

Original Research Article

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## Selective and Interbreeding of Freshwater Ornamental Shrimps with Endemic Aquarium Plants

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### Abstract

Shrimp are decapods or 10 limbed crustaceans along with crayfish, chera crabs and others. Historically only crayfish were kept, in the last 10-15 years shrimp and other crustaceans have become increasingly popular in home aquaria. Three endemic species Indian Dwarf Shrimp, "Rainbow" Shrimp *Neocardina zeylanica*, Indian White banded Shrimp *Caridina cf. babaulti* var. Indian white banded, Stripe Shrimp *Caridina cf. babaulti* and an exotic species Red Cherry Shrimp *Neocaridina denticulata sinensis* "red" were bred commercially by developing hatchery technology. Availability of these endemic species in the ornamental market will reduce the stress on wild population as they are wild caught for their commercial demand. The life cycle of the selected shrimp species were studied in detail and recorded. The Planktonic larval stages are completely suppressed and the larvae assume a benthic lifestyle after hatching, i.e., they are miniature versions of the adults. In the process of selective breeding, commercially sold feeds haven given better colour enhancement than natural feeds. Interbreeding among these shrimp species resulted in hybrids resembling their wild variants.

### Article Info

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### Introduction

The practice of shrimp keeping has developed among hobbyist to control algal formation in planted aquariums (Heerbrandt, T.C. and J. Lin, 2006). The main aim of the work is to focus on endemic freshwater shrimps that have ornamental value. Their biology, behavior, adaptability in aquarium conditions and color enhancement by feed formulations and selective breeding are being studied. Currently the industry is completely depends on the wild caught species (Raghavan *et al*, 2014). Breeding and production of high grade 'Rainbow shrimps', 'Stripe shrimps', 'White banded shrimps' will definitely reduce the stress on the wild population. Rainbow shrimps have potential market for their 'algae eating' ability despite its subdued coloration. By improving the pigmentation, its demand

can be increased both in domestic and international markets. For pigment enhancement, blanched carrot, cucumber, dried grapes and mixture of commercial shrimp foods were administrated and results were encouraging. The objectives of the present study are given below.

- Maintenance of fresh water ornamental shrimps brood stocks with endemic aquarium plants.
- Standardization of hatchery technology of fresh water ornamental shrimps.
- Selective breeding of fresh water ornamental shrimps for the production healthy strains with desirable character.
- Interbreeding of fresh water ornamental shrimps for the production of novel attractive strains for trade.

## Materials and methods

### Maintenance of fresh water ornamental shrimps brood stocks with endemic aquarium plants

Brood stocks of each species (three endemic species 'Rainbow shrimps', 'Stripe shrimps', 'White banded shrimps' and one exotic species-'Red cherry shrimps') were maintained in separate glass tanks (40 × 20 × 25 cm) supported with bio-chemical filter systems. Plants growth is aided with Compact Florescent lamp at a ratio of 1 watt per 5 liters, Mopani driftwood and suitable soil substrates. About 10 females and 10 males used were with average total length of 2.5±0.5 cm and placed into two separated tanks. Only females with saddle and healthy males were used. All the shrimps were procured from the koluthur, Chennai (Hub for aquarium trade). Each individual shrimps were carefully inspected for disease and abnormalities. The Shrimps were acclimatized for 15 minutes and then released into their respective tanks.

### CO<sub>2</sub> system

To aid plants growth in the brood stock tanks, CO<sub>2</sub> were supplied. Initially to reduce the running cost of the operation, improvised method was used where a mixture of water, sugar and yeast were used to produce CO<sub>2</sub>.

The main disadvantage of the improvised method is that the CO<sub>2</sub> supply cannot be fully regulated. The level of CO<sub>2</sub> was estimated by 'Drop Checker' with 4dkh Brmothymol blue solution. The desired CO<sub>2</sub> level is 30 ppm.

