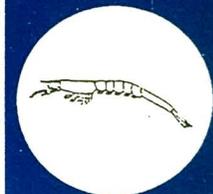
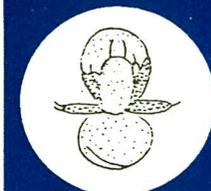


AQUACULTURE EXTENSION MANUAL NO. 19
FEBRUARY 1991



PRAWN HATCHERY OPERATIONS

**F.D. PARADO-ESTEPA
E.T. QUINTIO
E.L. BORLONGAN**



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SOUTHEAST ASIAN FISHERIES DEVELOPMENT CENTER
Tigbauan, Iloilo, Philippines

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PREFACE

Cognizant of the ever increasing need for fry for the prawn grow-out industry, SEAFDEC/AQD published in 1978 its first manual entitled "Design, Operation, and Economics of Small-scale Hatchery for Larval Rearing of Suppo *Penaeus monodon* Fabricius." As a result of continuing research on the simplification of prawn hatchery operations, a second manual, "Prawn Hatchery Design and Operation," was published in 1984. Since then, various hatchery techniques have been developed and practiced.

This updated version presents the underlying principles and step-by-step instructions on prawn larval and postlarval rearing. The terms used have been simplified for easy understanding. A Glossary of Terms has also been included for further reference. Mention of a company or product in this manual does not mean endorsement by the authors or SEAFDEC/AQD.

With few modifications, the techniques described here are not only applicable to *Penaeus monodon* but also to other penaeid and metapenaeid species [e.g., "hipongputi" (*Penaeus indicus* or *P. merguensis*), *P. japonicus*, *P. latisulcatus*, *P. semisulcatus*, and "suahe" (*Metapenaeus ensis*)]. We hope this manual will benefit present and prospective private investors, hatchery operators, technicians, teachers, and students.

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INTRODUCTION

Hatchery, as an initial phase in prawn culture, has become an indispensable step to meet the growing fry requirement of the industry. The first larval rearing techniques were adopted from the Japanese method utilizing large tanks (Shigueno 1975). The use of small tanks was later introduced following the Galveston method (Mock & Murphy 1971). Through the years, considerable progress in the development of a prawn hatchery technology has been achieved. A much improved and simplified technology which could be easily adopted by prawn growers was developed at SEAFDEC/AQD and is described in detail in this manual. The techniques may be modified depending on the problems encountered in a specific site.

SITE SELECTION

A prawn hatchery should be constructed on a suitable site. Several criteria should be considered.

Seawater Supply

The hatchery should be located near sandy and rocky or coralline shores where clean and clear seawater can be pumped easily and economically. It should be far from mouths of rivers and streams where flowing freshwater or brackishwater can abruptly lower salinity. It must also be far from possible sources of pollution like industrial, agricultural, and domestic discharges. The seawater near the hatchery site must exhibit only slight fluctuations in temperature, salinity, pH, dissolved oxygen, ammonia, and nitrite. These physico-chemical parameters must be within the ranges recommended in Table 1.

Table 1. Water quality parameters suitable for prawn hatchery

Parameter	Range
Temperature	27-30°C
Salinity	30-36 ppt
pH	7.5-8.5
Dissolved oxygen	>5 ppm
Unionized ammonia (NH ₃)	<0.1 ppm
Nitrite (NO ₂ -N)	<0.02 ppm

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Spawner and Broodstock Source

The source of spawners and broodstock should be identified before putting up a hatchery. Proximity to the source will minimize stress and expenditure in the transport of spawners and broodstock.

Availability of Electric Power

The hatchery should be located in areas where there is a reliable source of electric power. This is needed to run equipment and other life support systems in the hatchery.

Accessibility

Good roads near the hatchery will facilitate procurement of materials necessary for operations. Handling stress and transport expenses will be minimized during spawner procurement and disposal of fry if the market is near. Air transportation should be available when the market is far.

Freshwater Supply

Freshwater should be available in the hatchery because it is necessary for washing and rinsing of materials and tanks.

LIFE CYCLE

Familiarization with the biology of the species to be cultured is very important for hatchery management because it affects the program of daily activities. The life cycle of the giant tiger prawn, *Penaeus monodon*, in its natural habitat is shown in Fig. 1. The eggs are demersal and tend to sink while larvae are planktonic. Prawn larva thrives mainly offshore and undergoes three main stages: nauplius, protozoa, and mysis. At the postlarval and juvenile stages, the prawn migrates towards the estuary. As it grows, it starts moving to the shallow coastal waters. The adult prawn inhabits the open sea.

Sexes are separate and can be easily distinguished through the external genitalia located at the ventral side (Fig. 2): the thelycum in females and petasma in males. During mating, the male deposits the spermatophore inside the thelycum of the female. Mating can only occur between newly molted females and hard-shelled males (Motoh 1981). Spawning takes place throughout the year. The eggs are fertilized in the

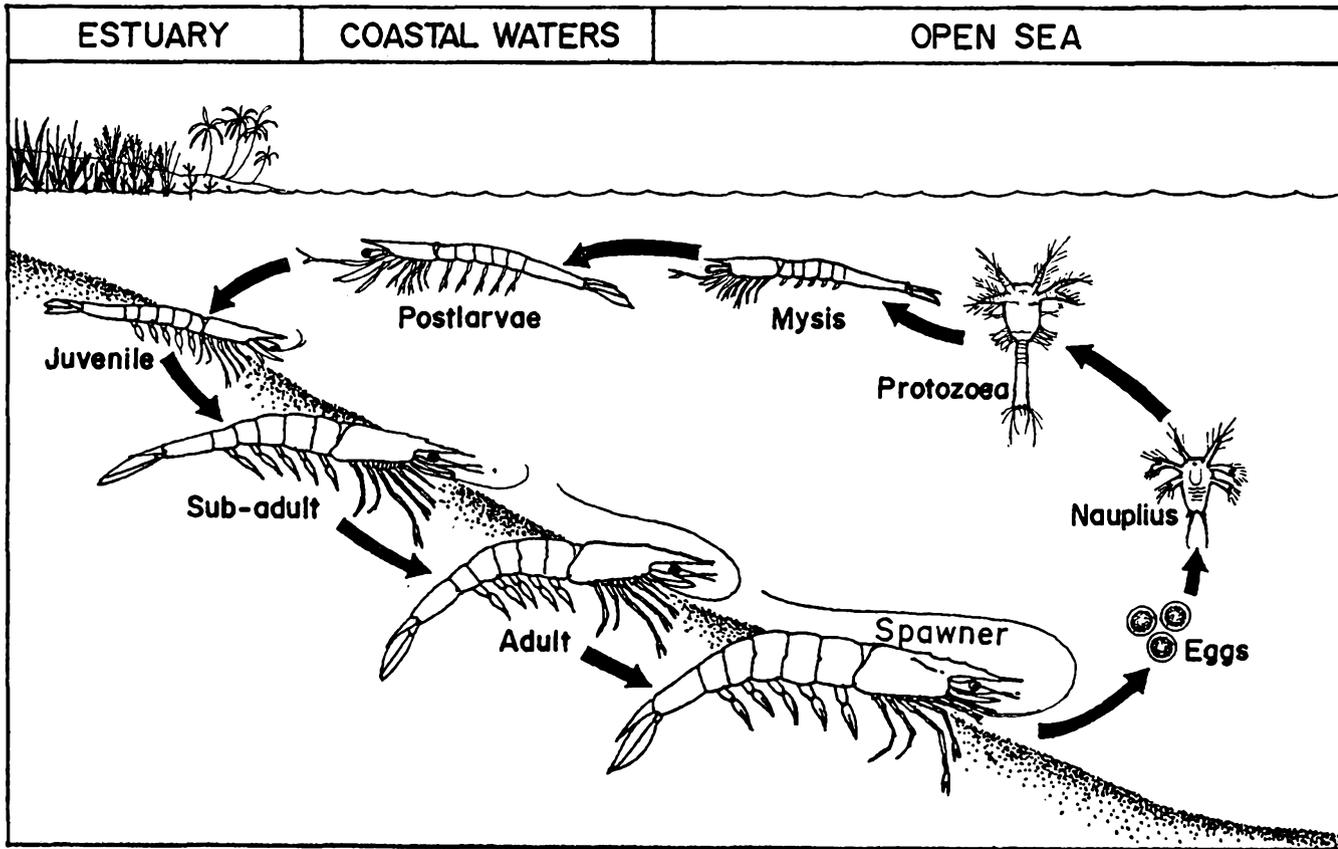


Fig. 1. Life cycle of sugpo, *Penaeus monodon* (Modified from Motoh 1981).

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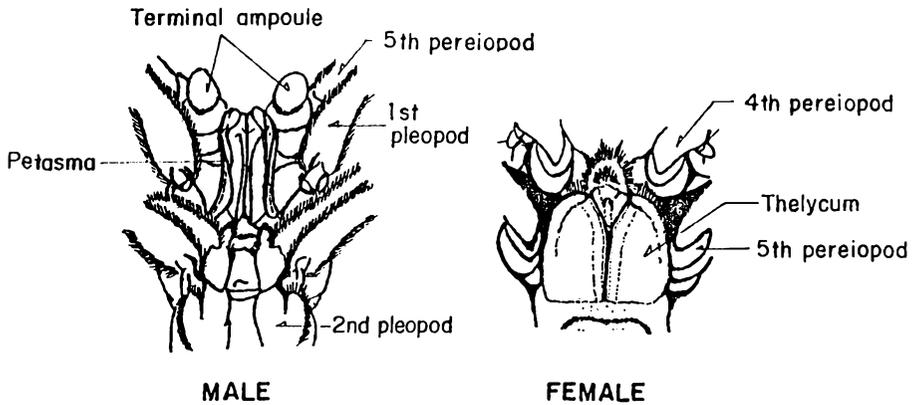


Fig. 2. External genitalia of male and female *P. monodon* (Motoh 1981).

water after the female simultaneously extrudes the eggs and the spermatophore. The number of eggs released by a single spawner varies from 248 000 to 811 000 (Motoh 1981). The biology of this species is described in detail by Motoh (1981) and Solis (1988).

Eggs

The eggs are small, spherical, and vary from 0.25 to 0.27 mm in diameter (Fig. 3). The developing nauplius almost fills up the entire space inside the egg. At 28-30°C, the eggs hatch 12-17 h after spawning.

Nauplius Stage

The nauplius stage is the stage after eggs have hatched. The prawn nauplius is very tiny, measuring from 0.30 to 0.58 mm in total length (Fig. 4). It swims intermittently upward using its appendages in a "bat-like" manner. It is attracted to light; and, in aerated tanks, it will concentrate in the most lighted areas if aeration is stopped. The nauplius molts through each of six substages for a total of about 1.5-2 days. The substages differ from each other mainly on the furcal spine formula. The latter indicates the number of spines at each side of the furca (Fig. 4).

Protozoa Stage

The protozoa can easily be distinguished from the nauplius stage. Its body is more elongated and measures from 0.96 to 3.30 mm in total length (Fig. 5). It consists of the carapace, thorax, and abdomen. The protozoa can also be distinguished by its movement; it swims vertically and diagonally forward towards the water surface.

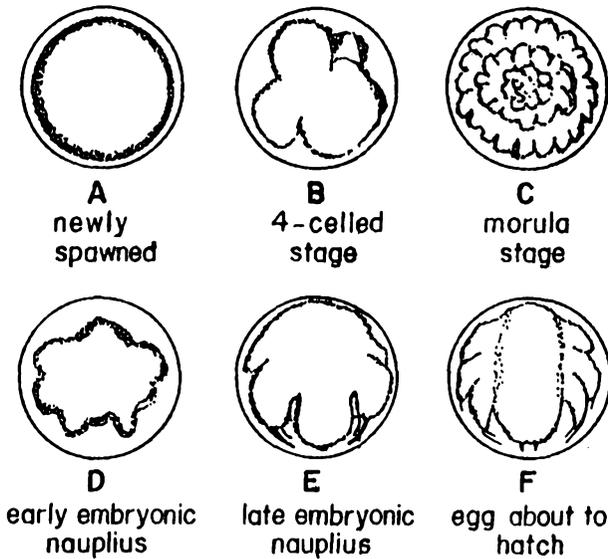


Fig. 3. Egg development of *P. monodon* (Motoh 1981).

