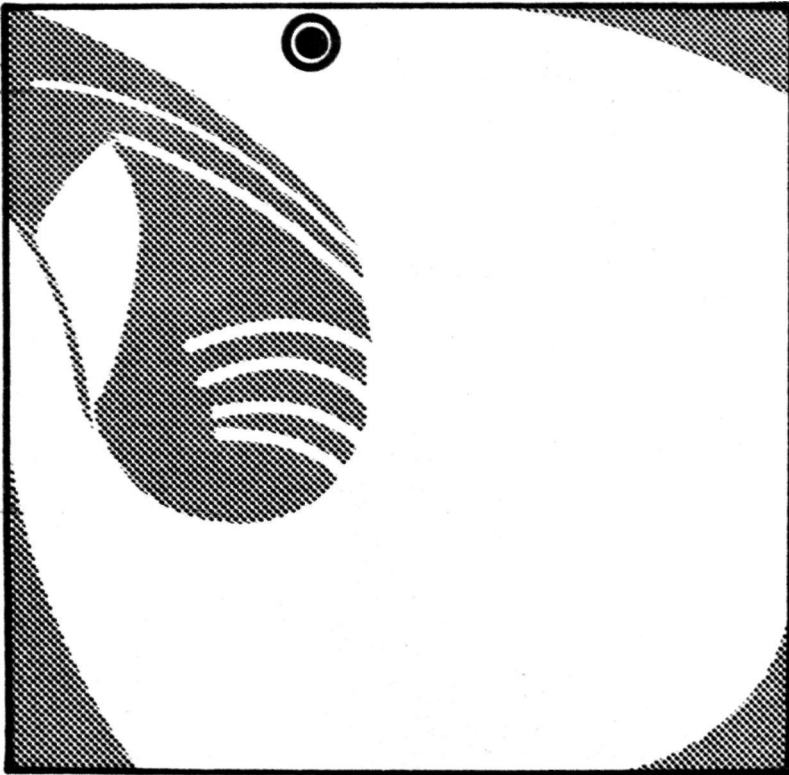


broodstock of sugpo

(Penaeus monodon Fabricius)



J.H.Primavera



AQUACULTURE DEPARTMENT
SOUTHEAST ASIAN FISHERIES DEVELOPMENT CENTER

COPYRIGHT 1985, 1989
AQUACULTURE DEPARTMENT
SOUTHEAST ASIAN FISHERIES DEVELOPMENT CENTER

ISSN-0115-5369

First edition, 1979
Second edition, 1980
Third edition, 1983

First printing, July 1983
Second printing, August 1984
Third printing, May 1985
Fourth printing, October 1985

Fourth edition, 1989
First printing, May 1989
Second printing, May 1996

All rights reserved by the publisher. No part of this publication may be reproduced or transmitted systematically in any form by any means without permission in writing from SEAFDEC Aquaculture Department.

Cover Design: Edgar Gonzales

PREFACE

It is heartening to note that since 1983, many hatcheries have started to use ablated females from wild or pond broodstock. To a large degree this is due to the price for wild spawners which at present ranges from P300 to more than P1,000.

In this edition, the material has been reorganized and updated to include recent findings and their corresponding references. The section on maturation pens has been deleted because of its limited applicability to commercial hatcheries.

J.H.P.
February 1989

PREFACE TO THE THIRD EDITION

The two major problems that plague the Philippine hatchery industry and account for underproduction are lack of technicians and lack of spawners.

Addressed to the latter problem, Fisheries Administrative Order No. 141 "banning the export of live gravid shrimps of the genus *Penaeus*" was jointly issued by the Ministry of Natural Resources and the Bureau of Fisheries and Aquatic Resources in December 1982. Nevertheless, the export of wild *Penaeus monodon* spawners to Taiwan continues given the lucrative prices of P5,000 to P10,000 commanded by a single wild spawner among highly competitive Taiwanese hatcheries. Middlemen for the export market can outbuy local hatcheries whose going rate is from P100 (Iloilo) to P400 (Metro Manila).

Parallel to a more effective enforcement by government authorities of the export ban, there is a need for private hatcheries to develop their own broodstock to provide part, if not all, of their spawner requirements. Using unilateral eyestalk ablation, completion of the life cycle of *P. monodon* was first achieved in the Igang, Guimaras pens of the SEAFDEC Aquaculture Department in 1975. Already, a dozen hatcheries in the country depend to some degree on broodstock, in contrast to six years ago when only the AQD hatcheries in Tigbauan, Iloilo and Batan, Aklan were using ablated spawners.

This manual aims to make available to prawn hatchery operators, government extension workers, fisheries students, and others interested

in aquaculture the latest technology on induced maturation and broodstock of *P. monodon*. More of a biological primer, the first sections deal with some basic aspects of the reproductive biology of the species including such topics as mating, maturation, spawning, and fecundity. This background material is followed by a detailed discussion of the broodstock tank and pen system, and the ablation process, which is the only reliable method to date of inducing ovarian maturation in captive *P. monodon*.

Lastly, a list of selected references on penaeid maturation is given for those interested in further reading.

J.H.P.
June 1983

PREFACE TO THE SECOND EDITION

The successful culture on a mass scale of any plant or animal species depends on a reliable supply of seed, among other factors. Fry supply is still the most serious problem in the development of the sugpo (*Penaeus monodon*) pond industry in the country and Southeast Asia. Traditionally, sugpo and other prawns and shrimps have been secondary products of milkfish ponds, the fry coming in with the tide. Some prawn farmers have gone into *P. monodon* monoculture using wild fry bought from coastal gatherers. The successful spawning of gravid females (with ripe eggs) from the wild paved the way for mass production of fry.

Nevertheless, the disadvantages of high costs and seasonal availability of wild females remain to this day. In contrast to only seven pesos for an immature female sugpo from wild or pond sources at present, a fully ripe spawner will fetch anywhere between P15 to P150 in various spawner collection sites in the country. The exorbitant prices are due to a lesser or greater extent to the high demand for our *P. monodon* spawners and/or nauplii in Taiwanese hatcheries. To protect our young and growing prawn industry, particularly the hatcheries, there is a need to promulgate laws that will ban the commercial export to foreign countries of our prawn spawners, larvae and postlarvae, as is the case with milkfish.

From accidental pond entry of wild fry, sugpo pond culture has progressed to stocking first of wild fry, then hatchery-reared fry from

wild spawners. The last step in the development of *P. monodon* seed supply should be the hatchery production of fry from females matured in captivity.

Using unilateral eyestalk ablation (removing or destroying one eyestalk), completion of the life cycle of *P. monodon* was first achieved in the Igang Station of the SEAFDEC Aquaculture Department in 1975. Yet most of the females spawned in the Tigbauan hatcheries up to 1976 came from the wild. Since 1977, the trend has reversed with more than 90% of gravid *P. monodon* females coming from ablated stock in marine pens and concrete tanks.

Already, the technology for prawn hatchery and pond culture on a semi-intensive scale has been extended to the private sector through various seminars and workshops of the Department and through publications. After lessons on how to rear the larvae to stocking and then to market size, it is only logical to follow up with techniques on how to produce the spawners. Moreover, with the extension of broodstock technology, the responsibility of spawner production is shifted to the private sector, thereby allowing research institutions to concentrate on other maturation-related problems.

J.H.P.
1980

CONTENTS

PREFACES

- I. REPRODUCTIVE BIOLOGY OF *PENAEUS MONODON*
 - A. Spawner and broodstock sources 1
 - B. Sex differentiation 1
 - C. Maturity 3
 - D. Courtship and mating 3
 - E. Ovarian maturation stages 5
 - F. Spawning and rematuration 5
 - G. Fecundity and egg quality 8
- II. BROODSTOCK TECHNOLOGY
 - A. Transport and stocking 10
 - B. Eystalk ablation 10
 - C. Sampling 13
 - D. Processing of spawners, eggs, nauplii 13
 - E. Procurement and transport of nauplii 17
 - F. Feeds and Feeding 17
 - G. Mortality 18
- III. MATURATION TANK SYSTEM
 - A. Construction 19
 - B. Water management 23
 - C. Construction and operating costs 23
- IV. SUGGESTED READINGS 24

I. REPRODUCTIVE BIOLOGY

A. Spawner and broodstock sources

Penaeus monodon (Tagalog, *sugpo*; Hiligaynon, *lukon*; Cebuano, *pansat*) is distributed throughout the Philippine archipelago. However, its abundance in any given area is seasonal and its availability to hatcheries depends on the catch of local fishery, e.g., by trawlers and native fishing gears.