### Lighting

After the initial setup, a photoperiod of 6 hrs is maintained. Then after couple of weeks, 8-9 hrs of photoperiod is maintained. Frequent change in photoperiod resulted in the formation of algal growth and throwing the plants off their routine and to compensate this automatic timer was used later. For lighting, Compact Florescent lamp at a ratio of 1 watt per 5 liters was used.

### Fertilizers

Not enough shrimps in the tank to provide fertilizers to the plants in this case, fertilizers tabs are inserted near the roots.

## Water parameters

The selected species prefer soft, acidic water with temperatures of about 26°C. For each shrimp species the water parameters are different. High temperatures will reduce the lifespan of the shrimps, lower temperatures increases the amount of dissolved oxygen, and higher temperature reduces the amount of dissolved oxygen. Although keeping shrimps in lower temperature helps prolong their life, it'll decrease their appetite and breeding rate. The 3 most important parameters are PH, GH, and KH. These 3 are the main deciding factors in establishing optimal parameters for the shrimps. Maintaining stable parameters is one of the most important aspects in keeping shrimp. Shrimp need clean water, so 30 percent of water was changed at least once every two weeks. Aeration was provided throughout the study period. Shrimp are especially sensitive to chlorine or chloramines in city water supplies, so care was taken to use a dechlorinator or ammonia neutralizer to remove these chemicals. Quality products that neutralize ammonia are important. If chlorine and/or ammonia aren't eliminated, it would result in mortality. Invertebrates especially shrimp are very sensitive to copper. So care was taken not to treat an aquarium with any copper-containing products. Copper is not the only thing that can harm shrimp. Many of the anti-parasite products on the market will have devastating results on shrimp, so this is just another good reason to have a shrimp-only aquarium. The kits or apparatus used were; DO meter (dissolved oxygen and temperature) and API aquarium freshwater test kit (pH, ammonia, and nitrite). Measurements were taken twice a week.

### Shrimp feed

For pigment enhancement, blanched carrot, cucumber, dried grapes and mixture of commercial shrimp foods were administrated and results were encouraging. Some of the commercialized colours enhancing foods are Hakari Crab bites, Spirulina, Tetra Colour bites and Hikirai Agar wafers.

### Endemic plants

Aquatic plants are not only an ideal background to showcase the aquatic habitat, but they're also improving water quality by allowing biological filtration, removing nitrates, and oxygenating the water. Plants maintained in the brood stock tanks are as follows: *Cryptocoryne cognata*, *Rotala macarandras*, *Rotala indicas*, *Hygro difformis*.

## Standardization of hatchery technology of fresh water ornamental shrimps

Important details of shrimp species experimented in the present study are given below (Fig. 1).



Fig. 1: Shrimp species used in the study.

**Common Name:** Red Cherry Shrimp

**Scientific Name:** *Neocaridina denticulata sinensis* "red"

Holthuis (1993) stated that *N.d. sinensis* was placed under the genus *Caridina* before being reclassified under *Neocaridina*.

**Size:** up to 3 cm

**Origin:** Taiwan (Hung *et al.*, 1993) this red variety does not occur naturally anywhere, but originally bred in Taiwan. Wild form occurs in Southern China and Taiwan.

**Larval Development Type:** Completely Suppressed (Shokita, 1981; Shy, 1994): Larvae assume a benthic lifestyle after hatching, i.e. they are miniature versions of the adults (Yang, H.J. and H.S. Ko, 2003). There is no planktonic larval stage.

**Common Name:** Indian Dwarf Shrimp, "Rainbow" Shrimp

**Scientific Name:** *Neocardina zeylanica*

**Size:** 2- 2.5 cm

**Origin:** India (North East), might be more widely spread in south Asia.

**Larval Development Type:** Completely Suppressed (Shokita, 1981; Shy, 1994): Larvae assume a benthic lifestyle after hatching (Yang, H.J. and H.S. Ko, 2003), i.e. they are miniature versions of the adults. There is no planktonic larval stage. The hatchlings of this shrimp are extremely small and grow slowly.