The protozoa undergoes three substages (Fig. 5). The paired eyes of protozoa I (Z_1) can be observed as two dark spots in the upper portion of the carapace when examined under the microscope. These eyes become stalked at protozoa II (Z_{II}). At protozoa III (Z_{III}), the dorsal median spine at the sixth abdominal segment first appears. Observations on the increase in size from Z_1 to Z_{III} during the actual rearing activity can also help in the visual identification of the substages.

Mysis Stage

The mysis is shrimp-like with the head pointing downward. Its body measures from 3.28 to 4.87 mm in total length (Fig. 6). The telson and uropods are developed. The mysis swims in quick darts accomplished by bending the abdomen backwards. For mysis substages, the most prominent change is the development of pleopods. The pleopods appear as buds at mysis I (M_I) which protrude at mysis II (M_{II}), and finally become segmented at mysis III (M_{III}).

Postlarval Stage

The postlarva resembles an adult prawn (Fig. 6). At postlarva 1 (PL_1), the rostrum is straight and exceeds the tip of the eye. It usually has one dorsal rostral spine without any ventral spine. Plumose hairs are present

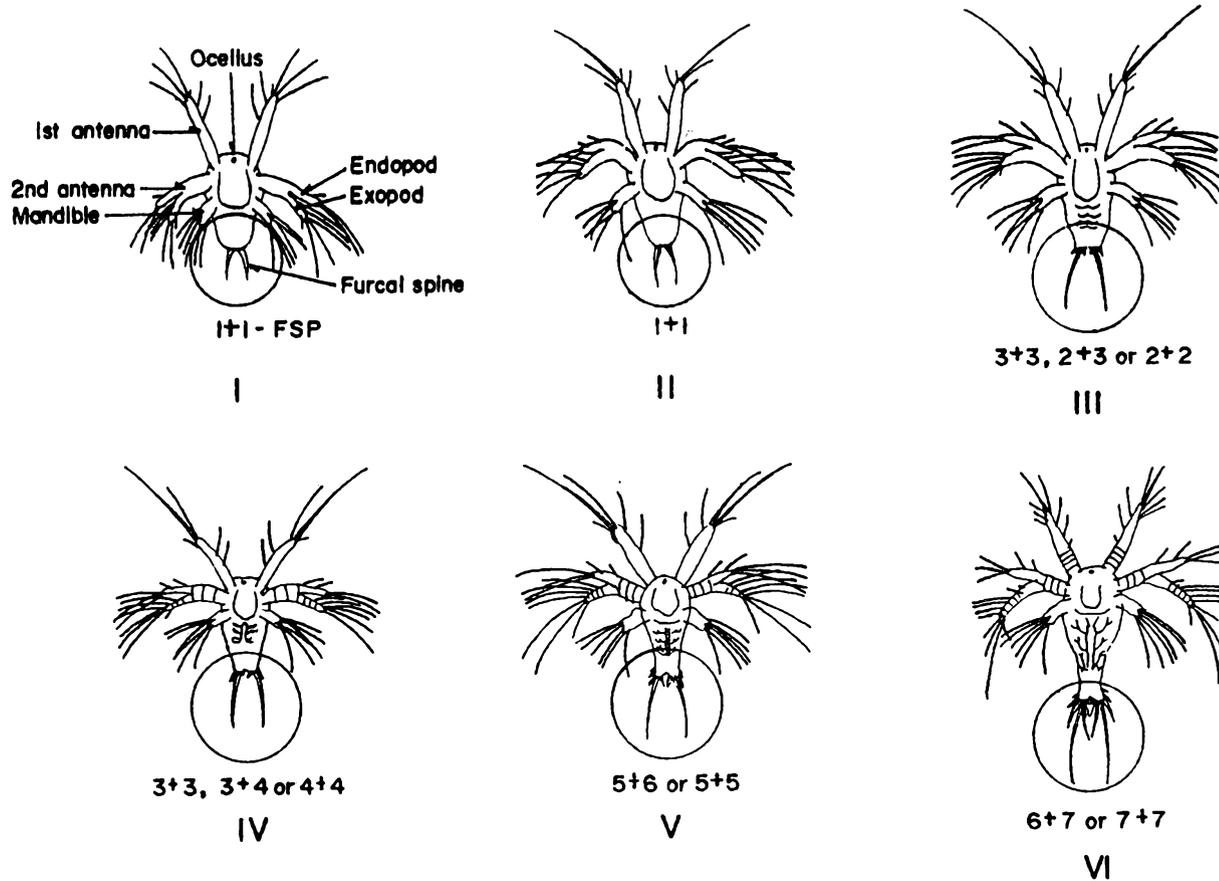


Fig. 4. Nauplius substages of *P. monodon* (Motoh 1981). The furcal spine formula (FSP) below each substage and the encircled portions indicate the distinct morphological changes that differentiate one substage from another.

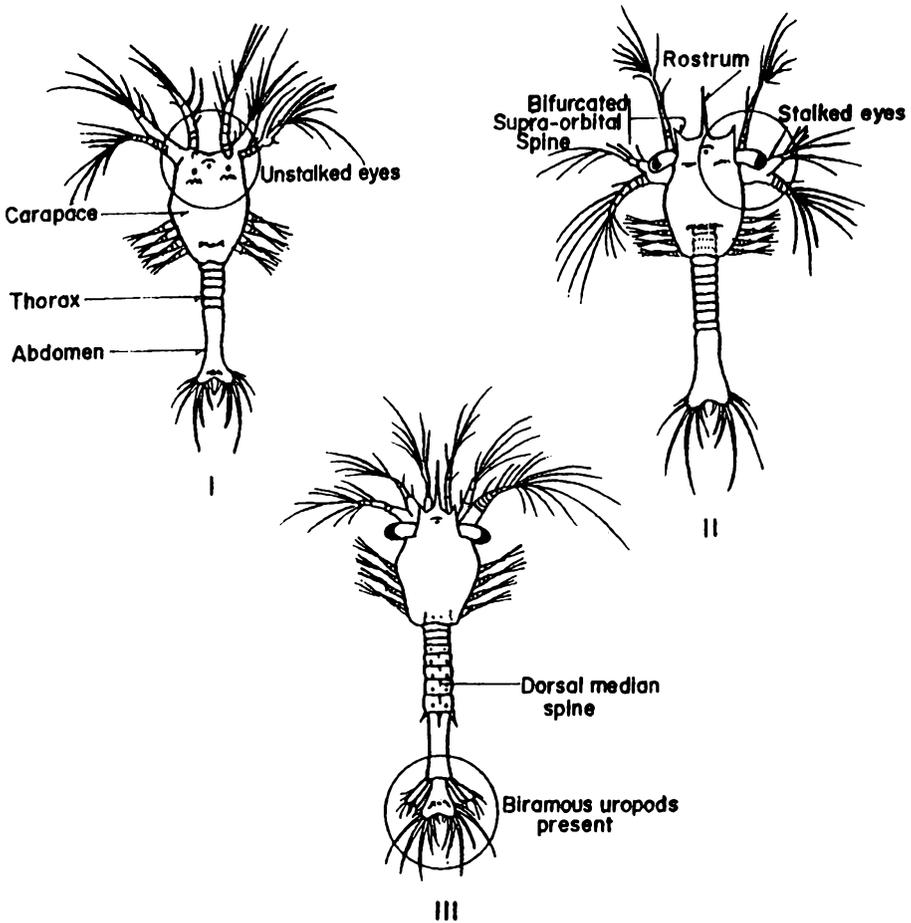


Fig. 5. Protozoal substages of *P. monodon* (Motoh 1981).

on the swimming legs. The number of days from this stage corresponds to the age of postlarva (e.g., PL₂ means second day after they have molted to postlarval stage).

HATCHERY DESIGN AND PLANNING

Size

The size of a prawn hatchery depends on two factors: financial capability and target production. For every million PL₁₅₋₂₀, a total effective

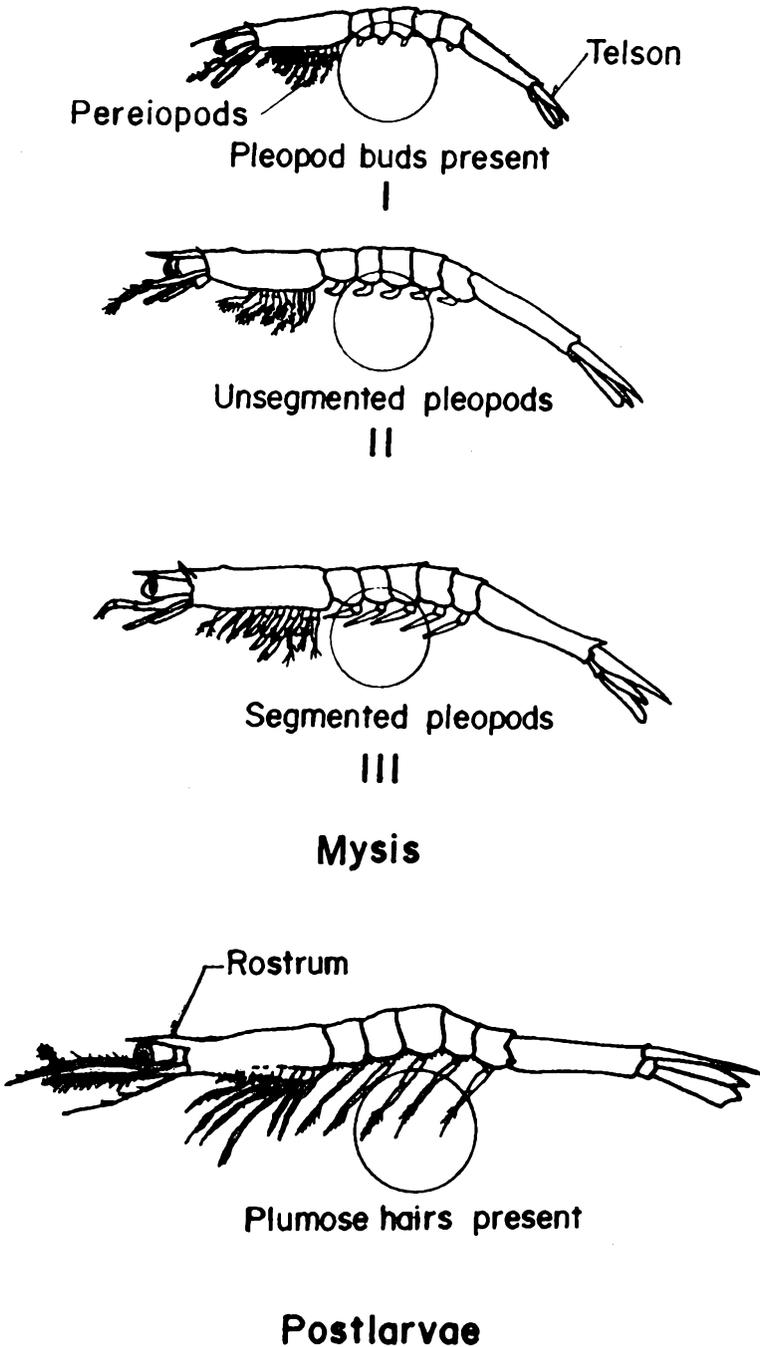


Fig. 6. Mysis substages and postlarval stage (Motoh 1981).

tank volume of 200 t for culturing larvae and postlarvae is required. This is based on the following assumptions: a) survival rate is 40% from nauplius to the early postlarval stage (PL_{1,5}) and 50% at nursery phase (PL_{15,20}); and b) stocking densities are 50 000 to 100 000 nauplii/t and 10 000 PL_{1,5}/t. This computation assumes that the larval tanks are also used as nursery tanks.

Facilities and Equipment

Larval and Postlarval Tanks. Containers used for culturing larvae and postlarvae may be of rubberized canvas, marine plywood, fiberglass, or concrete. These can either be circular, oval, or rectangular, depending on the operator's preference or financial capability. The capacity of each tank may be from 1-20 t but 10-12-t tanks are more economical and practical. Depth should only be about 1 m because tanks which are too deep are difficult to manage.

