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ARTIFICIAL REEFS AND SEAFARMING TECHNOLOGIES

CENTRAL MARINE FISHERIES RESEARCH INSTITUTE

INDIAN COUNCIL OF AGRICULTURAL RESEARCH
DR. SALIM ALI ROAD, POST BOX No. 1603, TATAPURAM - P. O.,
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Editor

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CULTURE OF SEA-CUCUMBER

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Introduction

The sea-cucumbers are entirely marine and distributed from the Arctic to the Antarctic region. They are distributed right from the supralittoral zone to the hadal zone. They inhabit a wide variety of habitats viz. rocky, muddy, sandy and coral. Most of them live among corals. They are well distributed on the reefs of the Indo-West Pacific region. Good concentration of them are found in the Philippines. Species belonging to the families Holothuridae and Stichopodidae alone are used for processing since they grow to a large size and the body wall is also thick.

In the seas around India more than 200 species are distributed. In the shallow waters 75 species are distributed. In the Indian waters the sea-cucumbers occur chiefly in the Gulf of Mannar and Palk Bay, the Andaman and Nicobar Islands and the Lakshadweep. In the Gulf of Mannar and Palk Bay *Holothuria scabra* (Pl. I) is the most important species for processing. In recent years *Actinopyga echinites* and *A. miliaris* are exploited in good quantities. *Holothuria spinifera* and *Bohadschia marmorata* are also distributed in small quantities. *Holothuria scabra* and *H. spinifera* are not distributed in the Lakshadweep. In the Lakshadweep *Holothuria nobilis* and *Thelenta ananas* are important species. The other species distributed in the Lakshadweep are *Bohadschia marmorata*, *Actinopyga mauritiana*, *A. echinites*, *A. miliaris*, *Stichopus variegatus*, *S. chloronotus* and *Holothuria atra*. In the Andaman and Nicobar Islands the most important species is *H. scabra*. The other species distributed are *Holothuria atra*, *A. mauritiana*, *A. echinites*, *A. miliaris*, *H. nobilis*, *Stichopus variegatus* and *S. chloronotus*. *Holothuria spinifera* is not distributed in the Andaman Islands. The record of *Thelenta ananas* from the Andamans needs to be checked. Sea-cucumbers are distributed in the Gulf of Kutch and also along the coast of the main land of India but they are not of commercial value.

The Central Marine Fisheries Research Institute started a Project on the seed production of *Holothuria scabra* in 1987 and they produced seeds for the first time in 1988 by inducing the sea-cucumber to spawn by thermal stimulation (James *et al.*, 1989). Since then seed of *H. scabra* is produced in the hatchery on a number of occasions. James and James (1993) have given prospects for sea-cucumber farming. James (1994 a, 1994 b) reviewed the seed production in Japan and China and also discussed the seed production of sea-cucumbers in India. Finally James *et al.* (1994 a, 1994 b) brought out a handbook on the hatchery techniques and culture.

Morphology and anatomy

Some information on the morphology and anatomy of the most important sea-cucumber *Holothuria scabra* from the Gulf of Mannar and Palk Bay is presented here.

External characters

The body is robust, elongated and cylindrical with blunt ends. The mouth is at the antero-ventral end, surrounded by a ring of 20 peltate tentacles, and the anus is at the postero-dorsal end. The dorsal side is convex and the ventral side flat. The total length ranges from 10-400 mm and the body weight ranges from 25 - 2000 g. The skin in large specimens is thick and slimy. On the dorsal side there are many small often inconspicuous papillae, which are thinly scattered. On the ventral side the tubefeet are densely arranged in an irregular manner. Each dark spot on the ventral side represents one pedicel.

In the live condition, it is grey to black on the dorsal side and white on the ventral side. Generally smaller specimens are totally black and larger specimens have a number of irregular yellow or white transverse bands on the dorsal

side. Mouth is oval, situated at the centre of the circlet of tentacles.

