INDUCED MATURATION AND SPAWNING OF INDIAN PENAEID PRAWNS

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ABSTRACT

Using the technique of unilateral eyestalk ablation, *Penaeus indicus, Penaeus monodon, Metapenaeus dobsoni* and *Parapenaeopis stylifera* have been induced to mature and spawn successfully under controlled conditions at the Narakkal Prawn Culture Laboratory of the Central Marine Fisheries Research Institute, Cochin. These prawns generally attained full maturity 10-16 days after eyestalk removal and spawned viable eggs that gave rise to normal, healthy larvae.

INTRODUCTION

Penaeid prawn hatcheries are being established in many parts of the world to meet the great demand for quality prawn seed for coastal aquaculture. A steady supply of spawners is essential for efficient programming of the hatchery operations. As the collection of spawners from the sea is a costly and uncertain operation, efforts have been made to induce the captive broodstock to mature under controlled conditions. The most successful technique for inducing maturation in captivity has been the removal of the eyestalk which is knwon to be the site of production and storage of Gonad Inhibiting Harmones (GIH). This method has been tried with varying degrees of success by Idyll (1971) and Caillouet (1973) in the U.S.A., Arnstein and Beard (1975) in the U.K., Alikunhi et al (1975) in Indonesia, AQUACOP (1975 and 1977) in Polynesia, Wear and Santiago (1976), Santiago (1977) Primavera and Borlongan (1978), Primavera (1978) and Primavera et al (1978) in the Philippines and Halder (1978) in India. The successful application of the eyestalk-ablation technique to induce maturation and spawning in *Penaeus indicus, P. monodon, Metapenaeus dobsoni and Parapenaeopsis stylifera* under controlled conditions was briefly reported earlier by Muthu and Laxminarayana (1979). A detailed account of this work is presented here.

MATERIAL AND METHODS

The work was carried out in the Narakkal Prawn Culture Laboratory of the Central Marine Fisheries Research Istitute, Cochin. The majority of the experimental animals were collected from the brackishwater ponds in the

Vypeen Island; a few spent spawners and immature adults from the sea were also used.

For removal of the eyestalk an electrocautery apparatus was used, as ablation with a pair of scissors led to profuse bleeding and heavy mortality. Cauterisation prevented bleeding and there was practically no mortality. No antibiotics were used to treat the ablated prawns. In the early experiments in which both the eyes were ablated the development of the gonad was very rapid but mortality was very high and even fully mature females did not release the eggs. So unilateral eyestalk ablation was resorted to and this proved very effective. The males were not ablated as they were fully mature even in the brackish-water environments.

The ablated females were kept along with an equal number of males in 12' diameter plastic lined pools in which the seawater was recirculated through a sub-gravel filter by air-lifts. The depth of the water in the pools was 75-90 cm. The pools were accommodated in a tile-covered shed without side walls and no artificial illumination was provided. *M. dobsoni* and *P. stylifera* were kept in 6' diameter plastic pools provided with 3 air-stones, the depth of seawater being 50 cm. The animals were fed with fresh clam (*Sunneta scripta*) meat. Live mysids were also introduced into the pools whenever available. The faeces and uneaten food were removed every day by shiphoning.

The temperature of the water in the pools varied from 24.5°C to 30.2°C and the salinity from 26.8 ppt to 38.6 ppt. (Table 1). The pH varied from 7.6 to 7.9.

TABLE 1. *Temperature and salinity of the seawater in the* 12' *diameter pools used for maturation of* P. indicus *and* P. monodon

Month	Temp. of pools °C	Salinity of pools ppt
November '78	26.2 - 29.2	28.0 - 28.5
December '78	26.0 - 28.5	31.3 - 34.9
January '79	27.5 - 29.0	29.9 - 30.6
February '79	27.0 - 29.5	37.1 - 38.6
March '79	25.8 - 30.2	31.6
April '79	27.1 - 30.2	33.6
May '79	28.0 - 30.2	28.1
June '79	25.8 - 29.0	31.8
July '79	24.9 - 27.2	28.2 - 29.1
August '79	24.5 - 27.5	26.8 - 31.5
September '79	26.0 - 27.1	27.6 - 32.2
October '79	26.0 - 27.3	28.4 - 28.9

. The fully developed ovary of the mature *Penaeus indicus, M. dobsoni* and *P. stylifera* could be clearly seen through the thin cuticle without removing the animals from the pool. But in the case of *P. monodon* the ovary could not be seen easily through the thick pigmented cuticle. Hence they had to be caught every week and examined against a bright source of light to determine the degree of development of the ovary.