Algal Tanks. Minute plants (phytoplankton) are needed as food for the early life stages of prawn. Daily procurement of algae directly from laboratories is expensive and impractical. Thus, a hatchery must have tanks where these food organisms can be cultured in large quantities.

Algal tanks must be shallow (ideally 0.5-m deep) to allow sufficient light penetration. A shed, not necessarily enclosed with walls, must be provided with transparent roofing to prevent contamination and dilution of the culture by rain while allowing light to pass through. Algal tanks must also be provided with screen covers during the night to prevent insects from getting into the culture.

The number and volume of algal tanks to be constructed depend on the daily algal requirement. This is usually 10-20% of the total volume of larval tanks if diatoms such as *Chaetoceros* or *Skeletonema* are to be cultured. Additional algal tanks are needed if *Tetraselmis*, a species of green algae, or other slow-growing species are to be cultured.

Spawning Tanks. Although spawners are usually placed in the larval tanks prior to spawning, it is advantageous to have smaller tanks with volumes ranging from 0.25 to 1 t where egg washing is done. Tapered bottoms are preferable since these allow homogeneous aeration necessary for hatching.

Artemia Hatching Tanks. *Artemia* or brine shrimp is a protein-rich live food organism given to prawn larvae starting at the mysis stage. *Artemia* is available in cyst form which has to be hydrated and incubated in tanks for at least 18-24 h. These tanks should preferably be of transparent material with a conical bottom so as to facilitate hatching of cysts and separation of cyst shells from the *Artemia* nauplii.

Reservoir. A reservoir or storage tank is necessary for chlorination and holding of filtered and treated water for daily use. This must have a

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total capacity of at least 50% of total larval tank volume. However, it is more convenient to have two storage tanks so that one may be cleaned and dried while the other is in use. An elevated storage tank that can distribute seawater to other tanks by gravity flow is advantageous.

Aeration System. Aeration is necessary in hatchery operations to keep food particles and algal cells in suspension and to maintain sufficient dissolved oxygen levels. Generally, aeration is supplied by a rotary blower (Fig. 7A). Since a rotary blower supplies a large volume of low pressure air, the depth of larval or algal tanks should not exceed 2 m. Another source of aeration is an air compressor (Fig. 7B) but this tends to emit oil and grease which may pollute the water in the tanks. It is also advantageous, especially

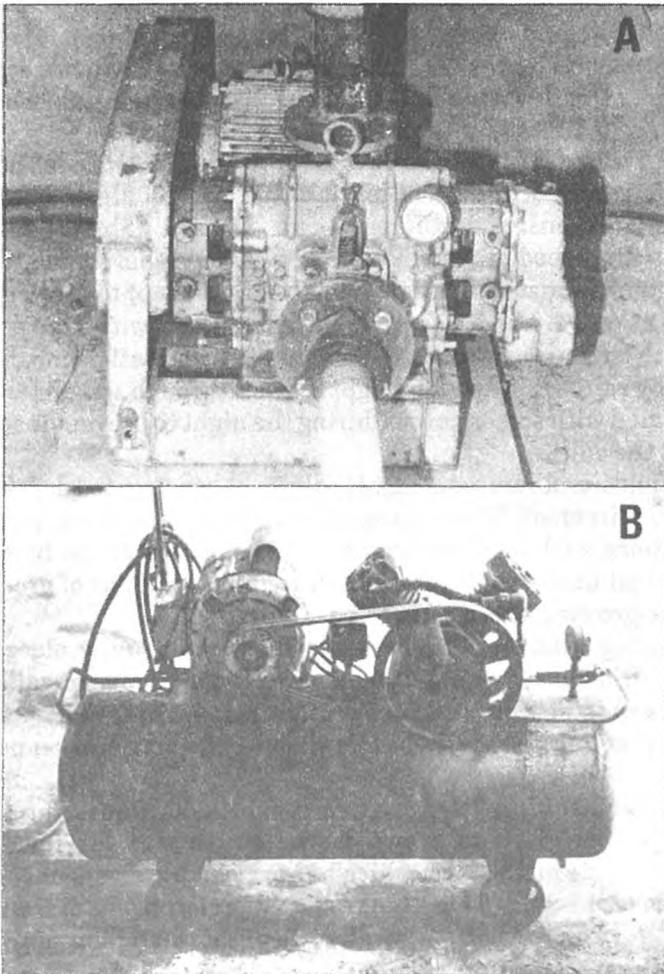


Fig. 7. Aeration equipment commonly used: rotary blower (A) and air compressor (B).

for large hatcheries, to install several units of lower capacity blowers (instead of one unit of high capacity) so that some units can be put off when not in use. Hatcheries with very small capacities (backyard type) may also use battery-operated aquarium aerators.

Continuous aeration is essential during operations. A stand-by generator will be very useful during power interruptions.

Seawater System. Seawater may be pumped directly from the sea or through a sump pit (Fig. 8). Water may be prefiltered through the sand in the sea bed or directly pumped to the hatchery. Commonly used prefiltration systems are shown in Fig. 9.

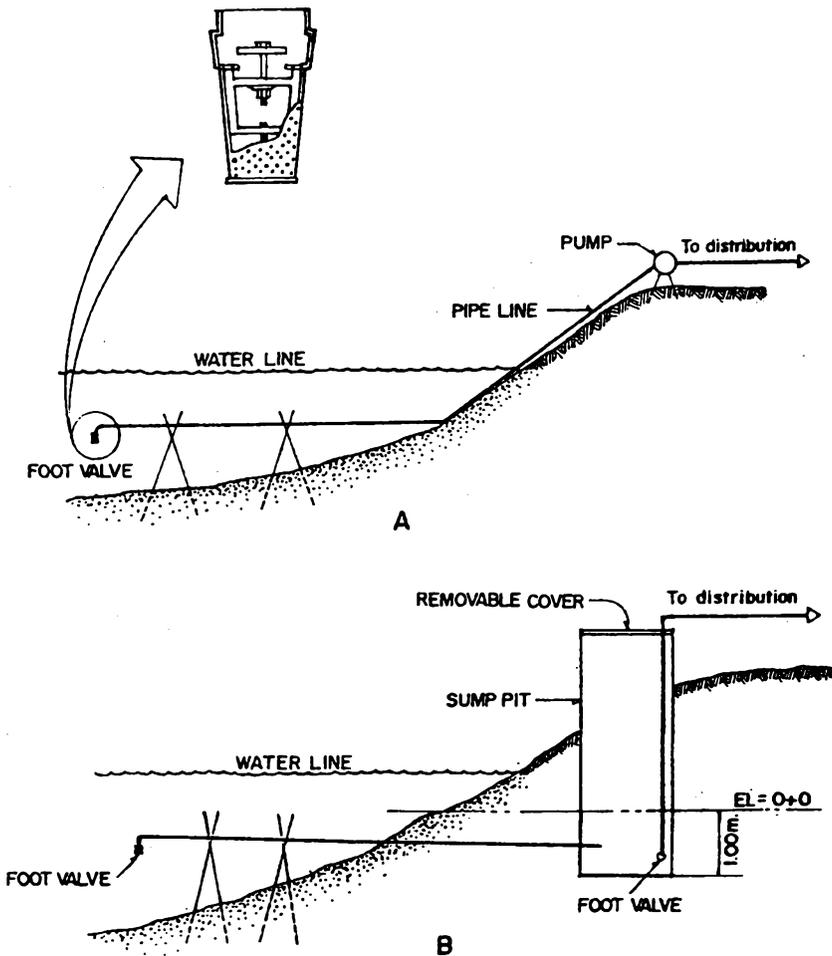


Fig. 8. Seawater intake systems: direct pumping (A) and sump pit (B) (SEAFDEC 1984).

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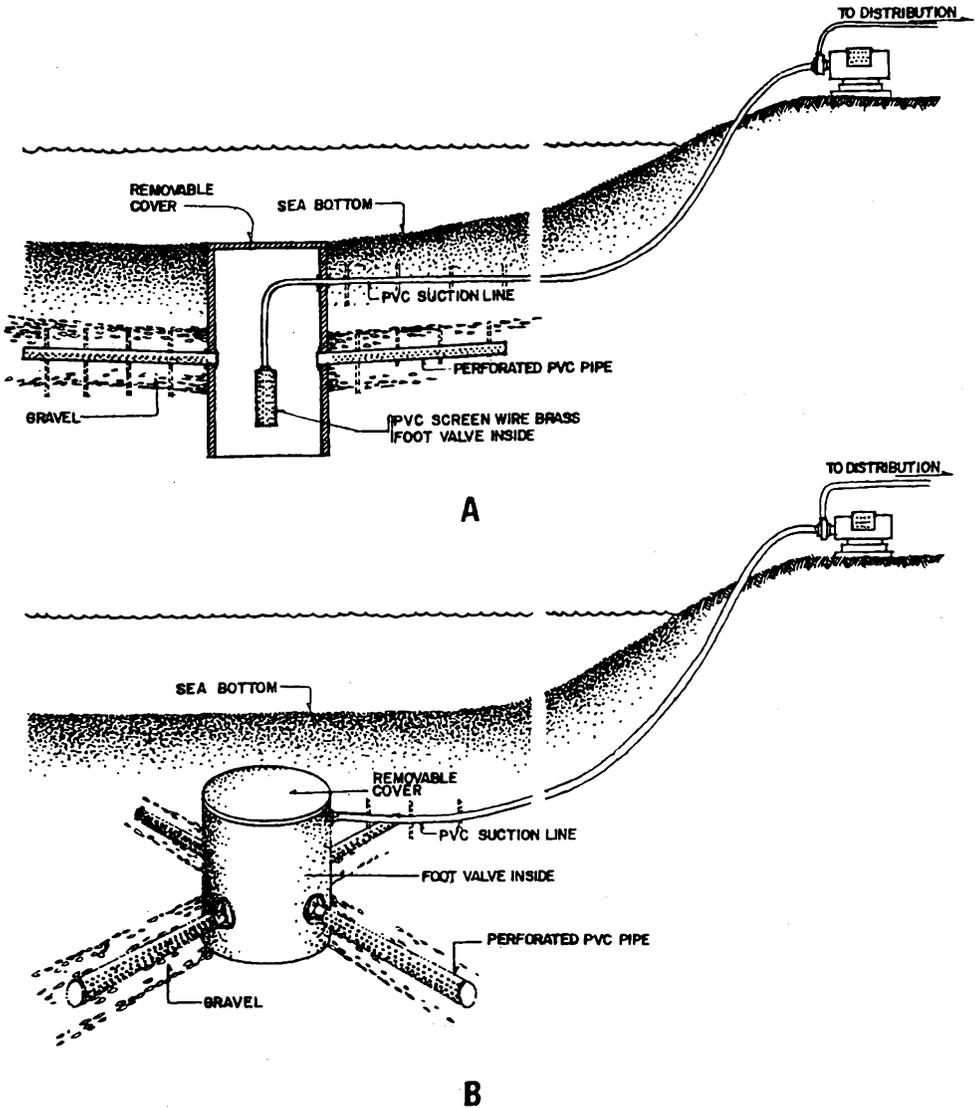


Fig. 9. Commonly used prefiltration systems: graded gravel (A) or concrete suction chamber with perforated PVC pipes (B).

Before the water is stored in a reservoir, it is passed through a sand filter which is usually elevated. The sand filter is made of graded gravel and sand which screen out particulate matter (Fig. 10).

Pumps. Water direct from the sea or from the reservoir is transferred to the larval tanks by either centrifugal or submersible pumps. The type and size of pump depends on the total volume of water required per day and the maximum pumping time. An engineer should be consulted regarding pump capacities.