Anatomy

At the base of the tentacles there are ten calcareous plates, of these five are radials and five interradials. On either side there are two long structures known as respiratory trees. They open at the posterior end into the cloacal chamber. At the base of the calcareous ring there is a circular ring water canal to which two or three polian vesicles which look like transparent sacs, hang freely into the body cavity. There is also a single stone canal. The intestine is highly coiled and opens into the cloacal chamber. There is a single bunch of gonadial tubules attached to the dorsal mesentery and this opens out through a single gonopore in the mid-dorsal region near the anterior end. There are five longitudinal muscle bands.

Feeding

It is omnivorous in habit. The peltate tentacles surrounding the mouth shovel the sand and mud continuously into the mouth. Nutrition is derived from the organic matter in the mud or sand. An analysis of the contents in the digestive tract has revealed the presence of marine algae, copepods, diatoms and molluscan shells. No food preference is noticed. The small intestine always appears to be relatively empty, whereas the large intestine is distended with a load of bottom material. The feeding seems to be a continuous process.

Reproduction

The sexes are separate and it is not possible to differentiate the males and females externally. Gonad is single, consisting of numerous thin, filamentous tubules united basally into one tuft. It is attached to the left side of the dorsal mesentery and hangs freely in the coelom. The tubules are elongated and branched. From the gonadal base, the gonoduct proceeds in the mesentery and opens to the outside in the mid-dorsal region near the anterior end. In advanced stages of maturity of filaments of the ovary are coloured brown in which the eggs or oocytes are visible as small white spots. The testis consists of long white beaded filaments. The gonoducts

released through the gonopore by the ciliary action of the gonoduct. Fertilization is external and the embryo passes through different larval stages before setting down to the bottom as juveniles.

Brood stock maintenance

The brood stock material is procured from the commercial catches meant for processing. This material can be collected only during January to April. Therefore the brood stock material has to be maintained properly so that it can be used for spawning experiments. Sea-cucumbers are kept in one tonne FRP tanks with 800 litres of sea water. Aeration is provided at either side of the tank. In one tonne tank 15-20 individuals are maintained. At the bottom of the tank a layer of mud of 100 mm thickness is provided for the animals as feed. The water is changed completely daily. A feed was prepared with prawn head waste, soyabean powder and rice bran. This has 6.5% protein and the sea-cucumbers accept this food well. Daily 50 g of this feed is put in the one tonne tank as supplementary feed. The brood stock is used for spawning during January to April.

Spawning

Natural spawning

During the breeding season mature animals release the gametes without external stimuli. During February, March, April, August and December, the sea-cucumbers spawned on changing the water in the brood stock tanks.

Induced spawning

First sea water is heated in GI buckets by using immersion heater. Pure filtered sea water is taken in 100 lt FRP tank and the initial temperature is noted. Then the heated sea water is slowly added to the 100 lt tank. While adding heated water, the sea water in the 100 lt tank is thoroughly mixed to get uniform temperature. When the temperature rises by 5°C, adding of hot water is stopped. The brood stock animals are first washed and then introduced into 100 lt tank. The normal temperature of the sea water in the hatchery is 28-30°C.

Usually the brood stock animals are introduced into the spawning tank around 1000 hrs. First the temperature in the spawning tank falls

by 1 or 2°C, since the sea-cucumbers release cold water held inside their bodies. After 2 to 3 hours first the males start reacting by raising the anterior end swaying. The mature males start releasing the sperms in white streams. When once the males start spawning they spawn profusely. This will induce the other males also to spawn. It is better to keep one or two spawning males in the tank and remove the others since they foul the water. Excess sperm suspension is harmful for normal development of embryos. The females release the eggs after one or two hours after the males release the sperms. The females release the eggs in one or two spurts to help in wider distribution of the eggs. The eggs on release first sink to the bottom. They are immediately fertilized by the sperms.