Th females with fully developed gonads were removed from the pool with a dip net and placed individually for spawning in 50-litre plastic basins or 3' diameter plastic pools containing filtered seawater with good aeration. Spawning took place either the same night or on the subsequent night. After spawning the females were removed and the number of eggs spawned and subsequently the number of hatched nauplii were estimated by counting aliquot samples. The size of prawns is given in carapace length (CL).

RESULTS

Penaeus indicus: A summary of the experiments with *P. indicus* is given in Table 2 and the details of individual spawnings in Table 3.

TABLE 2. *Summary of eyestalk ablation experiments with* Penaeus indicus

Exp. No.	Date of ablation	No. of females ablated	No. of females matured	No. that spawned	No. of successful hatchings	Source of specimens
1.	1-11-78	13	3	2	nil	Ponds
2.	3-11-78	3	2	nil	nil	Ponds
3.	20-11-78	5	1	1	1	Ponds
4.	30-11-78	15	4	3	2	Ponds
5.	1-12-78	8	4	3	3	Ponds
6.	16-12-78	2	2	2	2	Ponds
7.	28-12-78	3	1	1	1	Ponds
8.	31- 1-79	7	1	1	nil	Ponds
9.	7- 2-79	11	2	1	1	Ponds
10.	12- 3-79	25	3	3	3	Ponds
11.	24- 3-79	5	4	4	4	Ponds
12.	4- 4-79	12	4	2	2	Ponds
13.	24- 4-79	17	3	3	3	Ponds
14.	29- 5-79	29	2	2	nil	Ponds
15.	28- 7-79	6	2	2	nil	Ponds
16.	4- 8-79	10	1	nil	nil	Ponds
17.	1- 9-79	8	2	1	nil	2 form sea 6 form ponds
	Total	180	40	31	22	

In 17 experiments a total number of 180 females (178 from brackishwater ponds and 2 from the sea) were unilaterally ablated. The carapace length of the ablated females varied from 24 mm. to 44 mm.; but only females 30 mm. and above attained maturity (Fig. 1). Out of the 40 females (38 from the pond + 2 from the sea) that became fully mature, 31 spawned in the laboratory and 22 spawnings yielded normal and healthy nauplii which were reared to the postlarval stage. The number of eggs spawned varied from 10,600 to 2,93,300 depending upon whether the spawning was partial or complete. The hatching rate varied from 16.7% to 97.7%. A low egg count generally resulted in low hatching rate (Table 3). The prawns became fully mature 9-27 days after eyestalk ablation. Out of the 40 females 21 attained full maturity 11-15 days after eyestalk removal.

Penaeus monodon: In 8 experiments, unilateral eyestalk ablation was performed on 62 females (42-70 mm. CL) collected from brackishwater ponds. Only specimens 52 mm. and above became mature (Fig. 1). Although 9 out of the 10 females which attained full maturity spawned in the laboratory only one of them