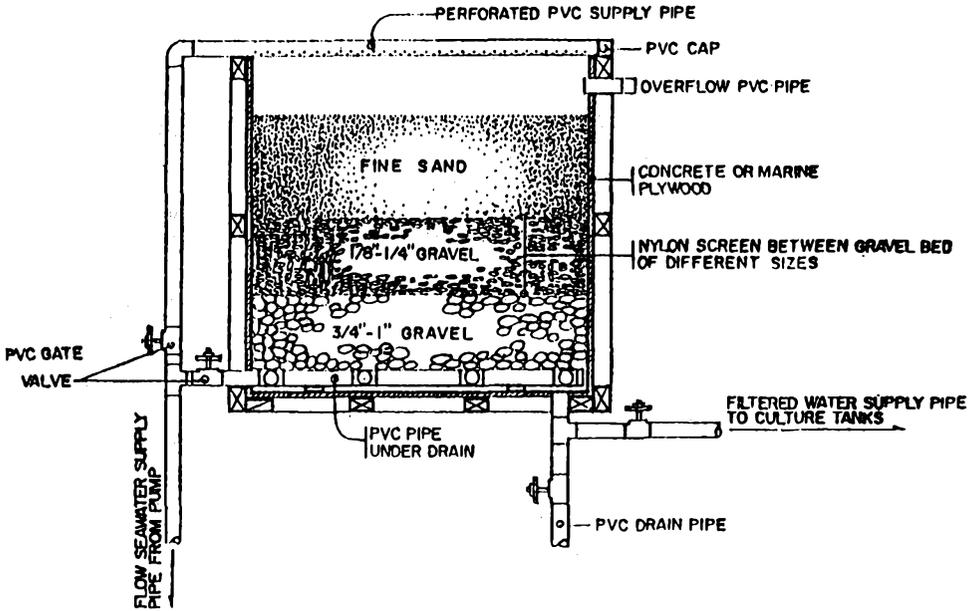


Fig. 10. Cross-section of a sand and gravel filtration unit.

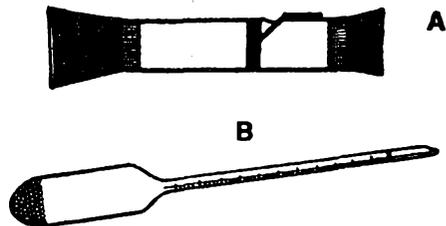
Layout

A sample layout of a shrimp hatchery is shown in Fig. 11. The algal tanks are constructed near the larval tanks for ease in feeding. If rectangular tanks are to be constructed, two tanks may share a common side but these should not be too long for the middle portion of the tank to be unreachable. There should also be sufficient space around the tanks for easier management.

Other Equipment and Accessories

Refrigerator. This is for storage of feeds, chemicals, and algal culture stock.

Refractometer (A) or hydrometer (B). This is for determining salinity of rearing water. A refractometer measures salinity directly. If a hydrometer is used, determine the temperature of the water and refer to density of water at this temperature before converting the measured reading to salinity.



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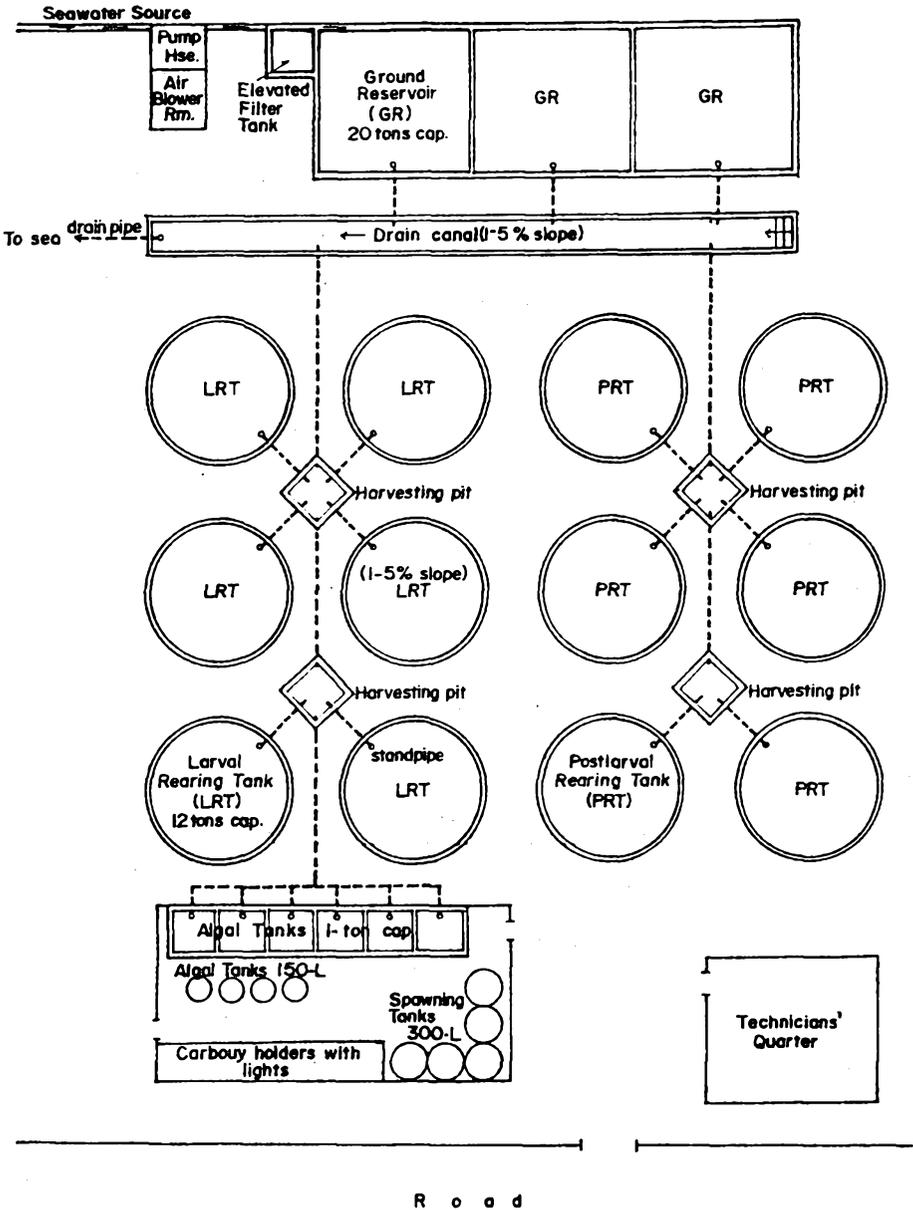
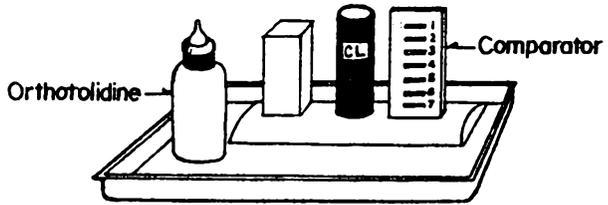


Fig. 11. A sample layout of a prawn hatchery.

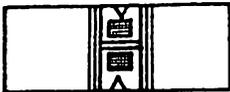
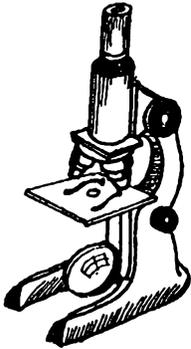
Thermometer. This is used to get temperature readings, especially before water change.



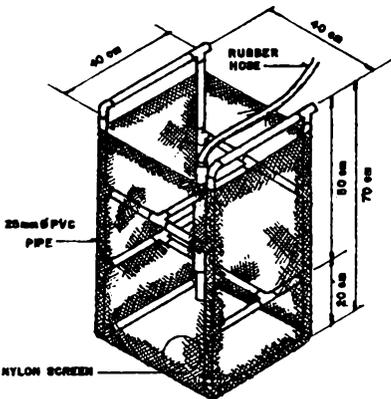
Chlorine test kit. The kit is used to determine residual chlorine concentration in the water, so that the amount of thiosulfate to be added during neutralization can be computed.



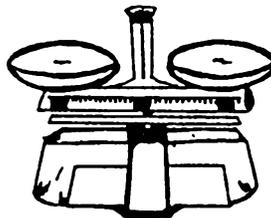
Microscope. This is for counting algal cells; also helpful in detecting abnormalities and diseases of the larvae at an early stage.



Hemacytometer. This is for determining the number of algal cells in a given volume.

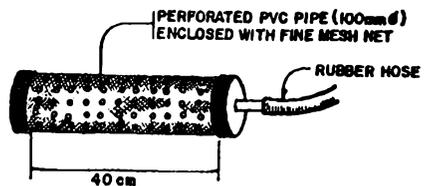


Blender. Processing of some microparticulate diets can be done by the blender.

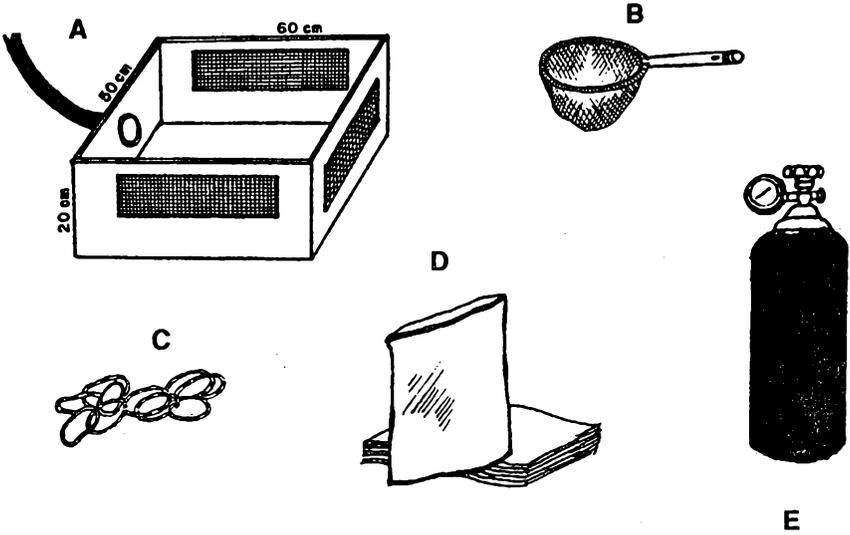


Weighing scale. Feeds and chemicals must be accurately weighed.

Drainers and hoses. For water change, mesh size of drainers must be smaller than the size of the larvae. Refer to Appendix A for the appropriate mesh sizes.



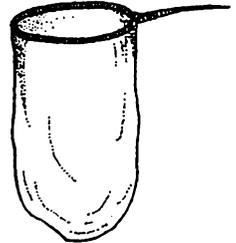
Harvesting boxes (A), scoop nets (B), rubber bands (C), plastic bags (D), filled oxygen tanks (E), pandan bags (F). These are used in harvest and transfer of animals.



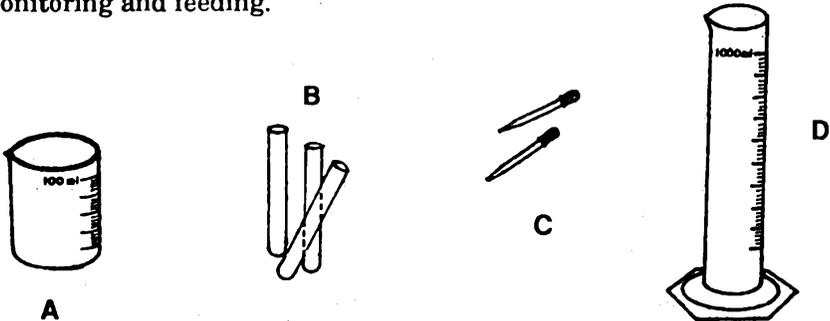
Filter bags. These are used to filter water prior to use of water for rearing. Mesh sizes are preferably 1 and 5 μ .



Basins and pails. Small-scale culture of algae, feeding, and transfer or harvest of animals require basins and pails.



Beakers (A), test tubes (B), droppers (C) and graduated cylinders (D). These are useful in monitoring and feeding.



HATCHERY OPERATIONS

Mortality during hatchery operations is oftentimes caused by poor quality of nauplii, unfavorable environmental conditions, diseases, and deficient or poor nutrition. The following section describes standard methods of management used in the hatchery. Disease prevention is emphasized through proper preparation, water treatment, and management.

Fig. 12 shows the daily activities involved during the entire hatchery run. These activities will be described in detail in the following sections.

Natural Food Production

Production of phytoplankton or algae for feeding must be planned and synchronized with the hatchery operations. Diatoms or other natural food must be ready as soon as the larvae molt to the first feeding stage (Z_1). The most common algae used as food are *Skeletonema*, *Chaetoceros*, and *Tetraselmis* (Fig. 13).