Prawns are caught in offshore waters by trawlers and baby trawlers (motorized pumboats), and in brackishwater areas by tide-dependent stationary gears such as fish corrals, lift nets, and lever nets. Offshore *P. monodon* has a characteristic reddish to pinkish shade in contrast to the dull green or greyish coloration of the catch from brackishwater areas.

Although most hatcheries use wild spawners, a good number rely on broodstock to meet part, if not all, of their need for spawners. The use of ablated females offers two distinct advantages—spawners are cheaper to produce (P30-100/immature female vs. P300-1000/wild ripe female) and can be had throughout the year.

Broodstock may come from pond-reared animals or wild stock caught in mangrove and offshore areas. The minimum weight is 80-150 g (7-12 pcs/kg) for females and 50-120 g (8-20 pcs/kg) for males. One major disadvantage in the use of pond broodstock is that the latter is generally unavailable because of the minimum pond age requirement of 8-10 months (or 10 months to 1 year from spawning). Only 3- to 6-month old *sugpo* are normally available from ponds. Another advantage in the use of wild broodstock is its fast turnover — 25-30 wild females can produce 3-7 million nauplii in only 4-6 weeks after ablation compared to 8-12 weeks for pond females — meaning less operating costs (feeds, power, etc.) for the operators. Also, wild broodstock can be maintained on squid and other fresh or frozen food alone while pond broodstock requires supplementary feed pellet for maturation.

B. Sex differentiation

Females are generally larger than males belonging to the same age group (Fig. 1). Sex is determined by looking at the ventral or underside of the prawn. Females have a flattened organ (thelycum) located between the bases of the last pair of walking legs (Fig. 2) where the

spermatophores are deposited. In contrast, males possess a paired structure (petasma) that aids in sperm transfer and swellings (terminal ampoules) between the bases of the fifth pair of walking legs. These swellings contain the spermatophores.

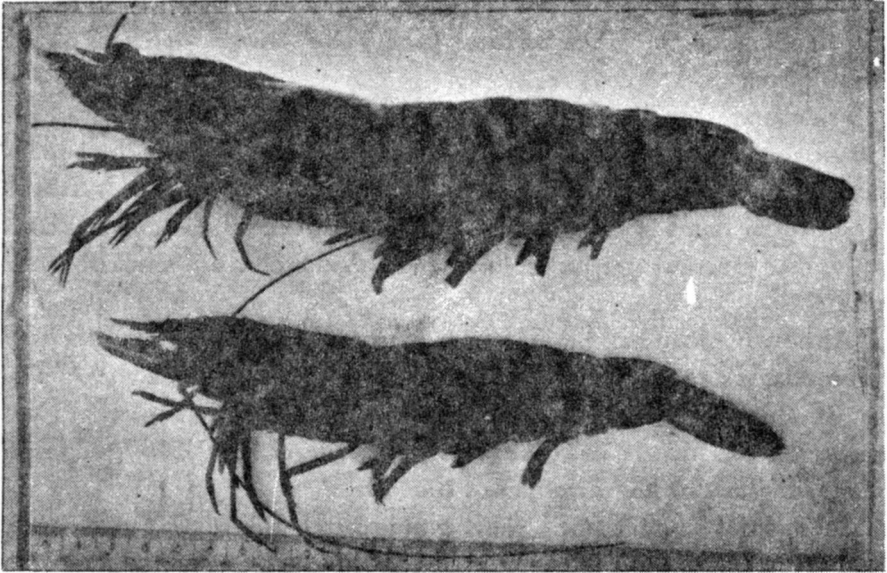


Fig. 1. Sugpo (*Penaeus monodon*) from pond stock. Female (above) is generally larger than male (below).

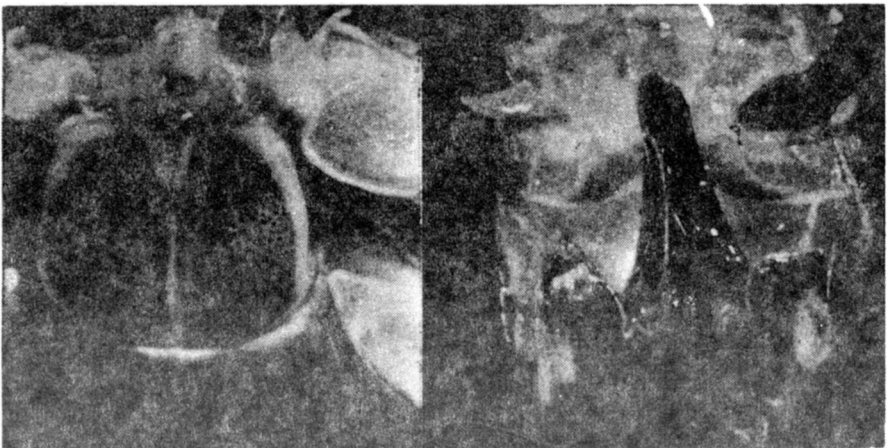


Fig. 2. Female *P. monodon* possesses a flattened thelycum (left). Male has a petasma (right).

C. Maturity

Development of the external sex organs precedes maturation of the ovaries and testes. Consequently, first mating occurs earlier than first spawning, the latter event usually at about one year in many penaeids.

In *P. monodon*, copulation takes place at 4-5 months and first spawning at about 10 months. The minimum recorded weight is 75 g for a wild spawner in the Tigbauan hatcheries and about 60 g for a wild-caught female with sperm present in the thelycum. In terms of maximum size of the species in offshore trawler catch, prawns weighing 300 g each are not rare. Fishing boat operators report a maximum weight of 600 g for a single female.

D. Courtship and mating

Courtship (precopulatory) behavior starts when the newly molted female attracts 1-3 hard-shelled males that follow her as she makes brief upward swimming movements over distances of 50-80 cm. Eventually, one male positions himself directly below the female. The pair engages in parallel swimming (Fig. 3a), during which the male turns to an upside down position, trying to attach his underside to that of the female (Fig. 3b). If successful, the male quickly turns around from this direct alignment to a perpendicular position (Fig. 3c), curves his body U-shaped around hers and flicks head and tail simultaneously (Fig. 3d). It is probably during this time that the spermatophores are inserted inside the thelycum.

Molting of the female is a prerequisite to copulation. Since *P. monodon* belongs to the group of penaeids possessing a closed thelycum, the spermatophores have to be inserted into containers within the thelycum. Insertion can take place only when the thelycum is soft, i.e., when the female has just molted. The sperm remain within the thelycum until the female spawns a few weeks later to release the sperm together with the eggs. If she does not undergo spawning because her ovaries remain immature, the sperm sacs are expelled with the shell during the next molting. Fresh spermatophores are inserted during the next mating.

Copulation generally takes place at night following molting. Among adult *P. monodon*, molting occurs about every 3-5 weeks.

