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MATURATION AND SPAWNING IN CAPTIVITY OF PENAEID SHRIMP: <u>Penaeus merguiensis</u> de Man, <u>Penaeus japonicus</u> Bate, <u>Penaeus aztecus</u> Ives, <u>Metapenaeus ensis</u> de Hann, <u>AND Penaeus semisulcatus</u> de Haan

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ABSTRACT

As no indigenous species of commercial penaeid shrimp live in the waters of French Polynesia, the preliminary step in developing shrimp aquaculture was to obtain maturation and spawning in captivity.

Five species have been investigated. Wild-caught juveniles from the New Caledonia lagoon (<u>P. merguiensis</u>, <u>P. semisulcatus</u>, <u>M. ensis</u>) and postlarvae from the Galveston NMFS laboratory (<u>P. aztecus</u>) and Fujinaga Institute (<u>P. japonicus</u>) have been used to obtain adult stock.

Shrimps are reared in circular tanks of 12 m^3 with **a** continuous water circulation through a coral sand bed. Rate of exchange is from 1 to 3 times a day. Throughout the year water temperature is between 25 and 29 C, salinity around 34.5 ppt, and pH 8.2. Solar energy is reduced by shade covers. Artificial pellets of different composition

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are given as food. In these conditions we have obtained: 1) about 1,000 spawnings and two generations in captivity from September 1973 to September 1974 with <u>P. merguiensis</u>; 2) spawnings of <u>M. ensis</u> in 1973; 3) four spawnings of <u>P. japonicus</u> in September 1974; 4) about 20 spawnings of <u>P. aztecus</u>, but only after removing one eyestalk; 5) maturation signs in <u>P. semisulcatus</u> but no spawning.

Experiments on larval rearing and growth of these species are in progress.

INTRODUCTION

Rearing of commercial penaeid shrimp in progress in many countries of the world is entirely dependent on the availability at a given time of wild-caught juveniles or ripe females ready to spawn. To begin our experiments on the feasibility of penaeid shrimp production in French Polynesia territories we were faced with the problem of the absence of commercial species in the surrounding waters. As the transportation of gravid females over a long distance is not feasible on a routine basis we have explored two approaches:

- investigation of the possibility of rearing tropical penaeid shrimp which show signs of maturation in ponds and which are found in New Caledonia waters, only six airflight hours away.
- investigation of the possibility of obtaining reproduction in captivity of some species which are commercially reared in other countries and which can be obtained as postlarvae from hatcheries elsewhere.

Success in these experiments was for us a preliminary, but necessary step to determining the feasibility of such a rearing in Polynesia. It opens also a wide field concerning selection and hybridization within the penaeid family.

The following experiments have been conducted in the Pacific Oceanologic Center (C.O.P.) which has been created by the National Center for the Exploitation of the Oceans (CNEXO), a French Government Agency.

The first facilities were in operation at Vairao, Tahiti Island, in March 1973. In May we received wild-caught juveniles from New Caledonia representing three species, <u>P. merguiensis</u>, <u>P. semisulcatus</u> and <u>Metapenaeus</u> ensis. In September <u>P. aztecus</u> postlarvae were kindly sent us by the Galveston Biological Laboratory of NMFS and in October <u>P. japonicus</u> postlarvae were purchased from Fujinaga Institute.

In 1 year, from September 1973 to September 1974, four species have reproduced in captivity. Records of such reproduction in captivity are few. It has been said that Fujinaga has obtained several generations of \underline{P} . japonicus in captivity but no details are

available. More recently Moore et al. (1974) succeeded with adults of \underline{P} . californiensis caught with undeveloped ovaries.

MATERIALS AND METHODS

At first we used various kinds of classical tanks to grow juveniles and postlarvae. We have now developed a round tank of 4 m diameter (Figures 1 and 2). Side and bottom are made of a translucent fiberglass sheet of 1 mm thickness. Height of the tank is 1.25 m for a water depth of 1 m. Holding capacity is around 12 m³.

The substrate consists of a 10 cm layer of peagravel in which concentric, semi-rigid, plastic perforated pipes (agricultural drains) are embedded. This layer is separated from a 5 to 10 cm thick second layer of mixed coral and black river sand by a screen of 1 mm nylon mesh to prevent mixture of the gravel with the sand.

The plastic perforated pipes (50 mm diameter) are fitted in a PVC tube (100 mm diameter) embedded in the sand; a similar tube (200 mm diameter) glued at a right angle forms the water inlet. This device allows water to enter the tank through the sand by gravity flow (Figure 1). Thus organic detritus particles are not trapped in the sand as in the classic circulation system using double bottom and airlift. The sediment has remained soft without any reduction signs after 9 months of continuous flow. Water discharge is achieved by means of a cylindrical screen in the middle of the tank. Two concentric tubes allow the bottom water to evacuate first. With a rate of exchange of 2 or **3** times a day the water stays perfectly clear.