Procedure 1. Production Schedule

1. Compute the total volume of algae needed for daily operations. This is dependent on the volume of larval tanks to be used and the species of algae to be cultured. The total volume of algae required can be computed as follows (see p. 20 for sample computation):

$$V_{AT} = \frac{2F_A \times N \times V_L}{D}$$

where: V_{AT} = total volume of algae required/day (in t);
 F_A = desired feeding density; usually 20 000-50 000 cells/ml for *Skeletonema* or *Chaetoceros* and 5000-10 000 cells/ml for *Tetraselmis*;
 N = number of days for culture to reach peak density;
 V_L = total effective volume of larval tanks (in t); and
 D = average peak density of species cultured

Average peak densities vary with site and species, but it is usually safe to assume that for *Skeletonema*, an average peak density of 300 000 cells/ml can be attained in 2 days. For *Chaetoceros*, this could reach one million (1×10^6) cells/ml after 1-2 days, and for *Tetraselmis*, 100 000 cells/ml in 3-5 days. For more accurate estimates, a trial run must be conducted at the hatchery site. For subculture or feeding purposes, use the algae at the exponential growth phase and not at the declining phase (Fig. 14). Algal

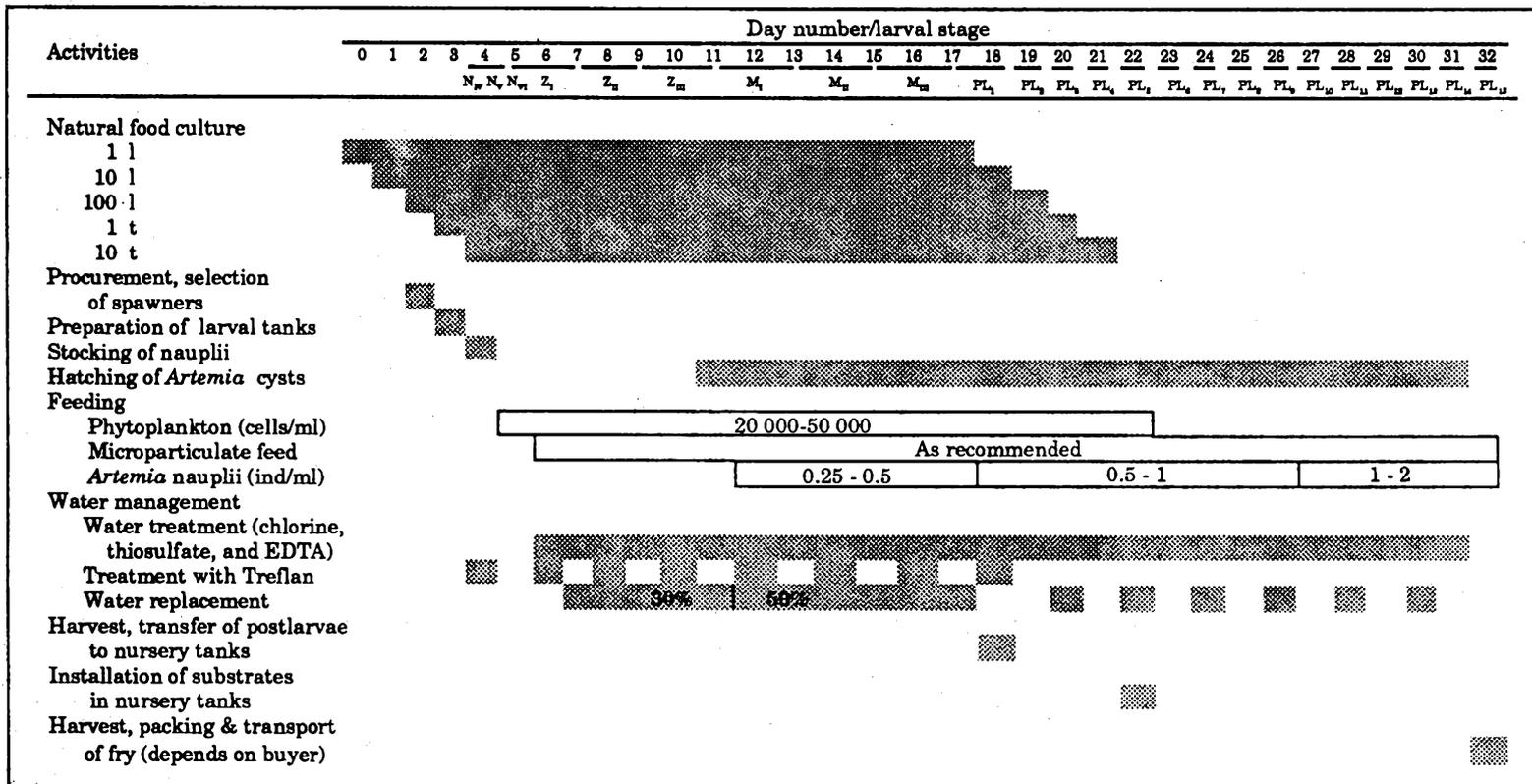


Fig. 12. Schedule of activities during a hatchery operation.

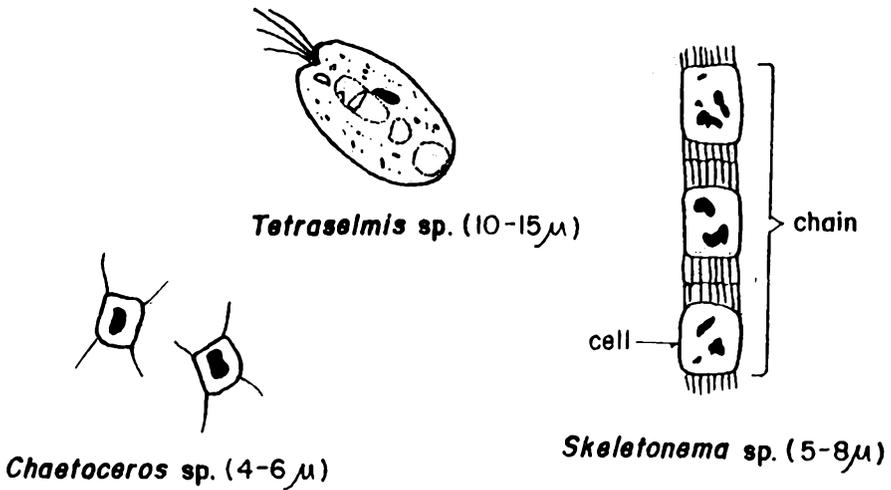


Fig. 13. Algae commonly used as food.

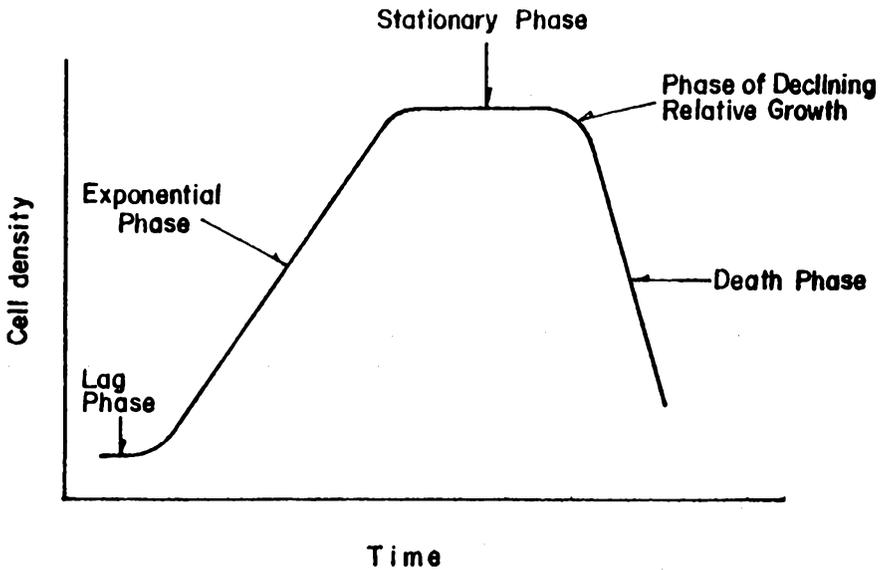


Fig. 14. Growth pattern of algae in a culture medium (Fogg 1975).

culture in a collapsed state is characterized by the presence of brownish suspended particles composed of clumped algal cells and bubbles on the water surface and sides of tank.

The volume of algae (based on the equation on p. 17) required for a 100-t hatchery where *Chaetoceros* or *Skeletonema* will be used is:

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$$V_{AT} = \frac{50\,000 \text{ cells/ml} \times 2 \times 100 \text{ t}}{1\,000\,000 \text{ cells/ml}} = 10 \text{ t}$$

If *Tetraselmis* is to be used, the same volume of algae is required but total algal tank volume should be three times that required for *Skeletonema* or *Chaetoceros*.

2. Make a schedule of production for this algal requirement. Algal starter should be 10% of the desired final volume. Fig. 15 shows one possible production schedule for a hatchery with a daily requirement of 10 t of algae.

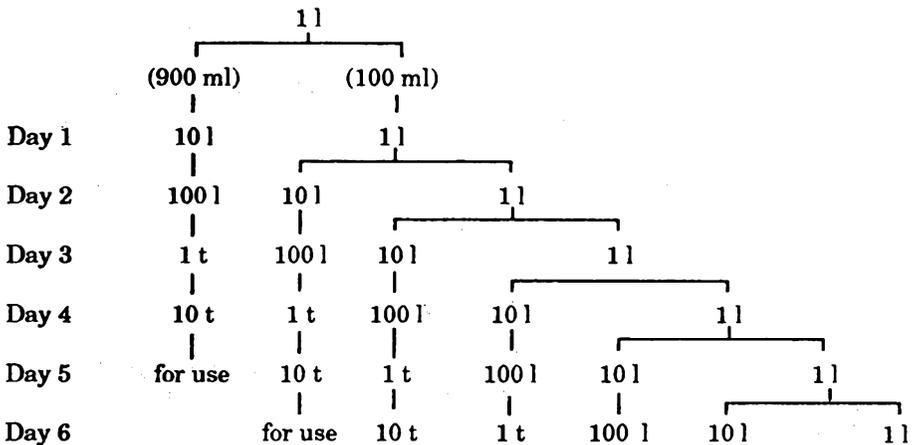


Fig. 15. Production schedule for an algal requirement of 10 t/day.

As shown in Fig. 15, 2 l of algae will be needed initially (1 l to scale-up and 1 l to refrigerate for future use). Based on this program, it takes 5 days before 10 t of culture will be ready for feeding.

Procedure 2. Mass Production of Algae

1. Procure the desired volume of algae based on the production schedule. Always procure twice the volume of algae needed on the first day of culture. Refrigerate the extra stock in a tightly closed double plastic bag or clean plastic container.

2. Prepare clean containers for the culture.

3. Fill the containers with the predetermined volume of water. If possible, use previously chlorinated and neutralized water (refer to procedures 10 and 11).

4. For every ton of algal culture, weigh or measure the following chemicals based on the corresponding requirements:

For *Chaetoceros* or *Skeletonema*:

- 100 g urea (46-0-0) (100 ppm)
- 40 g triple 14 (14-14-14) (40 ppm)
- 2.5 g ferric chloride (FeCl₃) (2.5 ppm)
- 2.5 g disodium EDTA (2.5 ppm)
- 6.0 g sodium metasilicate (NaSiO₃) (6 ppm)

For *Tetraselmis*:

- 80-100 g urea (46-0-0) (80-100 ppm)
- 10-15 g monoammonium phosphate (16-20-0) (10-15 ppm)

5. Dissolve each chemical separately in enough clean freshwater. Add to the algal culture water.

6. Add the starter and aerate. Make sure that the cultures are properly illuminated either by sunlight or artificial light. Depending on the species, the phytoplankton is ready for use either as starter or feed after 1-3 days of culture.

Preparation of Spawning, Larval, and Nursery Tanks

To prevent disease outbreak, the hatchery should be totally dried after several production runs. Tanks and facilities in the hatchery must also be cleaned well prior to a hatchery run. New tanks need to be filled with fresh- or seawater for at least a week to avoid mortalities due to toxic effects of chemicals used during construction of the tanks.