Early embryonic and larval development

First the fertilized eggs are siphoned out into a sieve which is kept partially immersed in a bucket of water. The tank is gently washed with fresh sea water from all sides to collect the eggs into the sieve. Now the eggs are washed well with fresh filtered sea water to remove excess sperms. The contents of the sieve is transferred to a 10 lt beaker upto the mark. Then the water in the beaker is gently stirred to bring in uniform distribution of the eggs. From this 1 ml sample is taken and transferred to the counting chamber. The eggs in the counting chamber are numerically enumerated. This procedure is to be repeated three times to get the average number of eggs in the 1 ml of sea water. The average number multiplied by 10,000 gives the actual number of eggs in 10 lt beaker. The fecundity is five lakhs to one million.

Three to five lakhs of eggs can be stocked in one tonne tank with 800 lt water. Two aerators are provided for one tonne tank. The eggs are round and spherical with diameter 160-190 μ . The cleavage is complete and holoblastic. After 40 minutes blastula is formed. First day no feed is given. After 26 hours the embryo hatches out. After 24 hours gastrula is fully formed. The gastrula is motile and oval shaped. After 48 hours early auricularia appears. As days advance the lateral projections become prominent in the auricularia. On the tenth day some of the auricularia transform into doliolaria. After three days pentactula stage is reached. The various types

of larvae and different stages of development have been illustrated and described elaborately by James *et al.* (1994 a), and they are again briefly described below.

Auricularia larva

Early auricularia is formed after 48 hours. After five or six days late auricularia is formed. It is slipper-shaped, transparent and pelagic in habit. It has a pre-oral loop anteriorly and anal loop posteriorly. These bands help in locomotion. The digestive tract consists of mouth, an elongated pharynx and sacciform stomach. The early auricularia larva measures on an average 560 μ . The later auricularia has an average length of 1.1 mm. On the tenth day some of the auricularia larvae metamorphose to doliolaria larva.

Doliolaria larva

The doliolaria is barrel-shaped with five bands around the body. These larvae measure 460-620 μ . Rapid changes occur inside the body and all adult features of the holothurian set in. This stage is short and lasts only for two or three days and subsequently transformed into a creeping stage known as pentactula.

Pentactula larva

The pentactula is tubular with five tentacles at the anterior end and with a single tubefoot at the posterior and which helps in the locomotion of the larva. The pentactula creeps over the sides and bottom of the tank. They actively feed on benthic algae and other detritus matter. The pentactula measures 600-700 μ . If they are fed on algal extract some of them reach a length of 10 mm in one month and transform into juveniles.

Larval rearing

Rearing tanks should be clean and free from bacterial settlement. They should be scrubbed well before use and after washing they should be dried. Strict control of rearing density of the larvae is to be observed. If the density of the larvae is more, they will form as a ball and sink resulting in death. Therefore rearing density should be controlled to ensure better survival rate. The desirable density should be controlled to ensure better survival rate. The desirable density