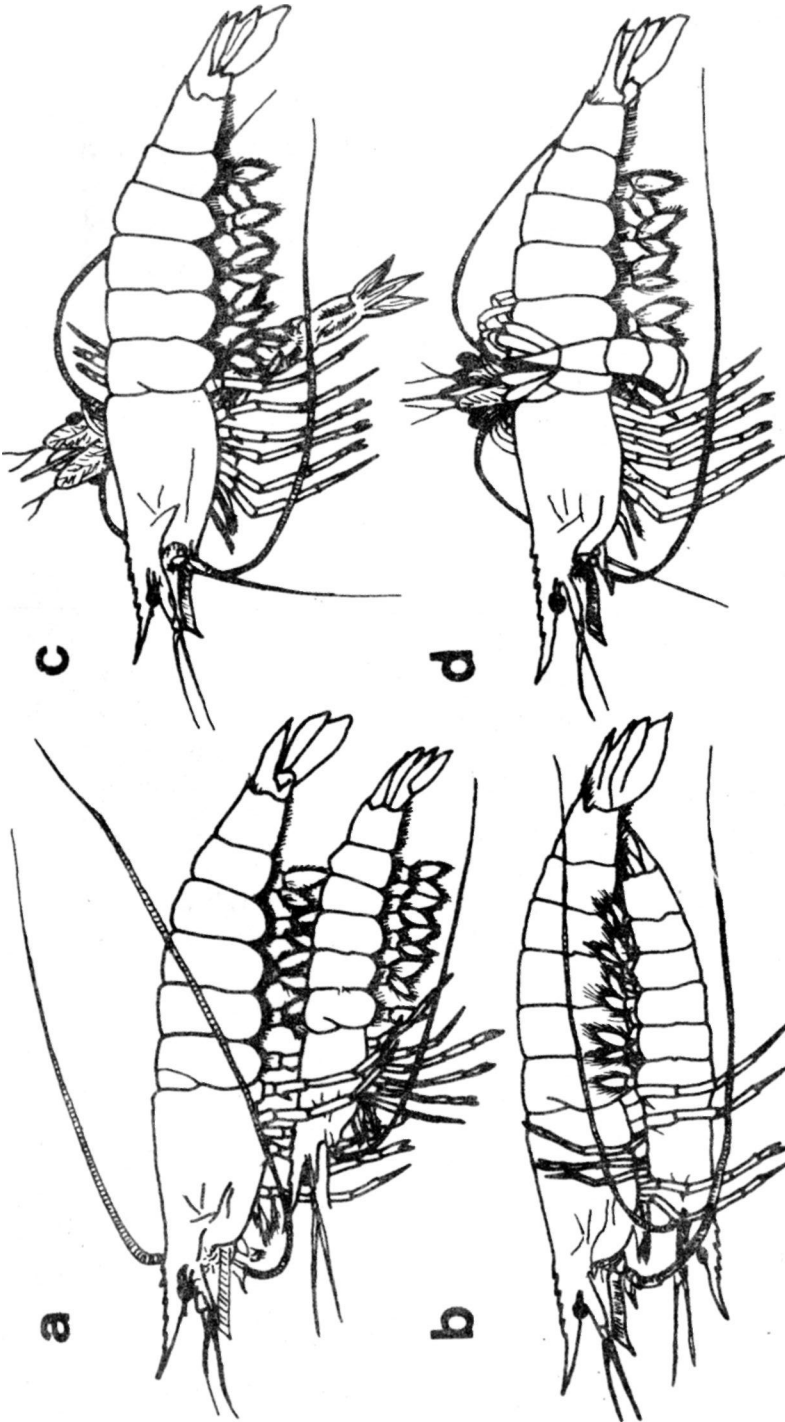


Fig. 3. Courtship and mating behavior in *Penaeus monodon*. (a) Female above-male below in parallel swimming; (b) Male turns ventral side up and attaches to female; (c) Male turns perpendicular to female; and (d) Male curves body in a U-shape around female and flicks head and tail simultaneously. (Redrawn from J.H. Primavera, *Crustaceana* 37:287-292. 1979.)

E. Ovarian maturation stages

The different maturation stages are (Fig. 4) distinguished as follows:

Stage I (Immature) — Ovaries thin, transparent, not visible through the dorsal exoskeleton. On dissection these appear as colorless strands without visible eggs. Some hatchery technicians refer to this as Stage 0.

Stage II (Early maturing) — Ovaries observed through the exoskeleton as a linear band as these start to increase in size, particularly in the anterior and middle lobes. Color of dissected ovaries ranges from cloudy white to light brown and grayish-green.

Some hatchery technicians refer to this as Stage I if the band is thin, and Stage II if the band has grown thicker.

Stage III (Late maturing) — Ovaries visible through the exoskeleton as a thick, solid, dark linear band as these expand considerably from the anterior thoracic to the posterior abdominal region. A somewhat "diamond" or "butterfly" outline can be seen at the level of the first abdominal segment. Dissected ovaries are mostly light olive-green, firm and granular in texture, and with visible clumps of eggs.

Stage IV (Mature or ripe) — The diamond-shaped expansion at the first abdominal segment is larger and more distinct; the linear band is thicker. Upon dissection, the ovaries appear dark olive-green and are so distended as to occupy nearly all available space in the body cavity.

Stage V (Spent) — Completely spent ovaries are limp and thin and outwardly appear similar to Stage I (immature) ovaries. Dissected ovaries are yellowish but become more and more white as regression continues.

Females that are partially spent have either the front or rear portion of the ovary still distended.

F. Spawning and rematuration

Spawning generally takes place between 8:00 p.m. and 6:00 a.m. although most females spawn between 10:00 p.m. and 2:00 a.m. Normally at rest or slow-moving at the bottom, a female about to spawn becomes restless and starts swimming upwards in circles. The eggs (and sperm) are released, often forcefully, as she swims and such a

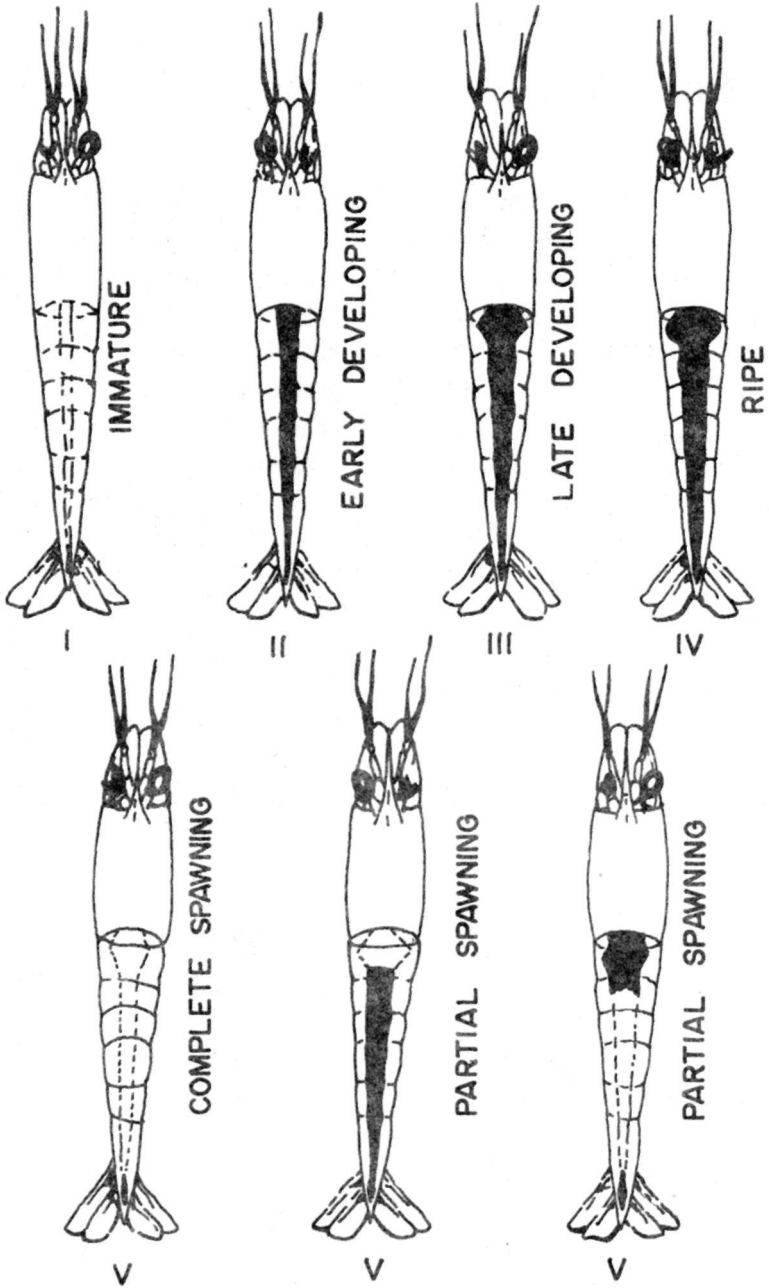


Fig. 4. Appearance through the dorsal exoskeleton of the ovary of *Penaeus monodon* at different stages of maturity.

release may continue even as she returns to the bottom. Active movements of the swimming legs or pleopods disperse the eggs and the non-motile sperm. Spawning lasts from 2 to 7 minutes.

Ovarian material released into the water together with the eggs form bubbles completely covering the water surface as an effect of aeration. After a few minutes, the bubbles break up and disappear within half an hour after spawning. The material turns into pink to orange scum forming along the sides of the spawning tank a thin to very thick ring which hatchery technicians often take as a sign of spawning (section II, D). In the absence of aeration, no scum is formed.