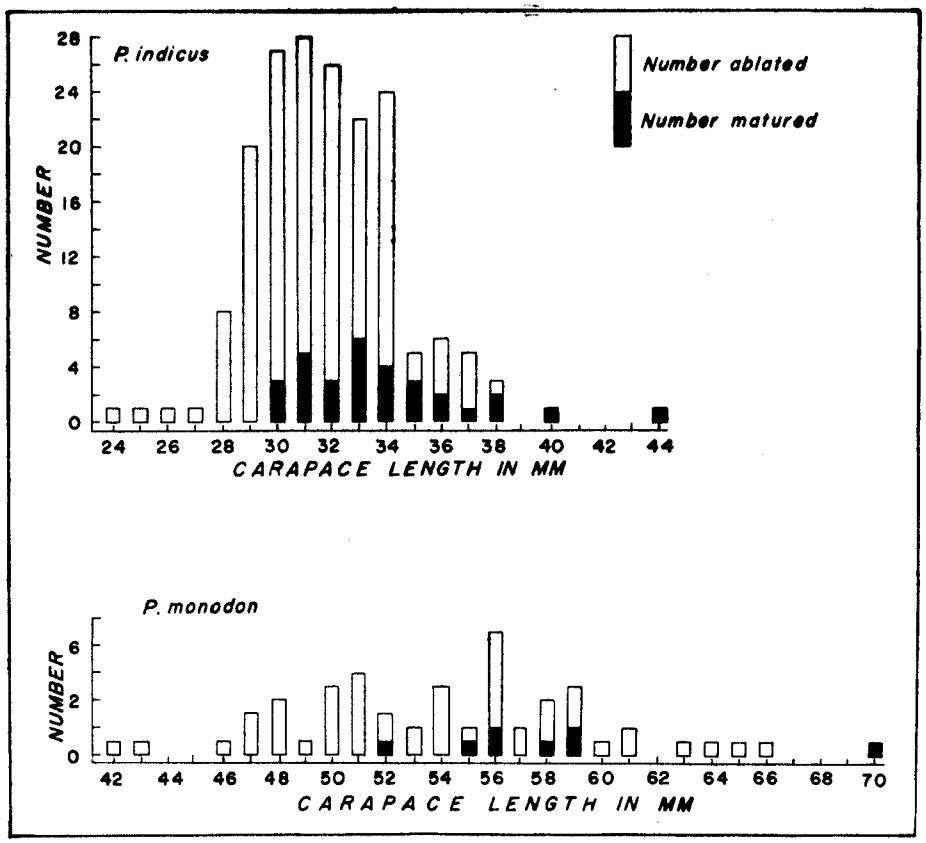

FIG. 1. Length-frequency distributions of eyestalk-ablated and matured prawns.

TABLE 3. *Details of successful spawnings of* Penaeus indicus *that matured after unilateral eyestalk ablation.*

Sl. No.	Carpace length mm	Date of ablation	Date of spawning	No. of days	Nature of spawning	No. of eggs laid	No. of naplii hateched out	Hatching rate %	Remarks
1.	35	1-11-78	13-11-78	12	partial	40,980	did not hatch	——	eggs unfertilized
2.	37	1-11-78	14-11-78	13	partial	15,180	did not hatch	——	eggs unfertilized
3.	33	20-11-78	2-12-78	12	full	61,430	50,930	82.9	—
4.	33	30-11-78	14-12-78	14	full	2,63,280	did not hatch	——	eggs unfertilized
5.	30.5	30-11-78	27-12-78	27	full	27,340	18,960	69.3	—
6.	34	30-11-78	27-12-78	27	partial	41,630	34,410	82.6	—
7.	30	1-12-78	21-12-78	20	full	1,27,240	84,760	66.5	—
8.	31	1-12-78	21-12-78	20	partial	13,640	2,280	16.7	—
9.	34	1-12-78	21-12-78	20	partial	10,620	2,080	19.6	—
10.	34.5	16-12-78	30-12-78	14	full	1,23,560	1,09,020	88.2	—
11.	30.5	16-12-78	30-12-78	14	full	1,03,830	1,01,440	97.7	—
12.	30	28-12-78	6- 1-79	9	full	62,130	56,000	90.1	—
13.	31	31- 1-79	18- 2-79	18	partial	34,300	did not hatch	——	eggs unfertilized
14.	29.5	7- 2-79	22- 2-79	15	partial	25,680	4,310	16.8	—
15.	36	12- 3-79	23- 3-79	11	partial	82,230	75,300	91.6	—
16.	33	12 -3-79	24- 3-79	12	partial	43,650	41,930	96.0	—
17.	30.5	12- 3-79	24- 3-79	12	partial	31,230	29,240	93.6	—
18.	34.5	24- 3-79	4- 4-79	11	full	1,29,600	1,17,840	90.6	—
19.	36	24- 3-79	4- 4 79	11	partial	43,900	17,520	40.0	—
20.	38	24- 3-79	5- 4-79	12	partial	67,160	18,440	27.4	—
21.	34	24- 3-79	5- 4-79	12	partial	63,730	20,830	32.7	—
22.	33	4- 4-79	15- 4-79	11	partial	60,720	44,140	72.7	—
23.	32	4- 4-79	15- 4-79	11	partial	35,840	14,160	39.5	—
24.	38	24- 4-79	10- 5-79	16	full	1,66,950	78,450	47.0	—
25.	32.5	24- 4-79	10- 5-79	16	full	66,300	57,600	86.9	—
26.	32	24- 4-79	9- 5?79	15	partial	14,540	10,920	75.1	—
27.	33	29- 5-79	12- 6-79	14	partial	56,420	did not hatch	——	eggs unfertilized
28.	34	29- 5-79	12- 6-79	14	partial	92,760	did not hatch	——	eggs unfertilized
29.	44	28- 7-79	10- 8-79	13	full	1,32,700	did not hatch	——	poor water quality
30.	40	28- 7-79	10- 8-79	13	full	1,20,000	did not hatch	——	poor water quality
31.	32	1- 9-79	28- 9-79	27	partial	15,700	did not hatch	——	poor water quality