Procedure 3. Tank Preparation

1. For tanks which have already been conditioned and previously used, proceed to step 5. For newly painted or constructed tanks, fill the tanks with fresh- or seawater and leave overnight.

2. Drain the water the following morning.

3. Repeat steps 1 and 2. Refill with water and let the tanks stand for 5-7 days.

4. Drain the water.

5. Brush and scrub the tanks with water and detergent.

6. Rinse thoroughly with freshwater.

7. Prepare a sufficient amount of 10% hydrochloric acid solution by adding 100 ml of hydrochloric acid to 900 ml water.

8. Splash this solution on tanks. Rinse thoroughly with freshwater.

9. Dry for at least 1 day under direct sunlight if possible.

10. Install aeration hoses with airstones in the tanks. Airstones are usually placed 1 m apart throughout the tank area.

11. Fill the tanks to the desired volume with clean seawater. Aerate moderately.

Selection and Stocking of Spawners

Nauplii to be reared to the fry stage can come from either a) broodstock - wild or pond-reared immature female induced to mature by eyestalk ablation; or b) wild spawners - female prawns caught from the sea with developed ovaries. The details on how to maintain and handle broodstock as nauplii source are described by Primavera (1983). The number of spawners needed for a hatchery run is dependent on the nauplii requirement. For every million nauplii, about 4-5 wild spawners or 7-8 female broodstock are needed.

Spawners procured as nauplii source must be carefully selected to obtain high fertilization and hatching rates of eggs. Stage of maturity should not be used as the sole basis for selection. Spawners must also be disease-free. To ensure development of the eggs, females should be mated to ensure release of sperm cells necessary for fertilization.

Procedure 4. Selection of Spawners

1. Hold the spawners gently but firmly with both hands. Hold the underside of the prawn against a light source (Fig. 16). Determine maturity



Fig. 16. Visual inspection against a bright light to determine the sexual maturity stage of *P. monodon*.

stage (Primavera 1983) based on the outline of the ovary as in Fig. 17. Choose spawners at either Stage III or IV.

2. Feel the shell of the prawn, especially the lower portion of the carapace (refer to Appendix B). Reject those with soft or injured shells or those with dull red color. A reddish color usually indicates that the animals are stressed.

3. Lift the carapace carefully to expose the portion of the ovary covered with a transparent membrane. The color of the ovary should be olive green and not whitish. The whitish color prevails when the animal is infected with a protozoan disease (white ovary disease).

4. Examine the thelycum at the underside of the animal. Choose females with opaque white and bulging thelyca as these indicate the presence of deposited sperm. Reject those with black spots on the thelycum.

5. Examine the legs. Reject those with cut or missing legs.

6. Place the selected spawners in a container with water. Stock one

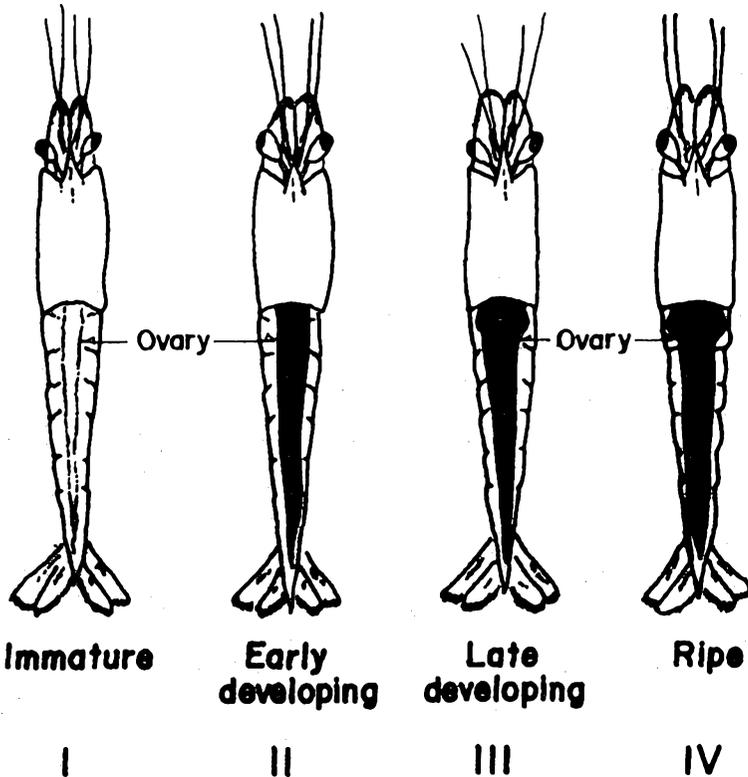


Fig. 17. Ovarian maturation stages as seen through the dorsal exoskeleton (Primavera 1983).

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spawner/ 3 l of seawater. Add 0.2 ml formalin/l of water in the plastic basin (200 ppm).

7. After 30 min, place the prawns in spawning tanks containing seawater. It is preferable to place one spawner per tank to avoid contamination of the whole stock in case one or a few are disease carriers. The water temperature should be within the range of 27-30°C and the salinity within 30-36 ppt.

8. Aerate and cover the tanks.

9. After spawning, remove spawners from tanks. This is done to lessen chances of contamination of eggs with bacteria from the feces of spawners.

10. Turn off aeration to allow the eggs to settle at the tank bottom. Scoop out the scum at the tank surface and sides.

11. If the spawning tanks are not too large, collect eggs in a 100-120 μ mesh nylon screen and rinse several times with clean seawater. If collection of the eggs is difficult due to the large tank volume, turn off the aeration temporarily and replace 50% of the water in the spawning tank. Make sure that the eggs are not siphoned out by draining only near the water surface and not at the bottom area where eggs will concentrate. Otherwise, drain with a 100-120 μ mesh net.

12. Resume aeration.

Stocking of Nauplii

During stocking and throughout the culture period, animals must not be exposed to abrupt changes in environmental conditions. The animals must be given time to gradually adapt to new conditions to avoid stress and mortalities.

Procedure 5. Stocking of Nauplii

1. Turn off the aeration in the spawning tanks. Cover the tank completely and leave only a small portion of the top exposed to light. Being attracted to light, the nauplii will concentrate on this area. With a small hose, siphon out the nauplii into a pail (Fig. 18). Aerate.

2. Get 3 100-ml subsamples. Count the nauplii in the subsamples. Get the average count and multiply by 10 in order to get the number of nauplii per liter (D_i) in the pail. Compute for the water volume to be placed in each tank using the following formula:

$$V = \frac{D_d \times V_L}{D_i}$$

where: V = volume of water in the pail to be placed in each tank (in l);

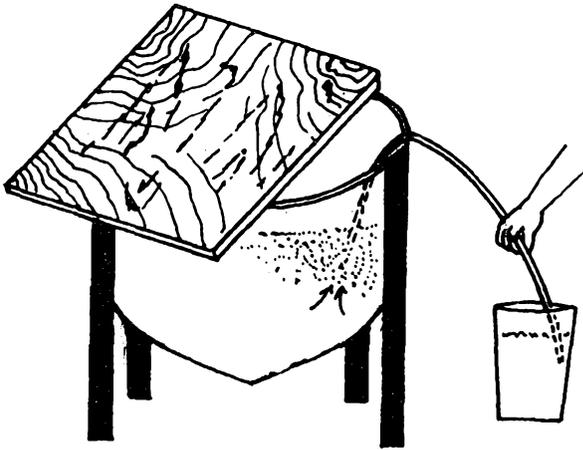


Fig. 18. Siphoning of nauplii into a pail.

D_d = desired stocking density (50-100/l);
 V_L = volume of larval tank (in l); and
 D_i = density of nauplii in the pail

3. Determine temperature and salinity of the water in the larval and spawning tanks. If the difference is more than 1°C or 5 ppt, respectively, proceed to step 4. Otherwise, stock nauplii directly in the tank.

4. Transfer the nauplii to be stocked to a clean basin in each tank.

5. Clean the outer part of the basin. Let the basin float on the larval rearing water for about 10 min. Place a liter of culture water every 5 min until conditions are similar to those in the rearing water. Allow the water to overflow until all the nauplii have been moved out into the rearing tank.

Feeding

Nauplii subsist on the yolk stored in their bodies. Since they do not require food, larvae start to feed at the first protozoal substage. Diatoms such as *Skeletonema* or *Chaetoceros* can be used for feeding the protozoa. Instead of diatoms, larvae at the second protozoal substage may be fed *Tetraselmis*. Artificial diets, called microparticulates because of their small particle size, such as MBD (refer to Appendix C for composition) or other commercially available diets can also be used as food during these sub-stages.

At the mysis stage, some animal protein must be present in the diet. The most commonly used protein source are newly hatched *Artemia* nauplii

and microparticulate diets which contain about 45-50% protein. The feeding schedule is summarized in Fig. 12.

When the animals reach the postlarval stage, egg custard, trash fish, mussel meat, or ground dried *Acetes* (small shrimp or "alamang") can be given to supplement the *Artemia* nauplii diet. Trash fish or mussel meat may be given either raw or cooked. Cooked trash fish is rubbed against a net to separate the muscle fibers while mussel meat is chopped or blended with water before feeding.

Procedure 6. Feeding of Diatoms and Other Algae

1. Get a sample of algae from the rearing water and algal culture tank. Place samples separately in clean test tubes.

2. Place a drop of sample on a clean hemacytometer and put a cover slip over (Fig. 19).

3. Examine samples under a microscope. Count the algal cells inside areas A, B, C, and D (Fig. 19). *Skeletonema* should be counted per cell and not per chain.

4. Compute for the algal densities using the following formula (based on Martinez et al. 1975):

$$D_a = \frac{(A+B+C+D) \times 10^4 \text{ cells/ml}}{4}$$

where: D_a = density of algae (in cells/ml); and
 $A+B+C+D$ = sum of the algal counts in these areas

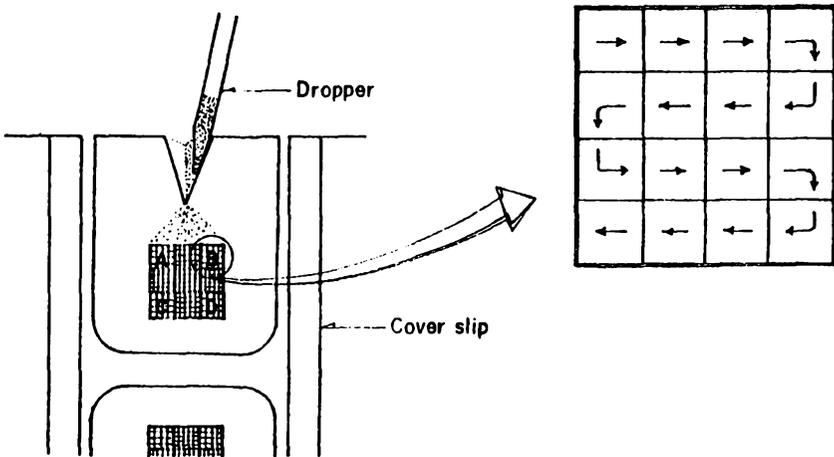


Fig. 19. Loading algal sample into the hemacytometer. The enlarged portion at the right shows the direction (arrows) for counting algae in areas A,B, C or D under the microscope.

5. Compute for the volume of algae you will need for feeding:

$$V_F = \frac{D_D - D_{LT} \times V_{LT}}{D_{AT}}$$

where: V_F = volume of algae to be used for feeding (in t);
 D_D = desired density for feeding;
 D_{LT} = density of algae in the larval tank;
 V_{LT} = effective volume of the larval tank (in t); and
 D_{AT} = density of algae in the algal tank

6. Get the amount of algae needed for feeding from the algal tanks with a clean scoop or pail or with a transfer pump if the required volume is large. Pour slowly and evenly throughout the larval tank.

Procedure 7. Feeding Artificial or Microparticulate Diets

1. Refer to the product label or brochure of the commercial diet for the recommended feeding regime for each larval stage. Weigh the amount needed for feeding based on the recommendation.

2. For most artificial diets (except microencapsulated diets), best results are achieved when these are mechanically blended with water prior to feeding. However, passing the diet with water through a net with the appropriate mesh size may be sufficient if no blender is available. For microencapsulated feeds, follow the recommended procedure on the label.