Of a given batch of ripe (Stage III and IV) females, some will spawn completely, others partially, while the rest may not spawn at all. Partial spawning or non-spawning is associated with stress due to transport, handling, crowding, etc. Unspawned and partial spawners will either spawn or continue spawning in the next 2-3 days or absorb their ovaries. Partial or complete spawning can be determined by holding the female against a bright light - some of the anterior or posterior portions of the ovary remain in partial spawners while complete spawners have no trace of the ovary (Fig. 4).

A gravid female that does not spawn for 2-3 successive nights but remains in Stage III or IV without regressing may have the "white ovary" or "milky ovary" disease. Infection by a protozoan (microsporidian) parasite causes the ovary to become whitish or milky and yet retain the diamond or butterfly outline visible externally.

In nature, *P. monodon* females probably have multiple spawnings in a year. Penaeid or marine prawns have a short life of 1-2 years, as documented for *P. merguensis* and *P. semisulcatus*. Data for broodstock that have matured without ablation show that on the average, *P. merguensis* spawns once every 2.6 months, and *P. japonicus* once every 2.8 months.

For a given number of ablated *P. monodon* females, 65% will spawn once, 35% a second time, and 15% a third time. Subsequent spawnings may take place as quickly as 3-5 days after the preceding one. Rematuration rates can be increased by reducing factors that cause post-spawning mortality among spent females, thereby increasing their chances to remature and spawn again. Nevertheless, both maturation and hatch rates decline progressively 6-8 weeks and 10-12 weeks after initial ablation of wild and pond stock, respectively.

G. Fecundity and egg quality

Fecundity or number of eggs in a complete spawning averages 300,000 (range: 100,000-800,000) for ablated females, and 500,000 (range: 200,000-1 million) for wild spawners.

Both wild and ablated spawners may produce good or bad eggs. To avoid wasting time and effort in rearing inherently weak larvae, the quality of eggs from a given spawning should be determined as early as possible. Toward this end, a system of classification of *P. monodon* eggs into five different types based on appearance (Fig. 5) has been established. Because technicians normally report to the hatchery at 8:00 a.m., the various egg types are described below according to their appearance in the morning (8:00-10:00 a.m.) after a spawning.

Type A₁ (good eggs) - nauplius undergoing normal development with distinct setae or bristles (only the multicell stage may be visible if the female spawned late, e.g., 5:00 a.m.); mean hatch rate (HR), 58%; larvae strongly phototactic, i.e., swim actively toward a source of light.

Type A₂ (not-so-good eggs) - development of embryo either delayed or abnormal in comparison to A₁ eggs of the same batch; mean HR, 32%; newly hatched nauplii may be weak.

Type B - bad eggs showing irregular cytoplasmic formations; 0% HR.

Type C - bad eggs with cytoplasm remaining a single undifferentiated mass; 0% HR.

Type D - bad eggs with very little remaining cytoplasm because of bacterial invasion; 0% HR.

There is a highly significant linear relationship between the proportion of A₁ eggs and hatch rates of ablated pond and wild stock.

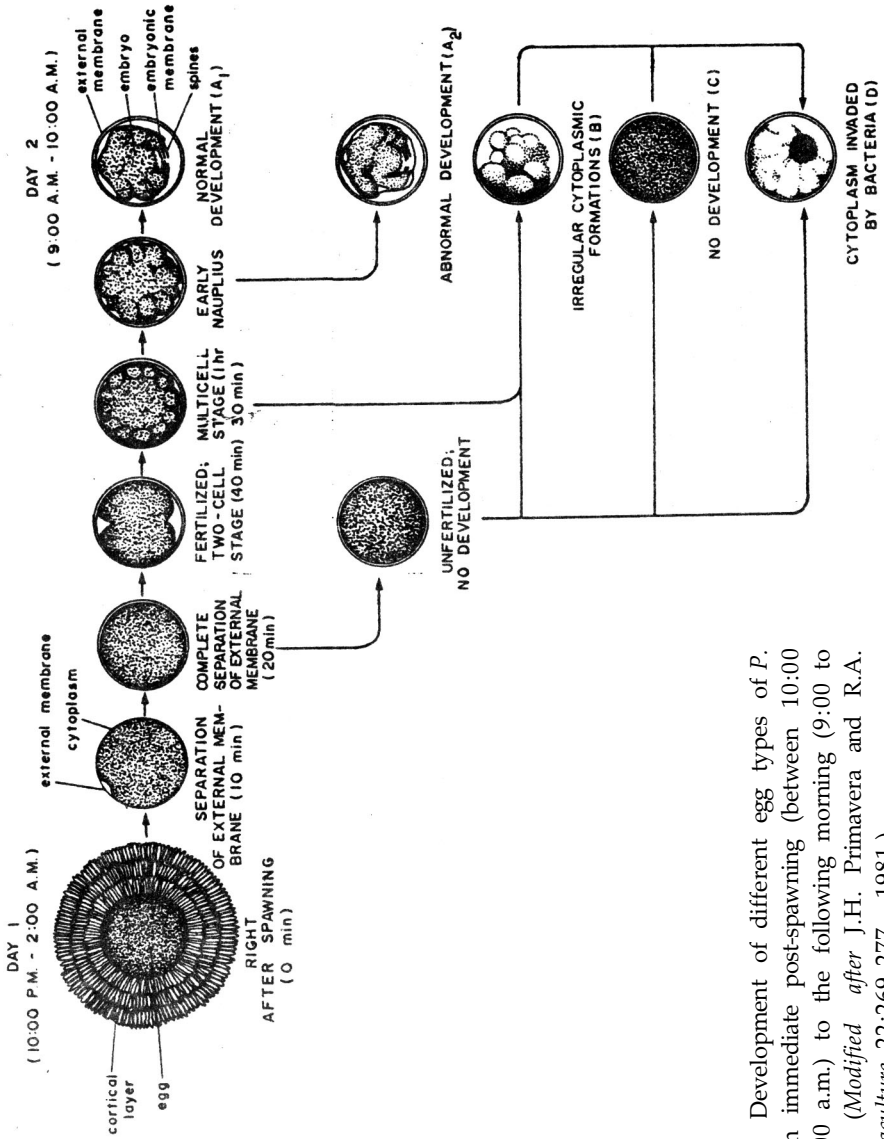


Fig. 5. Development of different egg types of *P. monodon* from immediate post-spawning (between 10:00 p.m. and 2:00 a.m.) to the following morning (9:00 to 10:00 a.m.). (Modified after J.H. Primavera and R.A. Posadas. *Aquaculture* 22:269-277. 1981.)

II. BROODSTOCK TECHNOLOGY

There are three major approaches to induce ovarian maturation in penaeids — eyestalk ablation, nutritional, and environmental. This section covers both ablation (II, B) and nutrition or feeding (II, F) approaches. The environmental approach is discussed elsewhere (section III).

A. Transport and stocking

Ideally, the broodstock tank or pen should be located close to pond and wild sources of *P. monodon* in order to minimize stress in transport. Broodstock may be transported in containers provided with aeration. A one-ton PVC or canvas tank can accommodate up to 400 adult prawns (see section I, A for sizes) if travel time is only 1 hour or shorter. With transport periods of 4-5 hours, not more than 200 prawns should be transported during each trip. Early morning or late afternoon transport is recommended to avoid high daytime temperatures that may add to stress of the animals. Broodstock may also be transported by boat.

Upon arrival, transfer the prawns to holding tanks with water of the same salinity and temperature as the water in the transport container and then acclimate them for 1-2 weeks. Once the prawns have sufficiently recovered as shown by active behavior and little or no additional deaths, disinfect for 1 hour in 50 ppm formalin, then stock.

Adult prawns are stocked at a ratio of 1-2 females to 1 male. Males provide "insurance" in case females after ablation shed off their shells with the enclosed spermatophores. (In all-female broodstock tank, maturation and fecundity rates were found to be normal but hatch rates were zero because the eggs were unfertilized.) Otherwise, some hatchery technicians stock all-female tanks after checking the mated condition of prawns by the external appearance of the thelyca. Up to 60 prawns per tank (12 m³) can be stocked although lower densities are desirable for larger area per prawn.

B. Eyestalk ablation

P. monodon females, unlike males, do not attain maturity in captivity unless they undergo ablation or destruction of one eyestalk. In

the eyestalk are found the production and storage sites of a gonad-inhibiting hormone which prevents the maturation of ovaries. In nature, some environmental factors cause the decrease of this hormone as the prawns migrate from estuaries to offshore areas where they normally spawn. Eyestalk ablation eliminates this substance or at least reduces it to a level at which maturation of the ovary can take place.