yielded viable and healthy nauplii (Table 4) which were reared up to the postlarval stage. The number of eggs varied from 44,480 to 7,47,500 (Table 5). Fertilized eggs were obtained on 7 occasions, but the embryo did not develop in 6 cases due to poor water quality. On the one occasion that the nauplii were obtained the hatching rate was 96% and this prawn spawned within only 10 days after ablation; all the others attained maturity after a period varying from 12-66 days.

TABLE 4. *Summary of eyestalk ablation experiments with* Penaeus monodon

Exp. No.	Date of ablation	No. of females ablated	No. of females matured	No. that spawned	No. of successful hatchings	Source of specimens
1	12-3-79	8	1	1	nil	ponds
2	30-3-79	4	1	nil	nil	ponds
3	4-4-79	1	1	1	nil	ponds
4	12-5-79	2	nil	nil	nil	ponds
5	28-7-79	4	nil	nil	nil	ponds
6	11-8-79	1	1	1	1	ponds
7	24-8-79	7	2	2	nil	ponds
8	1-9-79	35	4	4	nil	ponds
	Total	62	10	9	1	

TABLE 5. *Details of successful spawning of* Penaeus monodon *that matured after unilateral eyestalk ablation.*

Sl. No.	Carpace length mm	Date of ablation	Date of spawning	No. of days	Nature of spawning	No. of eggs laid	No. of naplii hateched out	Hatching rate %	Remarks
1.	58.5	12-3-79	24- 3-79	12	partial	44,480	did not hatch	—	eggs unfertilized
2.	52	4-4-79	20- 4-79	16	full	3,08,100	did not hatch	—	eggs unfertilized
3.	66	11-8-79	21- 8-79	10	full	7,47,500	7,19,500	96.0	—
4.	70	24-8-79	16- 9-79	23	partial	1,36,800	did not develop	—	poor water quality
5.	56	24-8-79	22- 9-79	29	partial	75,500	did not develop	—	poor water quality
6.	58	1-9-79	24-10-79	54	partial	3,76,000	did not develop	—	poor water quality
7.	56	1-9-79	3-11-79	64	partial	3,50,080	did not develop	—	poor water quality
8.	59	1-9-79	1-11-79	61	partial	1,65,280	did not develop	—	poor water quality
9.	55	1-9-79	5-11-79	66	partial	1,24,200	did not develop	—	poor water quality

Metapenaeus dobsoni: Three immature females (22-26 mm. CL) obtained from the ponds were unilaterally ablated. All of them attained full maturity and spawned 9-16 days after eyestalk removal and yielded healthy nauplii (Table 6).