Procedure 8. Feeding with Brine Shrimp

1. Determine the total volume (in ml) of the larval tanks where *Artemia* will be used for feeding. Refer to Appendix D for equivalents.

2. Multiply the tank volume (in ml) by the recommended quantity in Fig. 12 in order to give the number of *Artemia* nauplii required for feeding.

3. Determine the amount of *Artemia* cysts to be incubated. Use the following formula:

$$W_A = \frac{N \times HE}{1\ 000\ 000}$$

where: W_A = weight of *Artemia* cysts (in g) to be incubated;
 N = number of *Artemia* nauplii required for feeding (determined in step 2); and
 HE = hatching efficiency of given batch (in g cysts/million nauplii; refer to Appendix E)

4. To disinfect cysts, dissolve thoroughly 0.2 g calcium hypochlorite/l of seawater (or 200 ppm hypochlorite) in the *Artemia* hatching tank. Aerate vigorously.

- 5. Place the cysts in the *Artemia* hatching tank.
- 6. After 30 min, harvest and wash cysts thoroughly. Rinse the hatching tank.
- 7. Resuspend the disinfected cysts in the *Artemia* tanks with clean seawater. Aerate vigorously. Tanks should be illuminated to ensure efficient hatching.
- 8. Harvest the *Artemia* nauplii after the recommended incubation time specified on the product label.

Procedure 9. Harvesting Artemia Nauplii

1. Place the *Artemia* incubation tank near a light source. Cover with a black cloth or carbon paper, leaving a small portion at the bottom uncovered (Fig. 20). The positively phototactic nauplii will concentrate on this area.

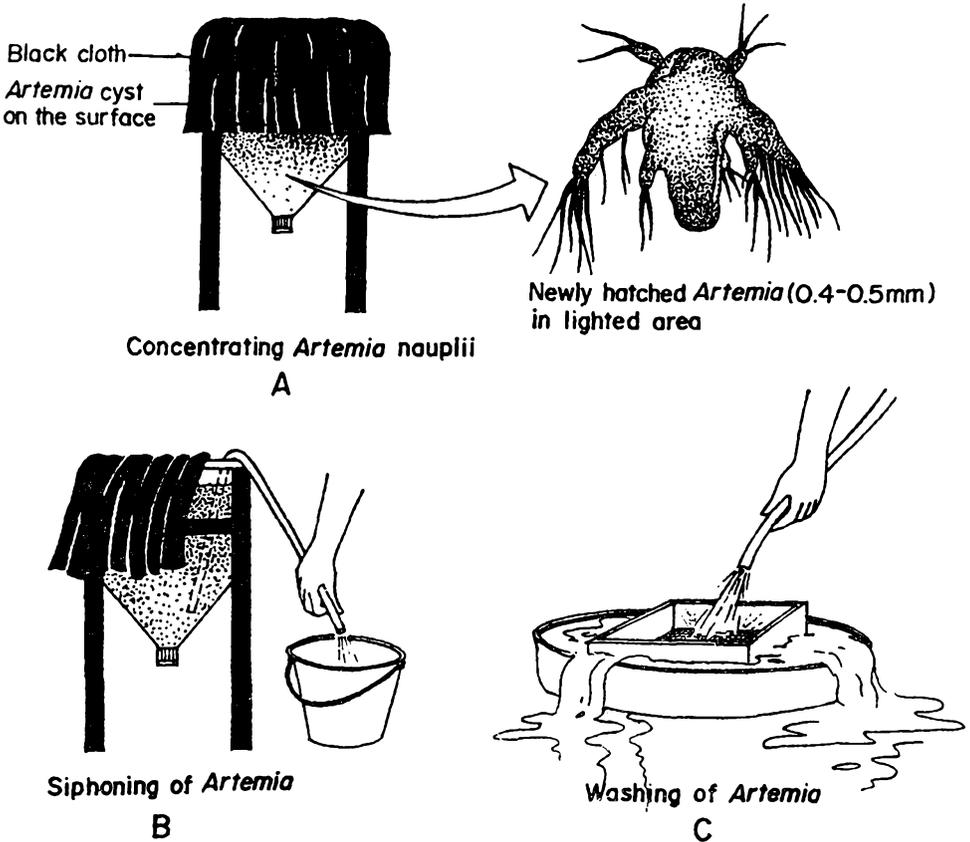


Fig. 20. Steps in harvesting newly hatched *Artemia*.

2. With a small hose, siphon out the *Artemia* nauplii in the lighted area and collect in a net with a mesh size of 150 μ .

3. Rinse and clean the harvested nauplii with sea- or freshwater. Store in a clean container with aerated seawater for not more than 24 h, or feed directly to shrimp larvae.

Water Management and Treatment

The quality of the culture medium in the larval rearing tanks deteriorates after sometime due to the accumulation of feces, decomposition of uneaten food and dead larvae. Regular water change dilutes the concentration of toxic metabolites which may have accumulated in the tank. Water change is done daily starting on the second day of the first protozoal substage (Z_1). About 30% of the water volume is replaced at the protozoal stage and 50% at the mysis and postlarval stages (Fig. 12). The difference in water temperature and salinity between the old rearing water and fresh seawater should not be more than 1°C or 2 ppt, respectively.

Rearing water is disinfected before use to eliminate or reduce harmful microorganisms. An oxidizing agent such as hypochlorite kills or retards the growth of possible disease-causing agents. However, it is also toxic to larvae or postlarvae. Water intended for replacing rearing water is therefore neutralized after chlorination and before use.

Procedure 10. Disinfection of Rearing Water

1. Pump seawater into your reservoir via the sand filter.
2. Determine and weigh the amount of calcium hypochlorite needed for disinfection using the following formula:

$$W_{Cl} = \frac{10 \times V_w}{C_{Cl}}$$

where: W_{Cl} = weight of hypochlorite (in g);
 V_w = volume of water to be treated (in t); and
 C_{Cl} = percentage of hypochlorite in the product

3. Dissolve hypochlorite in a pail of clean water. Aerate vigorously for faster dissolution.

4. Add the hypochlorite solution to the water which is to be treated. Aerate vigorously for homogeneous mixing.

5. Neutralize 12 to 24 h after disinfection with hypochlorite.

6. Dissolve and add 5 - 10 g of the chelating agent disodium EDTA (ethylenediaminetetraacetic acid) to a ton of previously neutralized water. Use within 6 h after neutralization.

Procedure 11. Neutralizing Chlorinated Water

1. Determine the amount of residual chlorine in the water. Use a commercially available chlorine test kit (procedures for residual chlorine determination are described in manuals which are given out with the product).

2. For every 1 ppm of hypochlorite, neutralize with an equivalent amount of sodium thiosulfate. Dissolve the sodium thiosulfate in a separate container. Add to the chlorinated water.

3. Aerate vigorously.

4. After 30 min, measure the amount of residual chlorine in the water. If some amount of residual chlorine is still present, repeat steps 2 to 4. Be sure that residual chlorine is zero before using the water.

Harvest, Transfer, Packing, and Transport

It is important that proper procedures of harvesting, packing, and transport are observed to ensure high survival of prawn fry. The procedure followed during transfer of postlarvae (PL₁ or PL₆) to nursery tanks is similar to the fry harvesting method described in Procedure 12.

The number of fry loaded per bag will depend on the size and age of fry, travel time, distance, and means of transportation. During extended transport periods, water temperature must be reduced to decrease molting and metabolic rates and the incidence of cannibalism among prawn fry. However, there is no need to lower water temperature in transport bags when transporting at night or during cool weather.

Procedure 12. Harvest and Transfer of Postlarvae to Nursery Tanks or for Transport

1. Lower the water level in the tank to about 1/3 to 1/4 of total volume to reduce water pressure in the drain pipe, hence, minimizing stress on the postlarvae.

2. Allow the remaining water in the tank to flow to the harvesting pit or box. Scoop the postlarvae from the tank with a scoop net. Distribute harvested fry in white basins or small tanks (Fig. 21).

3. Postlarvae are stocked in nursery tanks when the difference between water temperature and salinity in the rearing and nursery tanks is not more than 1°C or 2 ppt, respectively. Follow Procedure 13 when transporting fry.

Procedure 13. Packing and Transport of Fry

1. Estimate the total number of fry by the comparison or volumetric method as follows:

a) comparison method - count individual fry in a basin. Based on visual comparison of fry density, place the same estimated number of fry



1. Scooping of fry from the tank



2. Distributing fry in basins for counting



3. Pouring of fry in double-lined plastic bags



4. Oxygenating the plastic bag

Fig. 21. Harvest, counting, and packing procedures of prawn fry.

in the other remaining basins. Multiply the initial number of fry counted individually by the number of basins.

b) volumetric method - scoop out the harvested fry in a container (100-300 l capacity) using a 500-1000 ml beaker and place equal volumes in each basin. Aerate vigorously while sampling. Do a headcount of the fry in one basin. Multiply the number of fry by the number of basins.

2. Place double plastic bags measuring 50 cm x 90 cm in pandan bags, styrofoam boxes or laminated boxes.

3. Put the water and fry from the basins into the inner plastic bags. A transport bag should contain 5-6 l of seawater. If transport time is less than 6 h, about 2 000 to 2 500 PL_{25} to PL_{30} can be placed in the bag. Decrease the number to 500/bag for older and bigger fry (PL_{40} to PL_{60}).

4. Saturate the inner bag with oxygen and tie the two bags separately

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with 3-4 pieces of rubber bands for each bag.

5. If travel time exceeds 6 h, lower water temperature to 20-26°C. Maintain this by placing wrapped ice on top of the plastic bags. Reoxygenate the bags and replace the water if transport time exceeds 12 h.

Upon reaching the farm site, acclimatize fry to the water temperature and salinity of the nursery or rearing ponds prior to stocking.

DISEASES

Disease-causing microorganisms are always present in the water. These may harm the larvae especially when the latter are exposed to stressful conditions such as inadequate nutrition, overcrowding, poor water quality, and sudden changes in temperature, salinity, and other physico-chemical parameters. General indications are incomplete molting, empty digestive tract, deformed extremities, reddening of the body, and sluggish movement. To avoid mass mortality of the larvae, disease prevention measures which have been described in previous sections of this manual must be observed. Po et al. (1989) presents more detailed practices for disease prevention in prawn hatcheries.

Information on common diseases, their manifestations and prevention and control (Table 2) has been gathered from various references and collated by Baticados (1988). The authors, though, discourage the indiscriminate use of drugs because of possible development of drug-resistant strains of disease agents.

ECONOMICS

The projected economic performance of a business venture can be assessed through economic indicators such as those computed below. Although price of fry has declined, values obtained indicate that operation of a shrimp hatchery is still profitable.

Investment Requirements for a Small-scale Prawn Hatchery

The capital investment needed for a small-scale hatchery with a target production of 500 000 fry/run is shown in Table 3. Larval tanks

Table 2. Diseases of *Penaeus monodon* larvae and postlarvae (Baticados 1988)

Disease	Stages affected	Signs	Prevention/control
Viral [caused by <i>P. monodon</i> Baculovirus (MBV)] disease	PL ₅ to adult	Postlarvae small and dark, loss of appetite, lethargy, reduced preening activity	Prevention through sanitation, proper nutrition; eradication by depopulation and disinfection
Viral [Infectious Hypodermal and Hematopoietic Necrosis Virus (IHHNV)] disease	PL	80-90% cumulative mortality, latent infection in larvae possible	Prevention through avoidance; eradication by depopulation, disinfection, and isolation of live prawns
Necrosis of appendages	Z, M, PL	Necrosis of appendages, twisted antennae or broken setae, "browning" of shell or tip of appendages; liquefaction of gut contents in zoea	Erythromycin phosphate (1 ppm); Streptomycin-bipenicillin (2 ppm); Tetracycline chlorhydrate (1 ppm); Sulfamethazine (3 ppm); Furanace (0.1 ppm) every other day; Chloramphenicol (1 ppm) every 3 days or 2-6 ppm every 2 days(prophylactic) or 2-10 ppm (therapeutic)
<i>Vibrio</i> disease	Z	Heavy mortality reaching 80%	
Luminous bacterial disease	N,Z,M,PL	Prawn luminiscent in the dark, heavy mortality	Prevention by chlorination of water, removal of bottom sediments, and more frequent water change
Filamentous bacterial disease	Z,M,PL	Bacteria on gills, setae, appendages and body surface; mortality due to hypoxia, impaired molting	0.1 mg copper/l (Cu/l), 24 h or 0.25 mg Cu/l, 4-6 h
Fungal (caused by <i>Lagenidium</i>) disease	Egg,Z,M, PL	Fungus replaces internal tissues of prawn, eggs do not hatch,	Prevention by water management; reduction of stocking density;

Continued next page

Table 2, continued...