Prawns should be ablated only when hard-shelled, never when newly molted (soft-shelled) or ready-to-molt (with whitish spots on shell). Select only healthy animals with clean shells, intact legs and tails, and uninfected gills. The procedure is as follows:

1. Hold the prawn gently but firmly with one hand. Check the sex (see I, B). Only females are ablated. Do not use prawns with broken or diseased external sex organs (petasma or thelycum).

2. Check the ovarian maturation stage by external examination. Only immature (Stage I) and early maturing (Stage II) females are ablated. Late maturing (Stage III) ripe (Stage IV) females are ready to spawn.

3. Examine the thelycum closely for presence (bulging, with a whitish vertical streak on each side) or absence (depressed, evenly colored with no whitish streak) of sperm sacs. Only females which appear to have sperm deposited in the thelycum are ablated; the rest are returned to the holding tank for mating with males.

4. Ablation is performed on either left or right eye. However, an already infected or otherwise damaged eye should be ablated to leave one unablated healthy eye.

5. Ablation is performed through of the following ways (Fig. 6).

- a. *Pinching* (Fig. 6a) — Make an incision on the eye with a sharp blade, squeeze out the contents, and crush the eyestalk 2-3 times to destroy the tissue.

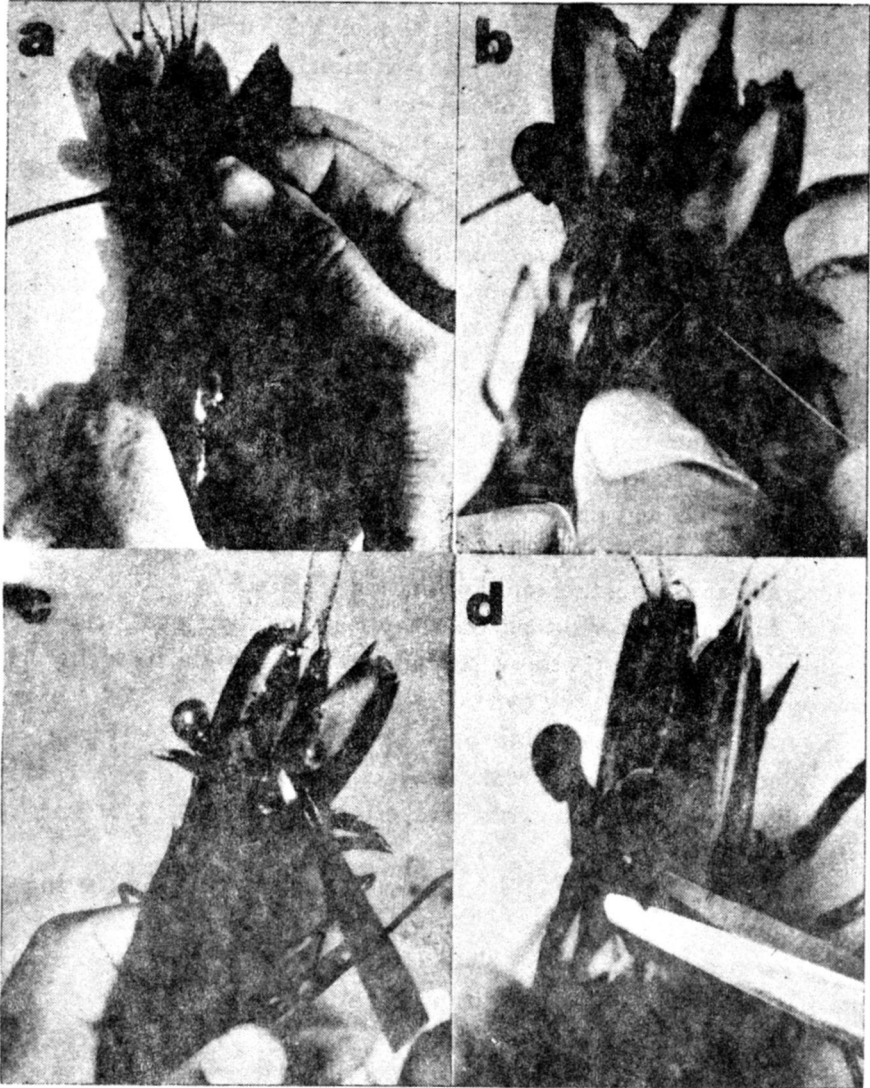
- b. *Ligation* (Fig. 6b) — Tie the eyestalk with a piece of string at the base close to the carapace. It should fall off in a few days.

- c. *Cautery* (Fig. 6c) — Ablate the eyestalk by squeezing with a pair of red-hot forceps or by using an electric cauterizer (nichrome wire, 5 volts).

- d. *Cutting* (Fig. 6d) — Cut off the eyestalk with a sharp pair of scissors about 3-5 mm from the base. Cauterize the cut eyestalk to prevent excessive loss of blood.

6. Ablation should be performed quickly and with the prawn underwater to minimize stress. After ablation, immediately release the prawn in the tank or pen. Mortality due to ablation stress should not be more than 10%.

Pinching is the preferred method because one person can do it alone. The eyestalk heals even without the use of antibiotics; the



external (corneal) layer forms the scar tissue in a week's time. Ligation needs two persons - one to hold the prawn while another ties the eyestalk. Cautery requires a cauterizer which may not be easily available. Cutting is inconvenient because it requires additional sealing by cautery; otherwise, loss of blood from the open (cut) eyestalk may lead to mortality. Application of antibiotics to the ablated eyestalk may prevent infection but also makes the ablation procedure more complicated.

Ovarian maturation follows a few days or weeks after ablation, and spawning may occur as quickly as 3 days after ablation. If ablation is done during the inter-molt, maturation and spawning will immediately follow. When ablation is during the early premolt, the females will first molt before they start maturing.

C. Sampling

First examination of broodstock in tanks is done 3 days after ablation. Thereafter, frequency of sampling is determined by the remaining number of early maturing (Stage II) females. Sampling in tanks is done at night or late in the afternoon when it is sufficiently dark. An underwater flashlight tied to a pole is held close to each prawn so that the light is perpendicular to the upper body portion where the ovary is located.

A maturation tank with 25-30 females (and around 20 males) can produce 4-7 million nauplii over 4-6 weeks for wild broodstock and over 10-12 weeks for pond broodstock.

Sampling in pens starts one week after ablation and continues every week thereafter. The sampling procedure starts when the divers release the inside net from its bottom anchor. The net is pulled to one side to form a pouch wherein the prawns are collected. During the weekly lifting, the nets are cleaned of barnacles, dead prawns, molted shells, and other debris. Holes are also repaired.

D. Processing of spawners, eggs, nauplii

Female with Stage III and IV ovaries are retrieved from the tank or pen and then prepared for spawning. Others with Stages I and II ovaries are left to mature further.

1. Spawners may be disinfected in 50 ppm formalin for 1 hour prior to transfer to the spawning tanks.

2. Conical spawning tanks (250-l capacity) (Fig. 7) are filled with 200 liters of newly prefiltered, chlorinated (5-10 ppm), and neutralized (with sodium thiosulfate) seawater. Addition of the chelating agent EDTA (ethylene dinitro-tetraacetic acid) to the spawning water at 5-10 g/m³ may also improve hatch rate.

3. Tanks should be provided with a cover and mild aeration so as not to disturb the spawner. Once the spent female is removed from the tank, aeration should be increased for better egg development. Salinity should range from 25 to 35 ppt.

4. For normal development, egg density in the spawning tanks should not be more than 2,500-3,000/l. This means a maximum of 500,000 to 600,000 eggs for 200 l, which is a suitable level for ablated females with an average of 300,000 eggs/spawning (section I, G). However, large wild spawners (5-6 pcs/kg; 150-250 g body weight each) that may yield more than one million eggs in a single spawning should be placed in wider spawning tanks with at least 400 l of seawater.



Fig. 7. Siphoning of eggs from 250-l spawning tank (left) to an egg washer; maturation tanks to the right.

5. The following morning, spawning tanks are checked for spawning. Although the presence of scum (*see* I, F) is a sign of spawning, the surest way is to take a water sample in a glass container and look for the eggs.