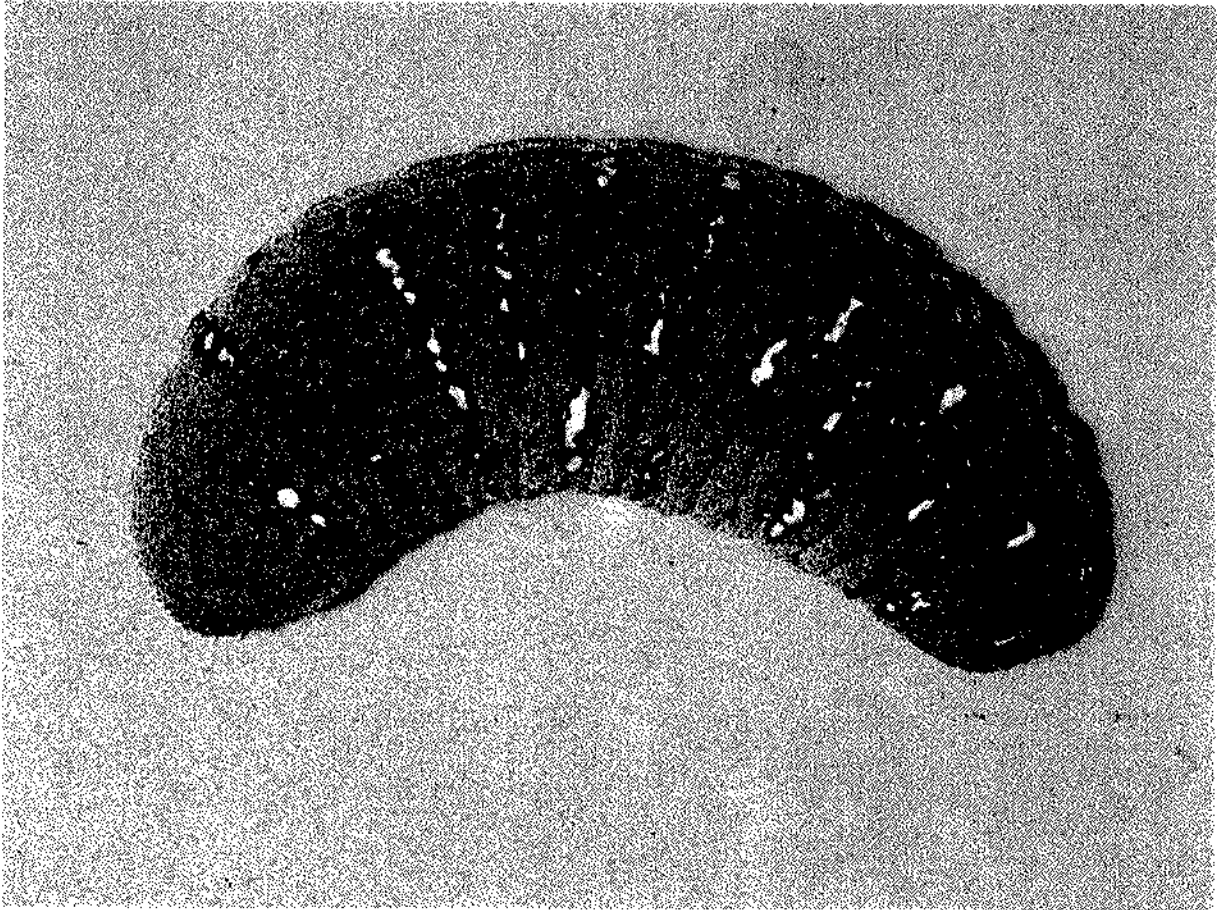


PLATE I. *Holothuria scabra*

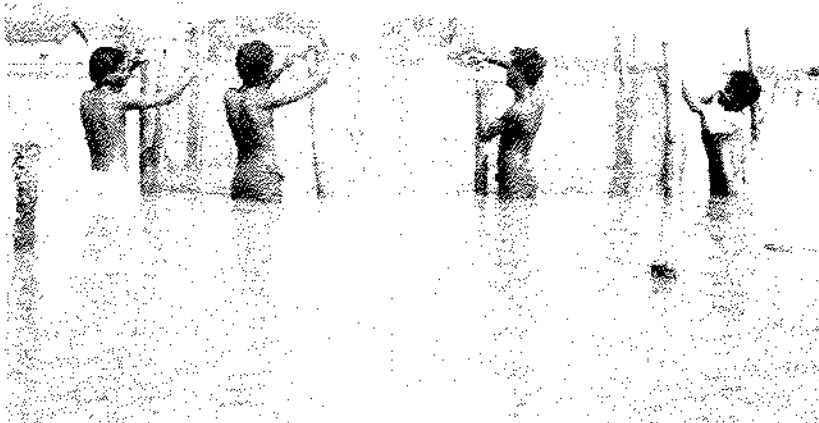


PLATE II A. Sea-cucumber seeds produced at the CMFRI hatchery at Tuticorin, B. Rectangular cage, C. Velon screen cage and D. Netlon cage.