**Common Name:** Indian White banded Shrimp

**Scientific Name:** *Caridina cf. babaulti* var. Indian white banded

**Size:** 2.5-3cm

**Origin:** India (North East), might be more widely spread in south Asia.

**Larval Development Type:** Completely Suppressed (Lai, H.T. and J.Y. Shy, 2009): Larvae assume a benthic lifestyle after hatching, i.e. they are miniature versions of the adults (Yang, H.J. and H.S. Ko, 2003). There is no planktonic larval stage. The hatchlings of this shrimp are extremely small and grow slowly.

**Common Name:** Stripe Shrimp

**Scientific Name:** *Caridina cf. babaulti*

**Size:** 2.5-3cm

**Origin:** India

**Larval Development Type:** Completely Suppressed (Lai, H.T. and J.Y. Shy, 2009): Larvae assume a benthic lifestyle after hatching (Yang, H.J. and H.S. Ko, 2003), i.e. they are miniature versions of the adults. There is no planktonic larval stage. The hatchlings of this shrimp are small and grow slowly.

## Materials used

Glass tanks (40 × 20 × 25 cm) supported with biochemical filters are used. External filters and power head filters are not used to avoid the suction of larvae. Hatchery tanks are maintained without soil substrate but mosses are maintained on bog wood and quart stone. The biofilm produced by the moss was used as nourishment for the larvae.

## Results and discussion

### Stripe shrimp *Caridina cf. babaulti*

The selected species reached first maturity at around 75 days old with TL 2.3±0.2 cm (Demas, 2007). Upon reaching maturity, the presence of visible ovaries can be seen through the cephalothorax region of the females. Appendix masculina on the second pleopod were observed in males (Kubo, 1938), Similar observation was reported by Englund and Cai (1999). Ten pairs of matured male and female shrimps were placed in breeding tank. Mating process usually take place 1-3 day(s) after the introduction of male and female into the same tank. Female molted prior to mating. Observation was carried until female shrimp become ovigerous.

Both molting and mating occurred rapidly in less than 10 sec. During mating process male and female will be facing each other and sperm were deposited into the genital opening of female using its appendix masculina. The eggs were fertilized when it passed through the sperm on the way to the brood pouch. The pouch was formed by pleopods and overhanging of pleura of the female. A day after mating, female was observed to carry fertilized eggs, fan and incubated in the brood pouch until hatch. According to Adiyodi and Adiyodi (1994) this kind of parental care will result in higher survival rate of larva produced. Similar to those reported by Schram (1986). Fertilized eggs were oval in shape, vary from greenish to yellowish in color. The egg size was comparatively large, with an average length 1.19 mm. Thin ribbon-like filament binds the eggs in grape-like bunches and attached it to the female's pleopod. The membrane can hardly be distinguished since it is thin, transparent and lying close to the surface. Smaller females produced fewer eggs compared to larger females (Ketse, 2006).

Number of larvae produced by each female was counted and recorded (Fig. 2). After a week, only healthy larvae were used for the next experiment.

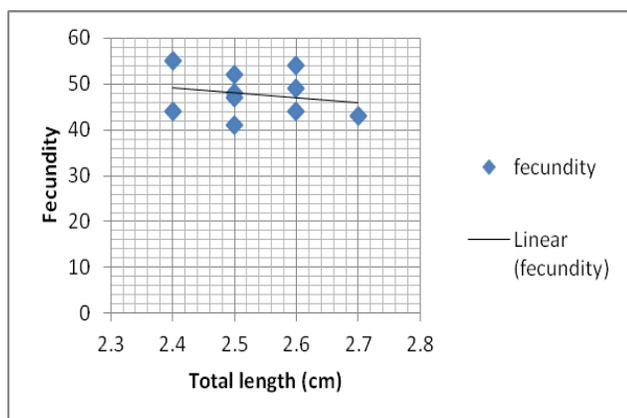


Fig. 2: Fecundity by length for *Caridina cf. babaulti*.