6. Females that do not spawn are returned to their respective tank or pen. If they remain at Stage III or IV for more than two consecutive nights without spawning, they may have the "white ovary" disease (section I, F). Spent females are either returned directly, or processed (length-weight measurements, tagging on the unablated eyestalk), if monitoring of individual females is required prior to return.

7. A random sample of approximately 200 eggs is obtained from the spawning tank, placed on a glass slide (Fig. 8) and examined under a compound or dissecting microscope. The eggs are classified according to the different types described in section I, G. If the proportion of good (type A₁) eggs is below 30-40%, the eggs are discarded by completely draining the spawning tank.

8. If at least 30% of eggs show normal development, the eggs are cleaned by first scooping out the scum and other spawning debris.

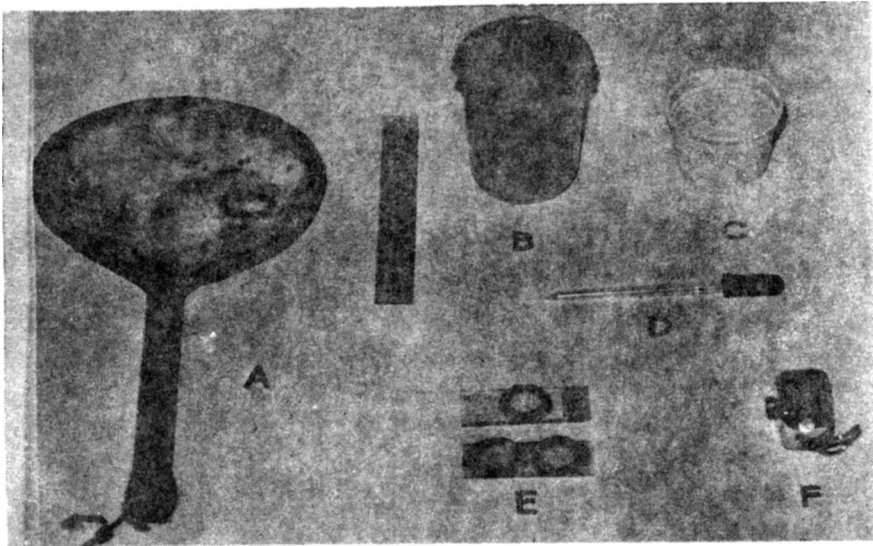


Fig. 8. Eggs are collected from spawning tank by means of flat net (A), then transferred to a 200-ml beaker (B) for counting using hand counter (F). If too many, eggs are subsampled in 50-ml plastic containers (C). Eggs are mounted in depression slides (2-mm deep wells) (E) and classified according to type (*see* Fig 5) under a microscope. Fine-tipped pipettes (D) are used to collect eggs.

Then the eggs are gently siphoned from the spawning tank into an egg washer with a series of two nets as follows (Fig. 7, 9):

Coarse net (0.35 mm mesh) — retains large dirt particles but allows eggs to pass through;

Fine net (0.25 mm mesh) — retains the eggs but allows finer particles to pass through.

Although the form of the egg washer may vary, the principle remains the same — the separation of eggs from both large and fine debris.

Always keep the eggs immersed in sea water (Fig. 9), and process gently to avoid mechanical damage. Water used for washing eggs should be chlorinated and EDTA-treated (*see 2 above*).

9. To prevent fungal infection, the eggs may be washed in 20 ppm laundry detergent for two hours and then thoroughly rinsed.

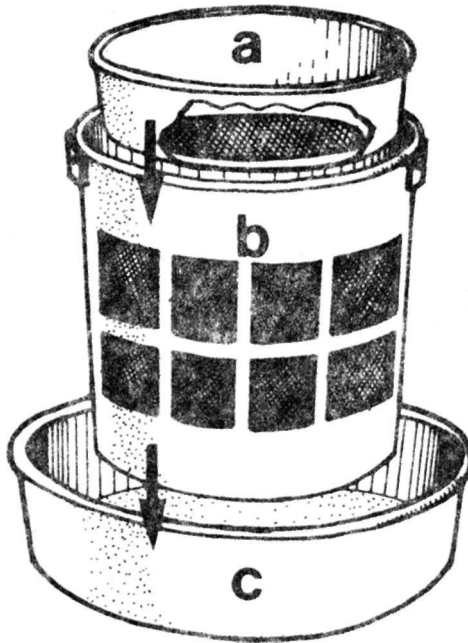


Fig. 9. Egg washer for *P. monodon* (arrows indicate flow of water). Container A (0.35 mm mesh) retains scum and large particles but allows eggs to pass through. Container B (0.25 mm mesh of windows) retains eggs but allows fine particles to pass through. Container C holds both A and B; it keeps eggs immersed in seawater.

10. When the eggs are cleaned, total egg count is estimated by taking three aliquot samples (200-500 ml) in a breaker (Fig. 9) after stirring the water in the spawning tank to ensure uniform distribution. The average egg count of the three samples converted to eggs/l is multiplied by 200 liters (volume of spawning tank) to give the estimated total number of eggs in each tank. The same procedure is used to estimate the number of larvae (nauplii). Nauplii are counted either at 4:00 p.m. of the same day for transfer to the larval rearing tanks as nauplius I or II (N_I , N_{II}), or in the morning of the next day as N_{III} , N_{IV} .

Percent hatch rate (HR) is computed as follows:

$$\% \text{ HR} = \frac{\text{total no. nauplii} \times 100}{\text{total no. eggs}}$$

11. To harvest larvae, aeration is stopped and the nauplii that gather at the surface (due to their positive phototaxis or response to light) are siphoned with a fine hose into a container for transfer to the larval rearing tank. With this procedure, weak nauplii that remain at the tank bottom are excluded.

E. Procurement and transport of nauplii

Where overland transport is too rough or stressful for wild spawners, and where hatcheries have an excess supply of larvae from ablated spawners, procurement and transport of nauplii may be resorted to.

The nauplii are packed in 20-l plastic bags filled with seawater at ambient temperature without oxygenation. Maximum density of nauplii is 20,000/l or 400,000/container for up to 4-5 hours overland transport. For longer transport by air, stocking should be lowered to 100,000 nauplii/container.

Transport of nauplii is effective because of a) the relative sturdiness of the nauplii, and b) the longer duration (1 ½ days) of the nauplius stage compared to the protozoa stage which requires feeding.

F. Feeds and feeding

A variety of fresh or fresh-frozen food should be given to prawn broodstock. A composite diet is better than a single-item diet. In

addition, pond-reared broodstock requires maturation pellet, unlike wild broodstock which can mature given fresh or frozen food alone (section I, A).

Broodstock feeds include squid, shrimp, mussel, trash fish, etc. Annelids or marine worms (locally sold as fish bait) are recommended because they are rich in certain polyunsaturated fatty acids predominantly found in mature ovaries of penaeids. Large quantities of food should be procured fresh and stored in a freezer at -8°C or lower.

Pellets (for maturation or grow-out) may also reduce the required amounts of fresh or frozen food especially if broodstock numbers are large. Since 1977, a maturation pellet (40-52% crude protein) has been recommended. Its present composition is as follows:

Ingredients	% in diet
Squid meal	30.00
Shrimp head meal	20.00
Fish meal	20.00
Cod-liver oil	6.00
Cholesterol	0.50
Wheat flour	5.50
Gulaman (<i>Gracilaria</i> spp.)	4.00
Rice bran	5.30
Vitamin mix	2.70
Mineral mix	6.00
BHT	0.03

Feeding in tanks consists of squid, mussel or annelids at 8:00-9:00 a.m. at 5% (wet weight) of biomass and pellets at 4:00-5:00 p.m. at 2% (dry weight) of biomass.

G. Mortality

Most deaths are primarily due to stress caused by molting, handling, disease, lack of food, etc. Cannibalism occurs only when one prawn is already weakened by any of the above factors. A prawn is in stress when it falls to its side and is unable to return to the normal upright position.

Weekly mortality rates range from 5 to 10% in tanks and pens depending on the presence of stress factors. Females have generally

higher mortality rates because they are exposed to greater stress due to ablation, spawning, and handling.

It is best to terminate or harvest a whole batch of broodstock and replace with a new one when female survival is less than 20% or when hatch rates of eggs consistently fall below 30-40%.

III. MATURATION TANK SYSTEM

A maturation system for *P. monodon* should provide the following environmental requirements: salinity of 28-35 ppt, temperature of 25-30°C, light intensity of 100 lux or less, and pH of 7.5-8.5.