of larvae is 300-700/lt. In one tonne tank with 750 lt of water 3,75,000 larvae can be stored. The number of larvae is estimated by using the same procedure as in the case of egg estimation. Normally the larvae are taken out once in three days so that the tanks can be cleaned thoroughly to avoid infestation of other organisms. On other days the water level is reduced to more than half by keeping the sieve inside the tank. The sediment must be removed to keep the water fresh. An upto date information on the survival rate at each developing stage is necessary.

Larval feeding

Suitable and high quality micro-algae and correct feeding rates are important for successful rearing. As the larvae progress in development the alimentary canal is well formed and the larvae must be given diet immediately. The effectiveness of the various micro-algae were tried. Better growth rate was obtained when fed on the micro-alga *Isochrysis galbana*. With this micro-alga the mortality rate was also found less. The larvae require different quantities of diet during different developmental stages. In general 20,000 to 30,000 cells/ml in the rearing tank water is maintained. The micro alga *Isochrysis galbana* cultured usually has a concentration of 80,000 cells/ml. When the bloom is good it reaches one million mark. *Pentactula* larvae settle down to the bottom of the tank. *Sargassum* is made into a fine paste and sieved through 40 μ . This settles down to the bottom of the tank. The *pentactula* larvae feed on the matter settled at the bottom of the tank. The culture of micro-algae for feeding of larvae of marine cultivable crustaceans, molluscs, fishes and holothurians have been elaborately given by Gopinathan in one of the chapters in this Bulletin.

Juvenile rearing

The juveniles when once they settle down to the bottom of the tank need food. They can no longer take planktonic food. If proper food is not available at the bottom of the tank they die. The juveniles are found to thrive and grow well on the algal extract of *Sargassum* spp. The seaweed *Sargassum* sp. is collected from the wild and first they are cut into small pieces. Then they are put in the mixer and made into fine paste by adding sufficient water. The fine paste is mixed with seawater and the same is filtered by using 40 μ

seive. This filtrate is now added to the tanks with juveniles. The extract soon settles down to the bottom of the tank and forms a fine film. The juveniles feed well on this extract settle down. In one tonne tank having an area of 2 m², 20,000 juveniles can be stocked.

The water in the tank is daily changed and the feed is given in the morning. Once in four days the juveniles are removed by using a siphon. Those which stick to the surface of the tank are gently removed with the help of a fine brush. It is observed that all the juveniles do not grow at the same rate though they belong to the same brood. Some of them grow faster than others and they are known as *shooters*. These are separated with the help of a brush and reared separately in 100 lt tank. When they reach a length of 10 mm, fine mud is also added to the tank. The *shooters* are regularly removed while rearing the juveniles. When they reach a length of 25 mm, formulated feed is also added to the diet of the juveniles. The fine mud provided for the juveniles is also changed when the juveniles are taken out to clean the tank (Pl. II A).

Farming

The sea-cucumbers are slow growing animals and they live for a number of years. It is expensive to maintain them in the hatchery till they reach marketable size. Therefore they are transferred to the sea when they reach a length of 20 mm. The juveniles were farmed in old one tonne tank, rectangular cage, velon screen cage, netlon cage and cement ring (Pl. II).

Old discarded one tonne tanks are used for farming the juveniles. These tanks are 2 m in length, 1 m in breadth and 50 cm in height. The tanks are taken to a suitable spot and filled with seawater to settle it to the bottom. Then the tank is fixed with the help of four casurina poles to keep it in the same position. Fine mud is provided in the tank to a height of 50 mm for the juveniles to feed and also to take shelter. The tank is covered by velon screen to prevent the entry of undesirable organisms into the tank.