### Selective breeding of fresh water ornamental shrimps for the production healthy strains with desirable character

#### Selective breeding of Indian White Banded Shrimp *Caridina cf. babaulti* var. Indian white banded

A dozen of mature Indian White banded Shrimps were procured from the Kolathur Market, Chennai, South Asia largest hub for ornamental fish industry. The sex ratio was 1:5 (Male: female) If the number of males is not enough, it will decrease the rate of successful mating. On

the other hand, to Prevent female death from being exhausted because of being chased by multiple males. Some more plants were grown to provide hidden places.

Traits selected for selective breeding are:

- Thickness of the body colour
- Shape of the tail
- Overall size of the shrimp
- Pattern of bands and spots
- Thickness of the white bands
- Redness of the legs

After successful mating, berried females were allowed to stay in the same tank for 3-4 days and then move them carefully and slowly to the tank rich of moss and micro-organisms. Using the genetic algorithm for selective breeding:

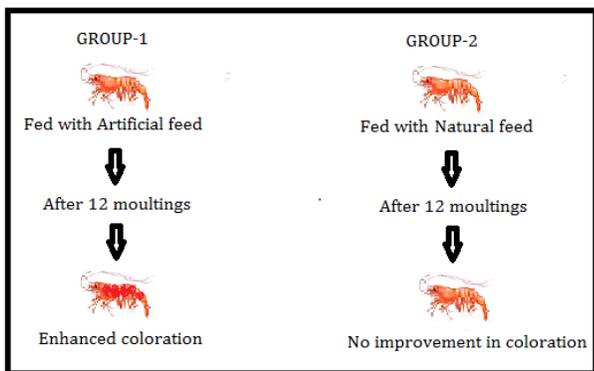
**Method 1:** Keeping the most desirable traits from the parents and passing it onto the offspring, not losing the traits in the offspring.

**Method 2:** All genes are randomly selected between two of the most elite male and female generations and part of the genetic info is exchanged. With the accumulation of the desired genetic information, we hope to achieve superior offspring.

Using these methods, F4 generations were able to retain and come up with desirable colour patterns.

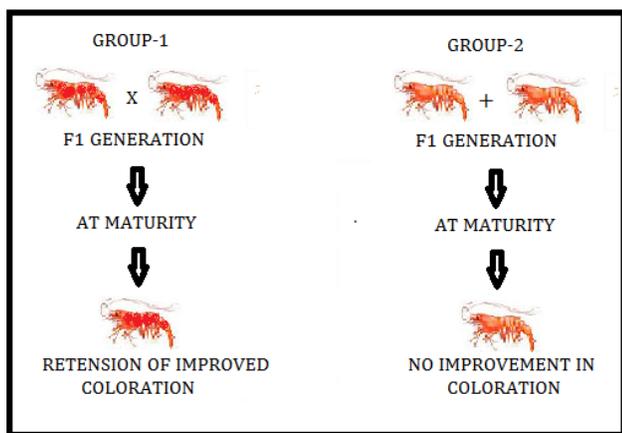
### Selective breeding and colour enhancement by feed formulations

Fifty larvae (one week old) were placed in a tank. Observation was carried out until all the larvae reached first maturity. Then two dozens of brightly coloured red cherry shrimps were separately maintained in two glass tanks. Both the groups of shrimps were given same amount of photoperiod, CO<sub>2</sub> injection, and plant composition. One of the groups (1) was exposed to different kinds of commercialized colour enhancing feeds such as Hakari Crab bites, Spirulina, Tetra Colour bites and Hikirai Agar wafers and the other group (2) had been administrated with natural food such as blanched carrot, cucumber and algae. After 12 moultings, the observations were made and recorded. The shrimps that were administrated with commercialized food showed improved red colouration compared with the group of shrimp fed with natural food (Fig. 3).



**Fig. 3:** Red Cherry Shrimps fed with artificial and natural feed.

The same procedure had been followed for the selected endemic species and the results were same. The F1 generation larva were segregated from the two groups of shrimps and separately maintained in two different tanks and they were allowed breed.

