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Course Manual

Winter School on

Recent Advances in Breeding and Larviculture of Marine Finfish and Shellfish

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PRELIMINARY EXPERIMENTS ON THE CULTURE OF THE BANDED CORAL SHRIMP STENOPUS HISPIDUS OLIVER

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Introduction

The banded coral shrimp, *Stenopus hispidus* Oliver, is the largest of the known "cleaner" shrimps which remove and eat parasites, injured tissue, and undesirable food particles from a large variety of reef fishes and helps to control gill, oral, and external parasites reef fishes as does by cleaner wrasses (Limbaugh *et al.*, 1961). *S. hispidus* occurs throughout the Indo-Pacific region (Holthius, 1946). It is usually found in pairs in a variety of reef habitats, including under coral ledges and in natural depressions in rock formations, where reef fishes come to be cleaned. In addition to its useful ecological role, the banded coral shrimp is also a beautiful and hardy specimen, which makes it very popular in the aquarium industry. Because of this popularity, a number of attempts have been carried out to breed it on a commercial scale, but none has succeeded due to high larval mortality and failure of the larvae to settle and metamorphose (Young, 1979). The purpose of this work was to study the reproductive cycle of *S. hispidus*, including maturation, mating, spawning, and hatching rate, to develop techniques for rearing larvae and inducing settlement and metamorphosis of larvae.

Materials and methods

Five mating pairs of *S. hispidus* were collected from Andhroth Island, Lakshadweep, and transported to Calicut. Subsequently they were kept in 120 L aquaria with biological filtration and crushed coral as substrate. Natural sea water was exchanged at a rate of about 25 % every day. Animals were fed a diet of frozen shrimp, mussel meat and bloodworms. Temperature was maintained between 26 - 29°C and the salinity between 33-37 ppt. Ammonia values ranged between 0.004 and 0.2 ppm. Larvae were obtained from eggs from wild-mated and captive-mated females. When eggs were close to hatching (as the colour turned to creamy white), the female was transferred to a 120 L aerated aquarium without any filtration or substrate. A 15 cm long 2"piece of grey PVC pipe was provided as shelter for the female. After eggs hatched, the female was returned to her mating aquarium and larvae counted by sampling 100 ml to estimate hatching rate. Larvae were kept in 120 L aquariums at an initial density of 20 larvae/L, and were fed *Artemia* nauplii maintained at a density of 5,000-8,000 nauplii/L. The bottom was siphoned clean every other day, alternating with days on which larvae were siphoned through a 360 micron screen, the tank washed and refilled and larvae returned to the tank along with new food.

Seawater was filtered through a 35 micron filter cloth, followed by Atman aquarium filter. Daily measurements of ammonia, temperature, and salinity, as well as food counts were made. Estimates of food density were made using 10 ml samples filtered through 53 micron mesh screen and counted in a gridded Petri dish using a dissecting microscope after preservation of the larvae in formalin. Larvae from the first matching of mating pair were maintained for 35 days in this manner. Five larvae were transferred to each of five 5L round beakers, each beaker containing a different sized coral substrate. Temperature, salinity, and ammonia were monitored daily in all beakers, with the same food concentration used in the 120 L aquarium maintained in the beakers.

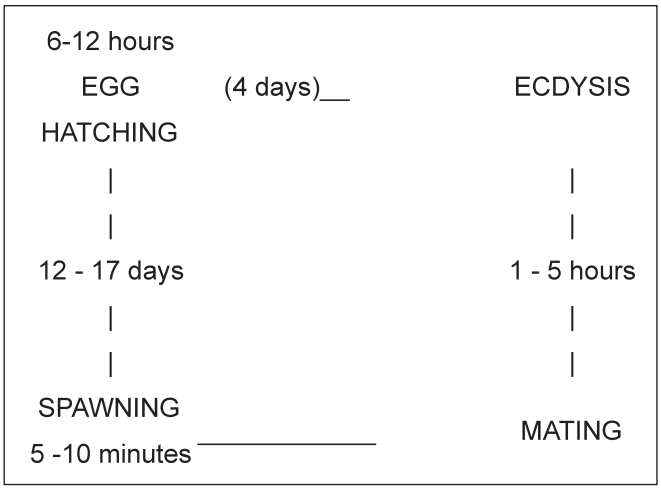
Table 1. Hatching rates of Live Larvae

Pair	Tank Volume	Hatching1	Hatching2	Hatching3
Pair1	120L	850	3	21
Pair2	120L	—	15	—
Par3	120L	560	22	10
Pair4	120L	430	43	300
Pair5	120L	600	37	240

Results

Mating in Captivity

All five pairs mated in captivity throughout the reproductive season which lasts from late April to early October in Lakshadweep. The reproductive time sequence observed in our experiments at Calicut is as follows.