The opening of a small lateral tap creates a circular movement of the tank water which concentrates fine detritus particles around the cylindrical screen where they are easily removed by strong aspiration when the central pipe is taken out for a few minutes.

Tanks are covered with different kinds of synthetic material developed to shade controlled agriculture systems; they transmit, depending on the model, between 10 and 40% of the incident light. These covers seem necessary to prevent a heavy development of benthic algae and also to attenuate the solar energy for non-burrowing species such as <u>P. merguiensis</u>.

Water is pumped directly from the lagoon. As there is a great renewal in the lagoon due to the swell action above the barrier reef, the water pumped from 10 m depth has oceanic characteristics with a low level or organic and inorganic particles. No particular filtration devices are used.

Temperature range throughout the year is 25.5 to 29 C. Salinity fluctuates around 34.5 ppt; pH is 8.2. There are 10 hours of light in July and 14 hours in December. Due to the water exchange rate in the tanks diurnal variations are negligible.

From 200 to 300 shrimp weighing between 10 and 25 g are maintained in each tank. The sex ratio is generally 1/1, but it seems that only 25% males is sufficient to obtain regular fertilization.

Examination of the ovarian development stages were made every day at 5:00 p.m. for <u>P. merguiensis</u> and at night for the burrowing species. Ovarian colorations visible through the exoskeleton allow one to select the ripe females which are transported in spawning tanks of 0.5 m³. An airlift device pumping from a conical bottom brings the eggs to four baskets which are constructed with a bottom of 207 μ plankton mesh (Figure 3). A perforated plastic plate prevents females from entering the conical bottom and eating the eggs. Females which have not spawned during the night are removed again to the maturation tanks during the day.

Shrimps were fed different kinds of artificial diets (Table 1) with an occasional supplement of fresh bonito.

RESULTS

Penaeus merguiensis

This species is one of the biggest Indopacific shrimp; average weight is 100 g (Racek, 1972).

In May and June 1973, we received 500 juveniles (2 to 6 g) from New Caledonia. First natural spawnings were observed in September; the first whole cycle in captivity was achieved in January 1974 and the second in August. More than 2,000 maturations have been observed in 14 months for females between 6 and 20 g. About 500 spawnings have been obtained. Percentage of viable eggs is quite variable between 10 to 80%. Mean number of eggs per spawning is 3,000 for a small female (6 g) and 20,000 for a bigger one (12 to 15 g).

With a sufficient number of maturation tanks, spawnings are obtained almost every day, but there seems to be a lunar periodicity with a peak lasting 1 week.

Most of the time females molting during the night are immediately impregnated. In the morning, the white and large expanded parts of the paired spermatophores are clearly visible outside of the "closed" thelyca. These soft parts disappeared during the day and the only remaining sign is a whitish mass under the thelycum plates. At this time the ovaries are undeveloped, small, and transparent. The first sign of maturation is a black line visible through the abdominal exoskeleton due to the color of scattered melanophores. This line is bifurcated in a "V" shape in the first abdominal segment. Ovaries increasing in size fill branches of the "V". In the early ripe stage a pale green mass is quite visible in the posterior part of the head and the anterior ovarian lobes are evident. One or two days later ovaries are still increasing in size; their color is olive to darker green, and sometimes expanded parts of the posterior lobes appear in the first and second abdominal segments (Figure 4). These females spawn during the night, usually before 10:00 p.m.

In the morning following the spawning the coloration has completely disappeared, and ovaries look like the undeveloped stage. These stages correspond to the five arbitrary stages described by Tuma (1967) for wild females.

Penaeus aztecus

We received in September 1973 postlarvae from the Galveston NMFS Laboratory and in June 1974 we obtained the first spawning of females between 15 and 20 g. The fertilization process is similar to $\underline{P}.$ merguiensis.

With this species held under our conditions, we never observed natural maturation. As we had noticed twice previously the beginning signs of maturation on females which had lost one eye, we decided to remove one eyestalk on 20 females of 300. Two weeks later the ovaries began to develop and spawning occurred. Ovaries of the control animals in the same tank did not develop. Maturation signs are not clearly visible by direct view through the water column, and it is necessary to handle the animal and look carefully at the abdominal cephalothoracic junction. Ovaries in the early ripe stage appear whitish without darker melanophores. One or two days later the coloration is green, and spawning takes place as the ovaries are in fact poorly developed. The posterior lobes are wider only in the first and second abdominal segments, very different from the aspect presented in the wild described by Brown and Patlan (1974). The number of eggs was between 10,000 and 20,000 with usually a lot of abnormal development. The hatching rate was between 0 and 50%.