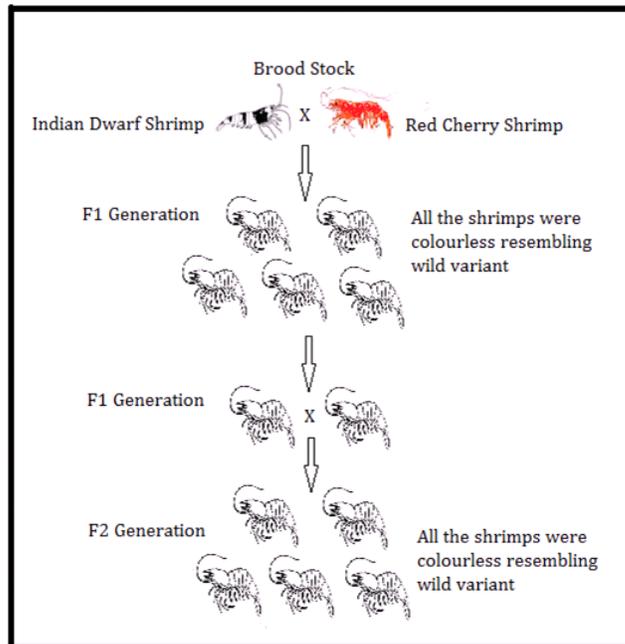


**Fig. 4:** F1 crossing in Red Cherry Shrimps.

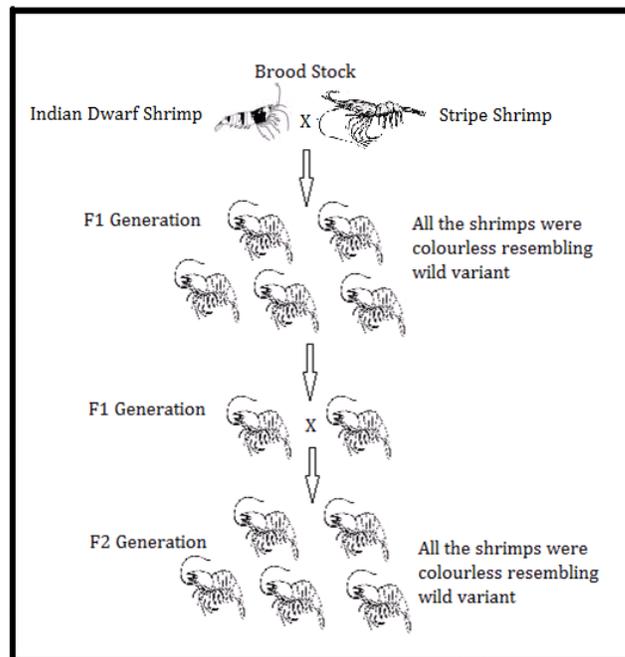
The F2 generation of Group-1 shrimps was able to retain the improved coloration from F1 parents in spite of feeding the F2 generation shrimp with natural feed. Where as in Group-2, shrimps of F2 generation did not show any major changes in their body coloration (Fig. 4).

**Interbreeding of fresh water ornamental shrimps for the production of novel attractive strains for trade**

The wild variant of the Red Cherry Shrimp *Neocaridina denticulata sinensis* is actually colorless and its Red color is due to intensive and repeated selective breeding and hence the name Red Cherry Shrimp. Interbreeding among these species resulted in colorless hybrids which represent their true wild variants. Same results were witnessed when F1 generation were interbred (Figs. 5 and 6).



