior to attaining that state of maturity. The only fish that should be kept at the farm past this point are those to be developed as broodstock and they should be individually isolated until the gender can be determined.

C. Prices and Outlook

When Banggai cardinalfish were first introduced to the aquarium trade in 1995, retail prices were about US\$100/fish. Breeder reports indicate that the capture and handling of wild fish was better early on during their exploitation and there was a fair chance that a wild fish would live long enough to breed in captivity. Prices have steadily declined since that time, despite concerns about over-exploitation of wild stocks. Likewise, the survival of wild-caught fish in the distribution chain and in the home aquarium has declined dramatically. Currently, the retail price for Banggai cardinalfish is approximately US\$15 each for wild-caught fish and US\$25 each for captive-bred fish. From our experiences the wholesale price for wild fish may be as low as US\$5 each. Rain Garden Ornaments has been consistently receiving a farm gate price of US\$7 each. If marine aquarium hobbyists were aware of the vast differences in health and vigor between captive-bred fish and the wild fish currently in the market, the price differential would be even greater. What is clearly needed is a continued source of information for the general public about the potential harm that collections from the wild can do - especially for this species because of its very limited geographical range. Artificially propagated marine ornamentals provide an alternative means in obtaining products for the industry with a far smaller environmental footprint.

While prices have fallen dramatically since its introduction, the long-term outlook for artificially propagated Banggai cardinalfishes looks promising. The current rate of wild stock exploitation is not sustainable (Allen, 2000) and hobbyists will eventually learn that captive-bred Banggai make much better aquarium fish. Using the system described, raising Banggai can be profitable at the US\$7 farm gate price and revenues can be much higher with creative marketing approaches.

D. Material Setup Costs

A summary of the production economics for the prototype is presented in Table 3. It is estimated that the materials to construct a system capable of generating US\$10,000 per year in gross revenues at US\$7 per fish would cost approximately US\$4,500. This

is with a life expectancy of about five years (US\$900/year) plus the cost of construction labor which is estimated to be at least 100 hours (20 hours/year). This also assumes that broodstock are maintained in aquaria or liner tanks and ponds are to be used for guppy production and cage grow-out of Banggai juveniles. The facility would have a footprint of at least 175-m². Direct costs for feed, electricity and miscellaneous supplies would be about US\$900 per year. Production labor, exclusive of construction and marketing, would require about 275 hours per year. Gross revenues less production costs and amortized construction materials would be about US\$8,200. With 275 hours of annual production labor and 20 hours of amortized construction labor, the gross return on labor would be US\$27.80 per hour invested. This is a very simplified analysis and does not include the costs of land used, investment costs, lost opportunity costs, and so forth. Profit margins could increase or decrease as the scale of the operation is increased or decreased. However, it does demonstrate that the commercial production of the Banggai cardinalfish can be easily incorporated as either a small-scale backyard enterprise or part of a company with a diversified number of products as Rain Garden Ornaments.

The work summarized in this manual also has an added benefit in that in the area around the natural range of the Banggai Cardinalfish there is a clear potential to augment or replace collections from the wild with aquaculture. The technology described herein is, by design, not very sophisticated and can easily be adapted for use in that region. Furthermore, we have demonstrated that the process can be simplified even further by employing open-pond production without the use of cages, feeders, and such. This open-pond approach may be more viable in areas where investment capital is limited and labor costs are lower. Clearly, artificial propagation of Banggai cardinalfish provides an economic alternative to fishermen engaged in collection of this species from the wild.

Table 3. Summary of production costs of Banggai cardinal culture prototype.

Category	Amount	US\$
Sale Price		\$7/Fish
Gross Revenue		\$10,000/Year
Facility Materials amortized over 5 years		\$900/Year
Facility Footprint		175 m ²
Construction labor		\$20 hr/Year
Production labor		275 hr/Year
Direct costs: feed, electric, supply, etc.		\$900/Year
Return on labor and other direct costs		\$27.80/Hour

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