Removing the two eyestalks induces complete development of the ovaries which become wider and darker green. Unfortunately spawning does not occur, and the females molt losing the spermatophores. Soon after there is a complete regression of the ovaries.

Penaeus japonicus

We received in October 1973 postlarvae from Fujinaga Institute, and in September 1974 we obtained natural spawning.

First maturation signs were noticed in May with ovarian posterior lobes increasing in size and showing a pink color. This development stopped as we isolated the females. We also tried removing one eyestalk; after 1 week we observed a slight development of the ovaries and also a pink color appeared, but regression soon occurred. In September we found animals with developing ovaries, both yellow and pink, and the spawning occurred for females which presented festooned posterior lobes. Four spawnings occurred, of which two gave 40,000 eggs each, all viable.

Metapenaeus ensis

This is the second most important Indopacific species of this genera by weight; average weight is 30 g (Racek, 1972).

In May and June 1973 we received 40 juveniles (4 to 8 g) from New Caledonia. We obtained in September the first spawnings, and we observed in July 1974 the first maturation of females born in captivity.

The thelycum of this species is "open", and no soft part appears on the female after fertilization. Ovarian development is quite similar to P. merguiensis with only a pink color in the developing stage, while early ripe and ripe stages are green.

DISCUSSION

Three species have spawned naturally under the described conditions. As for <u>P</u>, <u>aztecus</u>, the maturation process occurred only after the removal of one eyestalk. It seems a general rule that shrimps with only one eye mature sooner than those with two; we have obtained in this way mature <u>P</u>. <u>merguiensis</u> females of only 4 g.

First maturation time seems to be quite different according to the species. It takes about 4 months for \underline{P} . <u>merguiensis</u>, 8 months for \underline{M} . <u>ensis</u>, and around 1 year for \underline{P} . <u>japonicus</u> et \underline{P} . <u>aztecus</u>.

Light intensity seems to be an important factor for <u>P</u>. <u>merguiensis</u> which has a non-burrowing behavior in our culture conditions. More maturations were observed in a tank with a cover which allowed only 10% of the incident light to enter as compared with a cover which allowed 40% penetration.

Number of eggs seems to be normal for <u>P</u>. merguiensis as Tuma (1967) gives 100,000 for wild caught adults (100 g). We did not notice a reduction of the egg number with successive generations.

Tuma (1967), working on wild stocks of <u>P</u>. <u>merguiensis</u>, writes, "It appears that most adult females undergo insemination after each ecdysis until finally ripe ovaries occur in conjunction with insemination." In fact, in captivity insemination took place after each ecdysis but always with undeveloped ovaries. We have never seen a female molting with developed ovaries except for <u>P</u>. <u>aztecus</u> with the two eyestalks removed. We do not know whether fertilization is necessary to induce the maturation process.

It seems the whole process of maturation for <u>P</u>. <u>merguiensis</u> lasts about 3 to 4 weeks, but the time between early ripe and ripe ovaries does not exceed 2 days. We have observed the following sequence: molting, fertilization, ovarian development, spawning, molting, and a new fertilization. As we have not kept females isolated it is hard to say if one maturation is followed after spawning by another immediate maturation, but it is certain that some females with recognizable signs have spawned more than once. We observed the same sequence with <u>P. aztecus</u> and also more than one spawning for a single female.

It should be noted that we have obtained maturations under approximately the same environmental conditions as the experiments of Moore et al. (1974) on <u>P</u>. <u>californiensis</u>, i.e. the water had a strong oceanic character.

This report is the summary of preliminary observations we have made of five penaeid species. We recognize that there remain many questions to be answered by means of other experiments, but the success on a routine basis of reproduction with <u>P</u>. <u>merguiensis</u> allows us to develop such experiments and opens the way to investigations of selection and possibly hybridization in that group.

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Shigueno food		18 100	
Squid meal	47%	Polished rice	51%
Mysid shrimp meal	15	New Zealand fish meal	32
Yeast petroleum	20	Leucaena meal	2
Active sludge	5	Soya 50	2 3 3
Gluten	5 3 2 3	Meat meal	
a Starch	2	Cholesterol	0.5
Vitamin mix	3	Salt	0.5
Mineral mix	5	Guar	2
		Troca flesh	6
	100%		100%
45.111		17 000	
Shrimp meal (local)	31.5%	Troca flesh	33.3%
Norway fish meal	8	Meat and bone meal	33.3
Soya 50	3	Bonito meat	16.7
Polish rice	46	Chiken food	16.7
Brewer's yeast	5		
Fish soluble	5 2 1 0 2		100%
Lecithin	1		
Salt	0		
Guar			
Vitamin mix			

Table 1. - Artificial diets for penaeid shrimp