The rectangular cage (Pl. II B) is made of iron rods of 7 mm diameter. They are 1 m long and 60 cm wide and 15 cm high. They are in the form of rectangular boxes with lid. On the outer side

of the cage nylon rope of 2 mm thickness is knotted to the frame. The distance between two knots is 30 mm. The cage is lined with fine velon screen inside to prevent the sand or mud going out. The cage is fixed to the bottom of the sea at a depth of 2 m with the help of four casurina poles.

The velon screen cage (Pl. II C) was 2 m² in area. It was made of velon screen of 4 mm mesh to allow the free flow of water. The length and breadth of the cage was 2 m and 1 m respectively. The height of the velon screen cage was 2 m. The cage was fixed at a depth of 2 m on an algal bed. The bottom of the cage also has velon screen for easy and total retrieval of the juveniles. The cage was fixed to the ground by four poles one at each corner of the cage. The cage is further strengthened by four more poles at the middle of four sides to keep the cage in position during high gales. To keep the bottom of the velon screen cage stable four big stones were kept at four corners. After the juveniles were stocked the top is covered by velon screen and stitched so that fish and crabs may not enter the cage. Before stocking the juveniles, sufficient quantities of mud is put inside the cage which serves as food for the juveniles.

Netlon cage (Pl. II D) is cylindrical in shape with an area of 1.65 sq. m. Diameter of the cage is 1.5 m and the height of the same is 1.3 m. The mesh size is 5 mm. The netlon cage is erected in the sea at a depth of 1 m. The cage is fixed to the bottom with the help of four stout casurina poles. The top of the cage is covered by velon screen to prevent entry of other organisms. Every week during low tide two buckets of mud is put into the cage and this serves as food for the juveniles. The juveniles are examined every month to find out the mortality and also the increase in average weight of the juveniles.

Cement rings used in the construction of wells are used to grow the juveniles. The diameter of the cement rings was 75 cm and 122 cm. The cement ring is covered by velon screen.

The space in the rectangular cage is limited and therefore only small number of juveniles can be reared. The netlon material does not last long in the sea. Netlon with mesh 4 mm becomes brittle and does not last for more than three months.

Velon screen cages get blocked, because of the small mesh size. Therefore this has to be periodically brushed. Cement rings last for a very long time in the sea, but the number that can be stocked is again limited. Larger rings cannot be handled due to the weight.

Economics

A tentative expenditure both capital and recurring, investments and economics of sea-cucumber culture is given below :

Hatchery seed production

Total number of tanks required	: 6 (1 tonne capacity)
Stocking rate of auricularia larvae	: 3.75 lakhs/1 tonne tank
Total auricularia stocked in one run	: 1 million
Expected percentage production of juveniles (10%)	: 1 lakh
Survival of juvenile at the end of 18 months in the sea	: 40%
Net production of harvestable sea-cucumbers	: 40,000

Capital expenditure

	Rs.
A. Building and tanks	
Hatchery shed with light roofing (30 m 10 m)	1,00,000
Room for Generator/Compressor (27 sq. m @ Rs. 500/- per sq. m.)	13,500
Seawater sump 14 sq. m @ Rs. 750/- per sq. m	10,500
Sedimentation tank 8.4 sq. m @ Rs. 750/- per sq. m.	6,300
Filter bed 4.5 sq. m @ Rs. 750/- per sq. m	3,375
Pump house 14.6 sq. m @ Rs. 750/- per sq. m.	10,950
Overhead tank - 10,000 lt capacity	50,000
Total	1,94,625
B. Fibreglass tanks	
1 tonne capacity Broodstock/ larval/juvenile FRP tank - 6 nos @ Rs. 5000/-	30,000
100 lt capacity spawning FRP tank 4 nos @ Rs. 1000/-	4,000
200 lt capacity mixed culture FRP tank 1 no @ Rs. 1000/-	1,000
Total	35,000