The maturation pen prototype was tested by SEAFDEC AQD at its Igang, Guimaras and Batan, Aklan substations in the late 1970's. Site requirements include location in coves and other areas naturally protected from wind and wave action (Table 1).

Most commercial hatcheries that depend on broodstock use tanks in spite of the power requirement. The tank offers the advantages of more convenient and frequent retrieval of gravid females, easier maintenance, better security from poachers because it is located within the hatchery complex, minimal depreciation, and longer life span.

Majority if not all maturation tanks are integrated within a hatchery complex because of the power needed to supply seawater and aeration. This arrangement facilitates the immediate transfer of gravid females or nauplii to larval rearing tanks.

Freshwater is necessary for cleaning purposes and to lower salinity for acclimation of newly arrived broodstock.

A. Construction

Circular 4 m 0 x 1.25 m deep tanks (Fig. 10) are used for maintenance of prawn broodstock. Although wider and deeper tanks may be constructed, tanks of these dimensions are more convenient for sampling of the prawns. They also provide the minimum space required for maturation and mating of *P. monodon* (section I, D). Construction material may be of concrete hollow blocks, solid cement or concrete, ferrocement, marine plywood, or thick canvas lining with plywood support.

Table 1. Comparison of broodstock tank and pen systems for *Penaeus monodon*. (After J. H. Primavera and P. Gabasa. J. World Maricul. Soc. 12:345-356. 1981.)

	Land-based tank	Offshore pen
Site	power for 24-h flow-through	protected cove; water free from pollution
Dimensions; shape.	4 m 0 x 1.25 m; circular	16 x 16 x 4 m; rectangular
Area; volume	12 sq m; 12 cu m	250 sq m; 1,000 cu m
Total stock; sex ratio	50-7,0 at 1.5 female : 1 male	300 at 1-2 female : 1 male
Stocking density	4-6/sq m	1/sq m
Stocking sampling	nightly	weekly
Materials	concrete, wood, or canvas	bamboo and netting
Longevity	minimal depreciation	3-5 years

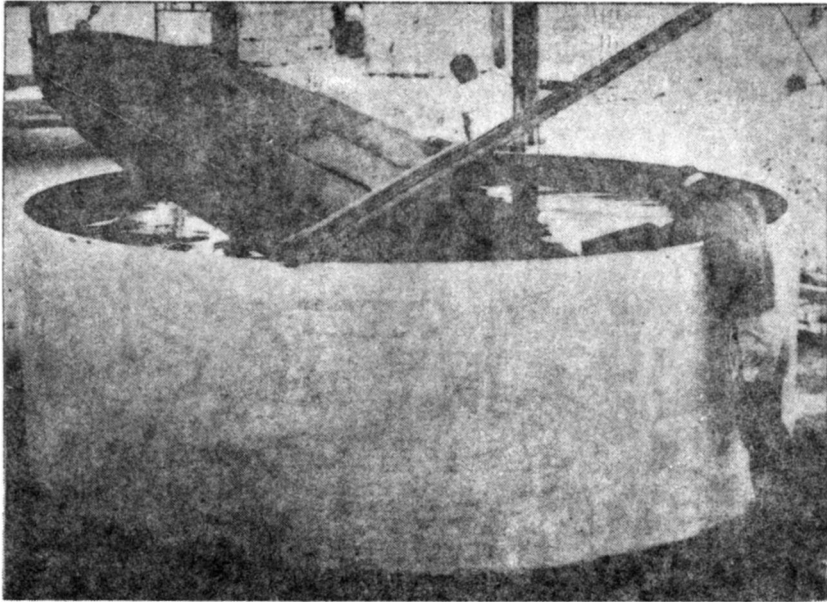


Fig. 10. A dark cover reduces light intensity and minimizes disturbance of prawn broodstock in a concrete maturation tank (12 m²).

The substrate consist of an upper layer of fine coralline sand (2-4 mm diameter) separated by a nylon screen from a lower layer of coarse (up to 2 cm) coralline material and stones; total thickness of substrate is 8-10 cm (Fig. 11). The prawns can be seen more easily against a white sand background as against dark sand. Some maturation tanks have a bare bottom without substrate with seawater introduced from the top.

Inner tank walls should be painted with nontoxic epoxy paint (black and other dark colors). The smooth surface is easier to clean and will not hold as many microorganisms as do porous concrete and other unpainted surfaces.

Tanks are generally located in a roofed structure with or without walls. A walled, completely enclosed building has a more stable air and, therefore, water temperature, particularly in the case of recirculating tanks. If the roof is a skylight without ceiling, light intensity can be reduced to approximately 100 lux by means of a dark cover (Fig. 10). The cover also minimizes disturbance of the broodstock.

An enclosed building with walls and ceiling will need an artificial light source such as one white, green or blue 40-watt fluorescent lamp

per tank. Wrap the lamp with black polyethylene netting material to reduce light intensity. Adjust photoperiod to 12-14 hours of light by means of an electronic timer.

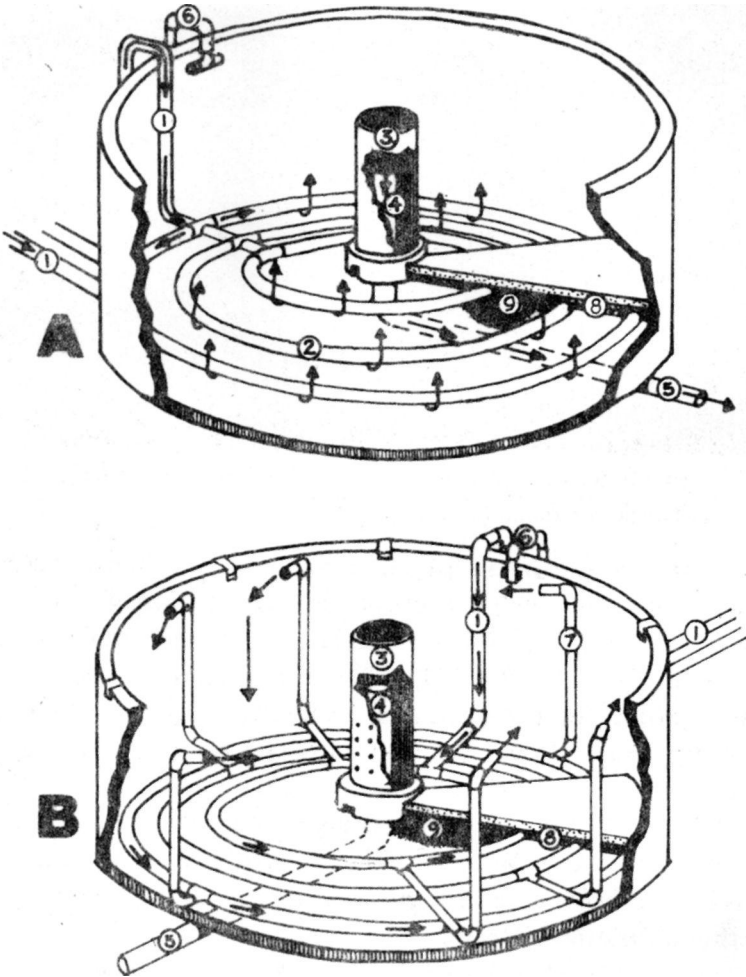


Fig. 11. Maturation tank, 4 m 0 x 1.25 m depth x 8-10 cm thick, for prawn broodstock with flow-through (A) or recirculating (B) water system. (1) Seawater line, 5 cm PVC pipe, (2) 3.2 cm PVC pipe, (3) 20 cm PVC pipe (central standpipe for drainage), (4) 10 cm PVC pipe (for drainage), (5) 10 cm outlet pipe, (6) aeration line, 5 cm PVC pipe (7) air-water lift, 5 cm PVC pipe, (8) rock and coralline sand substrate, (9) nylon netting. Arrows indicate flow of water. Tank may be made of concrete (hollow blocks or solid) or canvas-lined plywood.

B. Water management

The water supply system consists of concentric perforated (0.3-0.6 cm diameter holes) PVC pipes embedded in the lower stone layer (Fig. 11). Seawater let in through a vertical pipe is distributed through the concentric pipes and flows upwards through the coralline substrate. Water is drained through a double cylinder standpipe located at the center of the tank. This 24-hour flow-through system allows a daily exchange rate of 200-400% of the total water volume in the tank. Three to five airstones are provided in the event of pump breakdown.