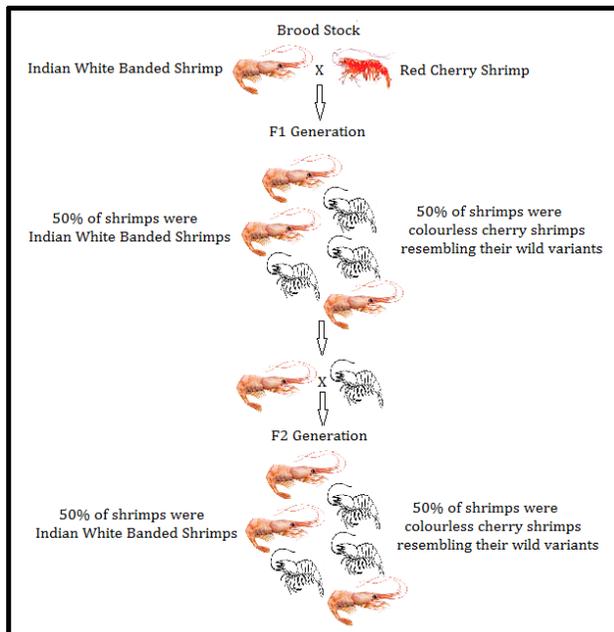
**Fig. 5:** Interbreeding between Indian Dwarf Shrimp and Red Cherry Shrimp.



**Fig. 6:** Interbreeding between Indian Dwarf Shrimp and Stripe Shrimp.

The wild variant of Indian White Banded Shrimp is light red in colour. So interbreeding of these species with Red Cherry Shrimps and Indian dwarf shrimps results in hybrids that 50% of the population is red and rest are colorless. Same results were witnessed when F1 generation were interbred Fig.7.

The life cycle of the four selected species were studied in detailed and recorded. In all the four species planktonic larval stages are completely suppressed and the larvae assume a benthic lifestyle after hatching, i.e. they are miniature versions of the adults. The study was successful in developing the hatchery technology and commercialising it. The life cycle of the four species were almost same and water parameters especially water temperature played a crucial part in breeding these species. The Fecundity of the shrimp species is proportionate to its total length.

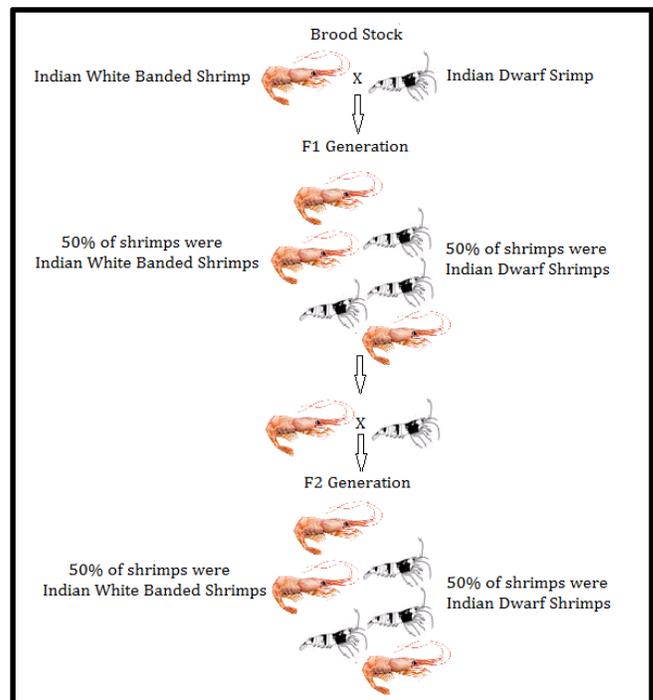


**Fig. 7:** Interbreeding between Indian White Banded Shrimp and Red Cherry Shrimp.

In colour enhancement procedures, commercially sold feeds have given better results than the natural feeds. Test samples that are fed with commercial feeds were able to retain enhanced colouration up to F2 generation.

Interbreeding among these shrimp species resulted in hybrids resembling their wild variants. Apart from Indian White banded Shrimp *Caridina cf. babaulti* var. Indian white banded which is red in colour by origin; other selected species are colourless by origin or by wild variant.

The wild variant of the Red cherry shrimp *Neocaridina denticulata sinensis* is actually colorless and its red color is due to intensive and repeated selective breeding and hence the name Red Cherry Shrimp. Interbreeding with Red Cherry Shrimps resulted in colorless hybrids which represent their true wild variants (Fig 8).



**Fig. 8:** Interbreeding between Indian Dwarf Shrimp and Indian White Banded Shrimp.

### Conflict of interest statement

Authors declare that they have no conflict of interest.

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