C. Major equipments

10 KVA Generator - 1 no	50,000
Air Compressor - 1 no	10,000
7.5 HP Electric pump - 1 no	15,000
1.0 HP Electric pump - 2 nos	7,000
Microscope, pH meter, Salinometer	15,000
Chemical balance	5,000
Furniture	25,000
ECE Controller, Silica cased immersion heater, jumo thermometer	4,000
Air Conditioner - 2 nos	40,000
Total	1,71,000
Total capital cost (A+B+C)	4,00,625

Recurring expenditure

A. Interest	
On Rs. 4,00,625 @ 15%	60,000
B. Depreciation	
On building and fibreglass tanks @ 5%	7,000
On equipment @ 10%	17,000
C. Salaries	
One Technician @ Rs. 2000/- per month for 18 months	36,000
Two helpers @ Rs. 500/- per month for 18 months	18,000
D. Contingencies	
Plastic ware, flexible PVC hoses, glassware bolting silk, etc.	5,000
Energy cost (Electricity and Diesel)	15,000
Chemicals	2,000
Other contingencies	5,000
E. Maintenance	5,000
F. Annual lease for land	3,000
Total recurring expenditure	1,73,000

Realisation of profitability of sea-cucumber produced

Total Sea-cucumber production	40,000 Nos
Cost of each Sea-cucumber	Rs. 20/-
Total amount realised (40,000 x 20)	Rs. 8,00,000
Less: Non-recurring and recurring expenditure (Rs. 4,00,625 + Rs. 1,73,000)	Rs. 5,73,625
Net Profit	Rs. 2,26,375

List of equipments required

<i>Equipment/facility</i>	<i>Quantity required</i>
Generator (10 KVA)	1
Air compressor 200 220 1420 RPM 50 HZ SI Rating output 0.75 KW AMP 73	1
Fibreglass rectangular Broodstock/larval juvenile rearing tanks (1000 lt capacity)	6
Fibreglass rectangular (100 lt capacity) spawning tank	4
Fibreglass tank rectangular (200 lt capacity) mixed culture tank	2
Binocular microscope	1
Mixie	1
ECE Controller	1
Jumo thermometer (0-50°C) (For thermal stimulation)	1
Silica cased immersion heater	1
Air-conditioner 2 tonne capacity	2
pH meter	1
Saline-refractometer (Temperature compensated) (0-50°C)	1
Chemical balance	1
Thermometer (0-50°C)	3
<i>Laboratory glassware</i>	
Beaker 10,000 ml	6
Beaker 5,000 ml	6
Beaker 3,000 ml	4
Beaker 1,000 ml	6
Beaker 500 ml	6
Beaker 250 ml	6
Conical flasks 250 ml	5
Oxygen bottles 125 ml	10
Volumetric pipettes (assorted sizes)	10
Burettes (10 ml)	2
Pestridishes (150 mm dia)	2
Embryo cups (50x50 mm)	6
Micro slides with cavity	2
Micro slides (in box)	1
Cover slips (in box)	2
Plankton counting chamber 1 ml capacity	2
<i>Plasticware</i>	
Plastic buckets (15 lt)	6
(5 lt)	6
(3 lt)	6
Basins (20 lt) capacity	6
Polyethylene flexible hoses (20 mm dia)	10 m

PVC Pipes (150 mm dia for sieves)	6
Polythene sheets (for mixed algal culture)	10 m
Bolting silk cloth	40 microns 1 m
	80 microns 1 m
	140 microns 1 m
	180 microns 1 m
	200 microns 1 m
Volen screen	1 mm mesh 30 m
	4 mm mesh 30 m
Tank cover cloth (Black)	30 m
Nylon rope (2 mm)	10 kg
Nylon rope (5 mm)	10 kg
Casurina pole (3 m length)	50 nos
Sea water draining distribution gride made of 50 mm and 25 mm grid	
PVC Pipelines and valves	as required
Aeration grid made of 25 mm rigid PVC pipelines with copper nozzles, 5 mm polythene tubes, plastic 'T' joints and regulators and diffuser stones	as required

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