At the beginning of the reproductive season it took longer for the cycle to be completed, especially between egg hatching and ecdysis (up to four days). Observed mating behavior is similar to that described by Muscat (1976), particularly pertaining to the protective behavior of the male for its female. It was observed that eggs first appear pink in color, but as they mature they change to a grey colour. From the observations, the sequence is as follows: the eggs first appear pink and as they mature they turn greenish, and finally a creamy white with black dots, which are the eyes of the larvae.

Comparison of hatching rates

Hatching rates from females which mated in the wild and which were collected while carrying eggs in their pleopods are much higher than for females which mated in captivity. In addition, the latter lost more eggs before hatching and had much higher mortality immediately after hatching. Also higher hatching rates were obtained when the mating pairs were kept in 120 L aquariums than when kept in smaller aquariums. Possibly, hatching rates are limited by the volume of the tank used, and this may be related to water quality. Larvae from the first hatching came from the eggs collected from females while carrying. Subsequent hatchings refer to eggs produced from mating in captivity. The female from pair 2 was carrying eggs when caught, but these fell from her pleopods shortly after being placed in the mating tank. This female mated in captivity, but produced very few eggs and died soon after her second mating in captivity.

Larval rearing

For the first 35 days the larvae were kept in a 120 L aerated aquarium.

Inducing settlement of larvae

All larvae died within a few days, with none settling.

Captive breeding of Stenopus hisphidous

Adults

2 day old Larvae

13 days old Larvae

Doctor shrimp larval rearing system

Doctor shrimp broodstock re-maturation tanks

Discussion

The experimental mating of *Stenopus hispidus* has been accomplished by Muscat (1976). Seasonally changing sea temperatures are known to synchronize the reproduction of most marine animals (Giese and Pearse, 1974), and Orton (1920) suggested that each species has a critical breeding temperature which must be reached before reproductive activities proceed. This is probably the reason for the reproductive cycle taking longer to complete at the beginning of the reproductive season. There is support for Orton's suggestion (known as Orton's rule) from laboratory manipulation of reproduction by temperature changes. Many summer breeding species, including shrimp (Little, 1968), have been induced to reproduce in winter. In preliminary experiments, *Stenopus* pairs maintained under natural conditions of temperature and salinity were spawned in almost all the months at Calicut, although the number of eggs was much less than that observed during the summer. This suggests that *Stenopus* can be raised throughout the year through manipulation of temperature and salinity. Many workers reported that photoperiod is a critical factor in the breeding of *Stenopus*. But in our experiments this was not found to be a critical factor, probably because of the tropical conditions.

Diet has been found to be an important factor in the maturation of penaeid shrimp. In present study also polychaetes were included in the diet of *Stenopus*, but kept no control to test if this diet affects the maturation of the banded coral shrimp. This is a subject for further experiments. Old carapaces were maintained in the tanks after both male and female molted, and they were eaten within two days by the shrimp. This suggests that it could be an additional source of calcium, which may be essential for hardening of the new carapace. Kleinholz and Brown *et.al*. (1980) reported this behavior from a number of decapod crustaceans. Feeding stops one day before ecdysis occurs and resumes one day after ecdysis is completed. The reason is probably that before moulting the animals are absorbing all mouth and gut parts, and after moulting these parts are still very soft to be used for some hours. This phenomenon has been observed in the common shore crab, *Carcinus maenas* (Linnaeus) (Robertson, 1960; Spindler-Barth, 1976), as well as in other decapod crustaceans (Travis, 1960).

All *Stenopus* females used were caught while carrying eggs and these produced a higher number of larvae. After the first mating in captivity, the next hatching was poor for all females, probably caused by capture and handling stress and/or adaption to a new environment (tank with limited area, artificial light) and diet. The next hatching showed an improvement over the second for those females kept in 120 L tanks. The water quality may be the reason for this difference, because in a larger tank the chemical and physical parameters are more stable and thus animals and eggs are subject to less environmental stress. This can also explain why the first hatching was much higher than subsequent ones. Penaeids in captivity have been reported to have reduced reproductive capabilities (Moore, 1974), and this is likely the case with *Stenopus* also.