Parapenaeopsis stylifera: Three spent females (104-109 mm. CL) obtained from the sea were unilaterally ablated. All of them matured and spawned 12-13 days after eyestalk removal. Two of the spawnings yielded healthy nauplii (Table 7).

TABLE 6. *Details of eyestalk ablation experiments with* Metapenaeus dobsoni

S. No.	Carapace length mm	Date of ablation	Date of spawning	Nature of spawning	No. of eggs laid	No. of nauplii obtained	Hatching rate %	Source of specimen
1	24	15-12-78	31-12-78	full	58,870	57,290	97.3	ponds
2	26	15-12-78	31-12-78	full	54,290	48,830	89.9	ponds
3	22	15-12-78	24-12-78	full	17,780	11,980	67.4	ponds

TABLE 7. *Details of eyestalk ablation experiments with* Parapenaeopsis stylifera

S. No.	Carapace length mm	Date of ablation	Date of spawning	Nature of spawning	No. of eggs laid	No. of nauplii obtained	Hatching rate %	Source of specimen
1	28.5	3-11-78	15-11-78	partial	15,780	did not hatch	—	spent female from sea
3	26.5	12-12-78	25-12-78	partial	38,150	33,590	88.0	—do—
2	25	3-11-78	16-11-78	full	92,760	88,730	95.6	—do—

DISCUSSION

The Indian prawns *Penaeus indicus, Metapenaeus dobsoni* and *Parapenaeopsis stylifera* have been induced to mature and spawn in captivity for the first time. The latter two species appear to be quite hardy as they attained maturity after eyestalk ablation even in a 6' diameter pool containing seawater which was kept aerated with air stones. But *P. indicus* and *P. monodon* did not attain maturity under these conditions. Positive results were obtained with these two larger species only after the sub-gravel fitter with air-lift recirculation was fitted in the pools. It is clear that the quality of the seawater is of prime importance in the maturation of these prawns. Recirculation of seawater through gravel filters, otherwise known as biological filters, is a well known method of converting the toxic ammonia excreted by the animals in the culture tanks to

harmless nitrates by the nitrifying bacteria that grow on the gravel surfaces (Spotte 1970). This method of purification of the seawater has yielded very good results during the present investigations.

P. monodon appears to take a longer time to mature in captivity than *P. indicus* which resembles *P. merguiensis* (vide Alikunhi et al 1975, AQUACOP 1975) in maturing 11-15 days after eyestalk removal. Primavera et al (1978) found that *P. monodon* takes generally 24-25 days to mature after unilateral eyestalk ablation while during the present study *P. monodon* took up to 66 days to mature. Frequent current failures leading to disruption of the recirculation system delayed the maturation process. However, the only viable spawning occured in the specimen which matured 10 days after eyestalk ablation. It is likely that delayed maturation may result in non-viable aggs.

The size of the prawn chosen for eyestalk ablation seems to have considerable effect on the process of maturation. The histograms in Fig. 1 indicate that none of the specimens of *P. indicus* below 30 mm. CL responded to this treatment. It is also seen that the percentage of prawns which matured after eyestalk removal was relatively high among the larger sizes. In *P. monodon,* however, this tendency was less marked. In this species the minimum size of female which showed a positive response after eyestalk ablation was 52 mm CL.

Although no definite correlation could be established between maturation and the salinity-temperature regime of the pools, it was found that, generally, the prawns attained maturity faster when the temperature of the pools was 28.5°C-30.2°C and the salinity 30-33 ppt.

During the present investigations, although maturation and spawning could be throughout the period from November 1978 to October 1979, viable nauplii could be obtained only during the period December 1978 to May 1979 (vide Tables 2 and 4) when the quality of seawater was good. During the rest of the period, which coincided with the south west and north east monsoon season, either the seawater was very turbid or it contained swarms of harmful dinoflagellates such as *Gymmodinium* sp and *Gonyaulax* sp.

The fact that both *P. indicus* and *P. monodon* collected from brackish-water ponds could be induced to mature and spawn in captivity makes it now possible for a prawn farmer to develop his own broodstock from the prawns grown in his ponds.

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