Disease	Stages affected	Signs	Prevention/control
		larvae weak, up to 100% mortality within 1-2 days	Treflan or trifluralin (0.2 ppm), 24 h treatment; disinfection of spawners with Treflan R, 5 ppm, 1 h; Malachite green, 0.006 ppm, only for mysis; disinfection of eggs with powdered Tide detergent (20 ppm), 2-4 h
Fungal (<i>Sirolopidium</i>) disease	N,Z,M	Same as above	Disinfection of equipment and tanks; filtration of water; treatment of spawners with Treflan (5 ppm), 1 h; sanitation; disposal of infected larvae
Fungal (<i>Haliphthoros philippinensis</i>) disease	Z,M	Same as above	Furanace, 1 ppm, 6 h
Fungal (<i>Fusarium</i>) disease	N,Z	Scanty white cottony strands attached to larvae; mortality	None reported
Fungal (<i>Hyphomyces</i>) disease	N,Z	Larvae move sluggishly	None reported
Ciliate (<i>Epistylis</i> , <i>Zoothamnium</i> , <i>Ephelota</i> , <i>Acineta</i>) infestation	all stages	Respiratory and locomotory difficulties, loss of appetite, fuzzy mat on shell sometimes formed	Chloroquin diphosphate, 1.1 ppm, 2 days
Gregarine disease	N,Z,M,PL	Interferes with particle filtration to gut or hepatopancreatic duct	Eliminate molluscan intermediate host
Heavy metal poisoning	N,Z,M,PL	Morphological deformities; mortality	Water change
Gas bubble disease	all stages	Gas bubble in gills, under cuticle or gut; prawn exhibits erratic swimming behavior	Mechanical aeration to reduce dissolved oxygen levels once disease is present

Table 3. Costs and returns of a hatchery with target production of 500 000 fry per run

Item	Quantity	Unit cost (P)	Value (P)
Sales	500 000	0.12/PL	60 000
Variable cost			
Nauplii	2.5 million	3 000/million	7 500
<i>Artemia</i>	12 cans	450/can	5 400
Artificial diets	1.5 kg	2 000/kg	3 000
Fertilizers & chemicals			1 500
Electricity	800 kwh	2.6/kwh	2 080
Marketing expenses	2% of revenue		1 200
Labor:			
Technician	1	3 500	3 500
Aides	2	1 000	2 000
Miscellaneous	5% of variable cost		1 309
			<u>27 489</u>
Fixed cost			
Repair & maintenance	5% of fixed assets		1 725
Depreciation			<u>11 017</u>
			12 742
Total cost			40 231
Net income/run			19 769
Net income/yr (6 runs)			118 614
Less income tax (35%)			<u>41 515</u>
Net profit after tax			77 099
Capital assets	P307 000		
Working capital (2 runs)	P 54 978		
Total investment requirement	P361 978		

are assumed to be made of canvas, with an economic life of only 2 years (Table 4).

Undiscounted Economic Parameters

1. Unit cost = total cost/total pieces
 = P40 231/500 000
 = P0.08/pc

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Table 4. Capital outlay and depreciation schedule using straight-line method

Item	Value (P)	Economic life (yr)	Depreciation/yr (P)
Land, 1000 m ² at P100/m ²	100 000	—	—
Hatchery building	20 000	5	4 000
Equipment	20 000	10	2 000
Hatchery fence	5 000	5	1 000
Larval & postlarval tanks	50 000	2	25 000
Algal tanks (10% of volume of larval tanks)	5 000	2	2 500
Reservoir	25 000	2	12 500
Generator & pumps	30 000	5	6 000
Aeration	15 000	5	3 000
Seawater system	20 000	3	6 700
Filtration	6 000	5	1 200
Electrical system	6 000	5	1 200
Miscellaneous	5 000	5	1 000
Total	307 000		66 100
Depreciation/run			P11 017

2. Variable cost /unit = variable cost/total pieces
 = P27 489/500 000
 = P0.05/pc
3. Return on investment (ROI)
 = net profit after tax/total investment
 = P77 990/P361 978 x 100
 = 21.30%
4. Payback period = total investment/(net profit after tax + depreciation)
 = P361 978/(P77 099 + P66 100)
 = 2.5 yrs.

For the values above, the operator will neither gain nor lose if he produces 335 258 pieces of fry during a run. This production is 67% of the target level of 500 000 fry/run. Israel et al. (1986) presented a more detailed description on the economics of small-, medium-, and large-scale hatcheries.

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GLOSSARY OF TERMS

- Ablation - incision and removal of the contents of the eye and eyestalk to induce gonad maturation
- Acclimatize - to gradually adapt to the environment
- Aerate - to supply with air
- Algae - refers to microscopic marine plants
- Ammonia - a metabolite excreted by shrimps and aquatic animals
- Appendages - include swimming and walking legs and also the mandibles in nauplii
- Broodstock - adult male or female prawn which can be induced to sexually mature
- Chlorination - treatment with chlorine (bleach) solution
- Cyst, *Artemia* - dormant stage of brine shrimp (*Artemia*) where eggs have a hard, thick protective outer layer; this may be hatched under suitable conditions
- Demersal - bottom dwelling
- Density - number of individuals or units per volume
- Density, algal - number of cells of algae per volume
- Diatom - microscopic algae with siliceous cell walls
- Disease - an abnormal condition affecting growth, function, or appearance of the animal
- Disinfect - to rid of harmful microorganisms
- Dorsal - referring to the top side of the prawn
- Effective volume - actual volume of water that a container can hold
- Fecal matter - waste excreted through the anus
- Feeding regime - feeding schedule specifying amount of feed to be given
- Formalin - a 37% formaldehyde solution used as disinfectant
- Furcal spine formula - indicates the number of spines on each side of the furca (refer to Fig. 4); a formula of 1+1 indicates that there is 1 spine on each side of the furca
- Hatching rate - the number of eggs which hatch into the nauplius stage; usually computed as the number of nauplii/total number of eggs incubated x 100
- Hatching efficiency, *Artemia* - grams of *Artemia* cysts that will produce one million nauplii
- Hypochlorite - a chemical used as the major component in bleach
- Incubation - to maintain under favorable environmental conditions to aid egg development and hatching
- Induce - to stimulate and cause
- Metabolism - the process of producing energy in a living thing through the chemical breakdown of food
- Microencapsulated diet - a microparticle enclosed in a special protein coat;

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refers to a type of artificial diet

Molt - to shed off the shell

Monitor - to check, record, and keep track

Neutralize - to counteract

Oxidize - to cause to combine with oxygen

pH - indicates acidity or alkalinity of a medium; a pH of 7 indicates neutrality; a value less or greater than 7 indicates acidity or alkalinity, respectively

Phototactic - response to light

Phototactic, positively - attracted to light

Phytoplankton - minute plants floating in water

Plumose - having hairs or feathery structures

Rear - to culture

Run, hatchery - period from the stocking of the nauplii until fry are harvested for stocking

Salinity - the concentration of dissolved salts

Scale-up - to progressively produce larger volumes

Seawater - water with a salinity of 30 to 40 ppt or ocean water

Setae - spines or hairlike projections

Spawners - mature females which are ready to spawn

Spermatophore - mass enclosing spermatozoa extruded by male

Starter, algal - inoculum; small volume of pure culture of algae used for starting mass production

Stock - to place in a tank

Stock, algal - reserve or supply

Toxic - poisonous

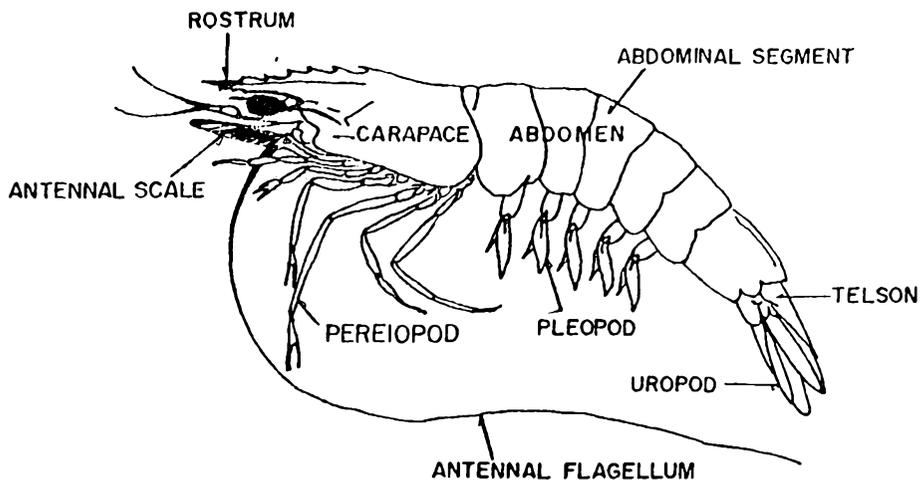
Ventral - refers to the underside

APPENDICES

APPENDIX A. Net Size for Different Larval Stages

Stage	Mesh size of net for draining (μ)
Eggs	100-120
Nauplius	100-120
Protozoa	120-210
Mysis	210-300
Postlarvae	300-350

APPENDIX B. Lateral View of *Penaeus monodon* Showing Important Parts (Motoh 1981)



**APPENDIX C. Composition of Kappa-carrageenan Microbound Diet
(Bautista et al. 1989)**

Ingredients	% Composition
Shrimp meal	35
Squid meal	30
Bread flour	11
Cod liver oil	8
Soybean lecithin	2.5
Cholesterol	1.0
Vitamin mix	6.0
Mineral mix	4.0
Butylated hydroxytoluene	0.5
Carotenoid	0.25
Cellulfil	1.75

APPENDIX D. Some Useful Unit Equivalents

1 gram (g)	= 1000 milligrams (mg)
1 kilogram (kg)	= 1000 g
1 kg	= 10 ⁶ mg
1 millimeter (mm)	= 1000 microns (μ)
1 centimeter (cm)	= 10 mm
1 meter (m)	= 1000 mm
1 m	= 100 cm
1 liter (l)	= 1000 milliliters (ml)
1 ton (t)	= 1000 l
1 t	= 10 ⁶ ml
1 t (water)*	= 1 cubic meter (m ³)
parts per thousand (ppt)	= mg/g or g/kg or ml/l or l/t or mg/ml or g/l or kg/t
parts per million (ppm)	= mg/kg or ml/t or mg/l or g/t

*Based on density of water which is 1 g/ml.

APPENDIX E. Hatching Efficiency* (g/million nauplii) of some Commercially Available *Artemia* Cysts

Brand name	Incubation period	
	24 h	48 h
Biomarine Fast Hatch	2.59	
Biomarine Standard	2.97	
Olympic	4.66	4.39
<i>Artemia</i> of Utah	4.88	4.65
<i>Artemia</i> 90	4.50	4.43
Argentemia	5.28	5.07
Aquafeeds	14.88	9.71
Sanders Premium Yellow Blue	5.40	4.65
Sanders Blue (A)	5.54	5.97

*Analyzed in the *Artemia* Laboratory, SEAFDEC/AQD.

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AVAILABLE IN 1991

Publications

- *Biology of Milkfish (Chanos chanos Forsskal)*. T.U. Bagarinao. Monograph.
- *SEAFDEC/AQD 1990 Annual Report*.
- *Seaweeds of Panay Island, Philippines*. A.Q. Hurtado-Ponce, M.R. Luhan, N.G. Guanzon, Jr. Monograph.
- *Identifying Milkfish Fry*. Aquaculture extension leaflet.

Video Tapes

- *SEAFDEC/AQD: Taking Strides into the '90s*. SEAFDEC/AQD research and development.
- *Getting Down to Sea Bass Tacks*. History, current culture systems, and constraints in sea bass culture in the Philippines.
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