Flow-through water is ideal but where seawater supply is limited, recirculation is necessary. Water can be recirculated through an external biomechanical filter or through a sand filter inside the tank itself using an air-water-lift system (Fig. 11). Still a further simplification is the use of aerators (6-10 airstones/tank) instead of the flow-through or recirculating system. Water is then replaced daily at 25-30% of total volume at the same time that dirt and excess feeds are siphoned out.

During typhoons and heavy rains when incoming water may be turbid, the water supply should be closed and aeration provided, otherwise silt will accumulate on the tank bottom. A combination of silty water and air failure in the tank can cause mass mortality of the broodstock in a very short time.

Even with a constant flow-through and regular siphoning of debris and excess food, the tank will tend to accumulate dirt over a long period. When the sand substrate turns dark brown, it should be disinfected with 50-200 ppm formalin for 12-24 hours followed by thorough rinsing. Sponges and other encrusting forms of marine fauna and flora may grow on the tank walls and sand substrate; they can be left alone if they cause no harm.

C. Construction and operating costs

Construction costs will depend on type of materials used. For example, concrete hollow blocks cost less than ferrocement or solid concrete.

Major operating costs will be for broodstock procurement and feeds (15 kg squid or mussel + 0.5 kg marine worms + 2-3 kg pellets/month/tank of 30 females, 20 males). Power and labor costs are in-

with a separate maturation unit of 10 tanks or more may need full-time personnel for the maturation tanks.

Miscellaneous costs include chemicals, nets, brushes, buckets, basins, and other materials.

IV. SUGGESTED READINGS

- Adiyodi, K.G. and R.G. Adiyodi. 1970. Endocrine control of reproduction in decapod Crustacea. *Biol. Rev.* 45:121-165.
- Alikunhi, K.H., S. Poernomo Am Adisukresno, M. Budiono, and S. Busman. 1975. Preliminary observation on induction of maturity and spawning in *Penaeus monodon* Fab. and *Penaeus merguensis* de Man by eyestalk extirpation. *Bull. Shrimp Cult. Res. Cent. Jepara* 1:1-11.
- Aquacop. 1977. Reproduction in captivity and growth of *Penaeus monodon* Fabricius in Polynesia. *Proc. World Maricul. Soc.* 8:927-945.
- Aquacop. 1982. Reared broodstock of *Penaeus monodon*. *Proc. Symp. Coastal Aquaculture* 1:55-62.
- Aquacop. 1983. Constitution of broodstock, maturation, spawning and hatching systems for penaeid shrimps in C.O.P. In: J.P. McVey (ed.), *CRC Handbook of Mariculture, Vol. 1, Crustacean Aquaculture*, pp. 105-122. CRC Press, Florida.
- Beard, T.W. and J.F. Wickins. 1979. The breeding of *Penaeus monodon* Fabricius in laboratory recirculation systems. *Aquaculture* 20:78-89.
- Clark, W.H., Jr., J.W. Lynn, A.I. Yudin, and H.O. Persyn. 1982. Morphology of the cortical reaction in the eggs of *Penaeus aztecus*. *Biol. Bull.* 158:175-186.
- Crocos, P.J. and J.D. Kerr. 1983. Maturation and spawning of the banana prawn *Penaeus merguensis* de Man (Crustacea:Penaeidae) in the Gulf of Carpentaria, Australia. *J. Exp. Mar. Biol. Ecol.* 69:37-59.
- Emmerson, W.D. 1983. Maturation and growth of ablated and unablated *Penaeus monodon* Fabricius. *Aquaculture* 32:235-241.
- Hiller, A.G. 1984. Artificial conditions influencing the maturation and spawning of subadult *Penaeus monodon* (Fabricius). *Aquaculture* 36:179-184.
- Lio-Po, G. and M.E.G. Sanvictores. 1986. Tolerance of *Penaeus monodon* eggs and larvae to fungicide against *Lagenidium* spp. and *Haliphthoros philippinensis*. *Aquaculture* 51:161-168.
- Middleditch, B.S., S.R. Missler, D.G. Ward, J.P. McVey, A. Brown, and A.L. Lawrence. 1979. Maturation of penaeid shrimp: Dietary fatty acids. *Proc. World Maricul. Soc.* 10:472-476.

- Millamena, O.M., J.H. Primavera, R.A. Pudadera, and R.V. Caballero. 1986. The effect of diet on the reproductive performance of pond-reared *Penaeus monodon* Fabricius broodstock, pp. 593-596. In: J.L. Maclean, L.B. Dizon, and L.V. Hosillos (eds.). The First Asian Fisheries Forum. Asian Fisheries Society, Manila, Philippines.
- Motoh, H. 1981. Studies on the fisheries biology of the giant tiger prawn, *Penaeus monodon* in the Philippines. Technical Report No. 7, SEAFDEC Aquaculture Dept., Iloilo, Philippines, 128 p.
- Posadas, R.A. 1986. The effect of salinity on the maturation and spawning of ablated *Penaeus monodon* Fabricius. M.S. Thesis, University of the Philippines in the Visayas, 50 p.
- Primavera, J.H. 1978. Induced maturation and spawning in five-month old *Penaeus monodon* Fabricius by eyestalk ablation. *Aquaculture* 13:355-359.
- Primavera, J.H. 1979. Notes on the courtship and mating behavior in *Penaeus monodon* Fabricius. *Crustaceana* 37:287-292.
- Primavera, J.H. 1985. A review of maturation and reproduction in closed thelycum penaeids, pp. 47-64. In: Y. Taki, J.H. Primavera, and J.A. Llobrera (eds.). Proc. First Intl. Conference on the Culture of Penaeid Prawns/Shrimps. SEAFDEC Aquaculture Dept., Iloilo, Philippines.
- Primavera, J.H. and R.A. Posadas. 1981. Studies on the egg quality of *Penaeus monodon* Fabricius based on morphology and hatching rates. *Aquaculture* 22:269-277.
- Primavera, J.H., E. Borlongan, and R.A. Posadas. 1978. Mass production in concrete tanks of sugpo *Penaeus monodon* Fabricius spawners by eyestalk ablation. *Fish Res. J. Philipp.* 3:1-12.
- Primavera, J.H., C. Lim, and E. Borlongan. 1979. Feeding regimes in relation to reproduction and survival of ablated *Penaeus monodon*. *Kalikasan, Philipp. J. Biol.* 8:227-235.
- Primavera, J.H. and P. Gabasa. 1981. A comparison of two prawn (*Penaeus monodon*) broodstock systems - land-based tanks and marine pens. *J. World Maricul. Soc.* 12:345-356.
- Pudadera, R.A., J.H. Primavera, and E. Borlongan. 1980. Effect of different substrate types on fecundity and nauplii production of ablated *Penaeus monodon* Fabricius. *Philipp. J. Sci.* 109-15-18, 44.
- Pudadera, R.A., J.H. Primavera, and T. Young. 1980. Effect of different sex ratios on maturation, fecundity and hatching rates of ablated wild stock *Penaeus monodon* Fabricius. *Fish. Res. J. Philipp.* 5:1-6.
- Pudadera, R.A. and J.H. Primavera. 1981. Effect of light quality and eyestalk ablation on ovarian maturation in *Penaeus monodon*. *Kalikasan, Philipp. J. Biol.* 10:231-240.

- Reyes, E.P. 1981. The effect of temperature and salinity on the hatching of eggs and larval development of sugpo, *Penaeus monodon* Fabricius. M.S. Thesis, University of the Philippines, 42 p.
- Santiago, A.C., Jr. 1977. Successful spawning of cultured *Penaeus monodon* Fabricius after eyestalk ablation. *Aquaculture* 11:185-196.
- Tolosa, R. 1978. Notes on the construction of a 12 cu m ferrocement maturation tank for prawn broodstock. *J. Ferrocement* 8:93-103.
- Villaluz, D.K., A. Villaluz, B. Ladrera, M. Sheik, and A. Gonzaga. 1972. Reproduction, larval development and cultivation of sugpo (*Penaeus monodon* Fabricius). *Philipp. J. Sci.* 98:205-234.

BROODSTOCK OF SUGPO (*Penaeus monodon* Fabricius)
Aquaculture Extension Manual No. 7 ISSN 0115-5369
Fourth Edition, May 1989



AQUACULTURE DEPARTMENT
SOUTHEAST ASIAN FISHERIES DEVELOPMENT CENTER