Earlier experiments indicated the need for strong aeration as a means of keeping the larvae suspended (strong aeration reduces contact time of larvae with bottom debris) so that they are not trapped in bottom debris and die. Frequent bottom siphoning also helps in this respect and maintains water quality. Eventhough we used strong aeration and debris was siphoned off every other day, mass mortalities of larvae trapped in debris occurred. Similar works carried out in Penaeid shrimps suggested larger tanks for larval rearing. This could also probably improve survival of *Stenopus* larvae because contact time with the bottom is reduced and water quality improved. Females were observed eating their own eggs from pleopods and larvae immediately after hatching. Young (1979) observed this and reported higher larval survival by placing the gravid female in a small cage inside the hatching tank. In addition, cannibalism occurred and accounted for losses of some larvae. A solution for this also may be stronger aeration, because this has proven to prevent cannibalism in American lobster (*Homarus americanus* Edwards) larvae (Bardach *et al.* 1972) by preventing accumulation of larvae on the bottom. Old food was always discarded because *Artemia*, according to Sorgeloos (1980) loses up to 20 percent of their energy reserves during molting. *Artemia* was not fed because results from an experiment by Maddox et al., (1976), showed that freshly hatched *Artemia* had higher nutritional value than older *Artemia* fed with algae. After two to three days the unfed *Artemia* have lost most of their nutritional value and therefore exchanged it for freshly hatched *Artemia* having a large yolk reserve. The causes for the deaths of larvae during induction of settlement is considered as follows.

1. Larvae may have become trapped in the bottom substrate.
2. Timing of introducing the substrate may have been inappropriate, or
3. The larvae do not need any specific substrate to settle.

In earlier experiments, Planktonic larvae were maintained with no substrate for 55 days while feeding normally.

Mc Sweeney also kept *Stenopus* larvae for similar long periods of time, suggesting that larval development is very long. Possibly no substrate is needed, or introduction of a substrate must be precisely timed. Further work on induction of settlement is required. Additional experiments are required to determine if a specific substrate is required for larvae to settle. If it is, the substrate must be identified and the timing of its introduction determined. Mating and larval production do not appear to be problems for mass culture of *Stenopus*. Temperature and salinity manipulation could allow for continuous production of this shrimp.

Diet studies, such as those carried out for *Penaeus* spp., could improve maturation and spawning in captivity. Artificial diets should also be considered. The use of larger tanks and stronger aeration should increase larval survival. Further experiments using tanks of different volumes and larval densities are in order.

Considering the importance of *S. hispidus* in the aquarium industry, further research is needed to perfect the breeding and larval rearing techniques for its commercial scale production under captivity.

References

Bardach, J. E., J. H. Ryther and W. O. McLarney. 1972. *Aquaculture: The Farming and Husbandry of Freshwater and Marine Organisms*. Wiley-Interscience, New York. 868 pp.

Brown, A., J. McVey, B. S. Middleditch, A. L. Lawrence, B. M. Scott and T. D. Williams. 1980. The maturation and spawning of *Penaeus stylirostris*, under laboratory conditions. Proc. 11th Ann. Meet. World Maric. Soc. 11: 488-499.

Giese, A. C. and J. S. Pearse. 1974. *Reproduction of marine invertebrates.* Academic Press, New York and London. Vol. I. 546 pp.

Holthius, L. B. 1946. The Stenopodidae, Nephropsidae, Scyllaridae and Palinuridae. The Decapoda Macrura of the Snellius Expedition. I Temminckia, 7: 1-178.

Limbaugh, C., H. Pederson and F. A. Chace, Jr. 1961. Shrimps that clean fishes. *Bulletin of Marine Science* 11(2): 237-257.

Maddox, M. B. and J. J. Manzi. 1976. The effects of algal supplements on static system cultures of *Macrobrachium rosenbergii* larvae. Proc. 7th Ann. Meet. World Maric. Soc. 7:677-698.

Moore, D. W., R. W. Sherry and F. Montanez. 1974. Maturation of Penaeus californiensis in captivity. Proc. 5th Ann. Meet. World Maric. Soc. 5: 445-447.

Muscat, G. 1976. Spawning and rearing of *Stenopus hispidus. The Marine Aquarist* 7(7): 31-34.

Orton, J. H. 1920. Sea temperature, breeding and distribution in marine animals. *J. Mar. Biol. Ass. U. K.* 12: 339-366.

Robertson, J. D. 1960. Ionic regulation in the crab *Carcinus maenas* (L.) in relation to the moulting cycle. *Comparative Biochemistry and Physiology* 1: 183-212.

Sorgeloos, P. 1980. Improvement in availability and use of *Artemia* as a food source for *Macrobrachium rosenbergii* larvae. International Foundation for Science. Provisional Report #9, Bangkok, Thailand, June 1980 (mimeo). pp. 123-132.

Travis, D. F. 1960. Matrix and mineral deposition in skeletal structures of the decapod Crustacea, *In* Calcification in Biological Systems. R. F. Sognnaes, editor. American Association for Advancement of Science 64: 57-116.

Young, F. 1979. Spawning of the banded coral shrimp, *Stenopus hispidus*. *Freshwater and Marine Aquarium Magazine* 2(3): 16-17.

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