

APPLIED ZOOLOGY

PEARL CULTURE

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1. INTRODUCTION:

Pearls are highly esteemed biological gems having smooth, lustrous and variously coloured deposits (nacre) around a grain of sand or other foreign particles in the shells of certain marine oysters and freshwater mussels. The nacreous deposit is composed of 82 - 86% calcium carbonate (aragonite crystals), 2-4% water and 10-14% organic substance conchiolin, which impart shining to the pearls. Pearls are three types: natural, cultured and artificial. Natural pearl is formed when a foreign particle viz., piece of sand, animalcule, small parasite, algae etc. enters the body of certain oysters/ or mussels by chance, and is not rejected out easily. Oysters or mussels start depositing a shiny coating on the particles layer by layer that ultimately results in formation of pearls. While the cultured pearls are produced by inducing oysters to deposit nacre around a surgically implanted foreign body of a particular shape and size into some identified locations. The artificial pearls are made of plastics, marbles, glass, talc, ivory or shell beads etc. They are painted with pearl essence, which is a mixture of enamel and silvery extract of fish scales (iridescent guanine -C₆H₅ON₅).

Natural pearls are an extremely difficult to procure. One has to slaughter thousands of pearl oysters/mussels in order to get just one good quality natural pearl. In the best pearl beds inside the Gulf of California, called "*placer*" in Spanish, the incidence of natural pearls was said to be in the range of 5 to 12%, meaning that for every 100 killed oysters one would find only 5 to 12 pearls, but only a 30% would be of good quality. So, out of those possible 12 pearls, only 3.6 good pearls could be obtained. This intensive fishing effort had adverse effects on the viability of the natural pearl oyster populations throughout the world. Moreover, over exploitation and water pollution has further added difficulty in obtaining natural pearls.

Pearls are generally used for decorative and jewelry purposes. They have also been used as medicine to cure insanity. They are powdered for pharmaceutical preparations like potions, balms, and salves to treat a wide variety of ailments. Other conditions for which pearls were prescribed for treatments include memory loss, insomnia, asthma, jaundice, liver ailments, heart problems, infertility and also in insect or snake bites.

For millennia, pearl have fascinated humanity the world over. In ancient times, the demand was met by natural production. However, to meet the rising demand of pearls in the modern world, entrepreneurs and researchers resorted production of pearls by culturing pearl producing oysters and mussels. Although first attempt was made by Chinese to produce cultured pearls, the Japanese became champions of producing cultured pearls successfully and Kokichi Mikimoto is credited for it. The truth behind the mystery of pearl formation in nature was unraveled in 1907 by Tokichi Nishikawa who gave the "pearl sac theory". According to him, the pearl secreting cells of the mantle migrate into the body of the oyster under the stimulus of a foreign particle and by series of cell division form a pearl-sac around the foreign body. The pearl-sac in turn secretes the nacre, which is deposited over the foreign body forming a 'natural pearl' in course of time. The subsequent scientists exploited this natural phenomenon by inserting a foreign particle called 'nucleus' along with the secretary mantle piece into the oyster/ mussel. After grafting the nucleus, oyster/ mussel are cultured under controlled conditions. Formation of a mature pearl takes about 12-24 months depending on the size of nucleus.

The technology has now spread from Japan to throughout the world, and with suitable modifications in the technology many countries are producing cultured pearls of high quality. India is blessed with number of molluscan species, which produce gem quality pearl both from marine and freshwater environment.

2. TAXONOMY AND DISTRIBUTION OF OYSTERS AND MUSSELS:

The pearl producing oysters and mussels are molluscan bivalves having a protective exoskeleton in the form of two calcareous valves united by an elastic hinge ligament. The true pearl oyster belongs to the order Dysodonta and the family Pteriidae. Members of this family have a straight hinge with 1-2 small tooth-like thickenings, a cavity below the anterior angle for the

byssus, and scaly surface on the outer shell. This family includes two genus: *Pinctada* and *Pteria*. In *Pinctada* spp. the hinge is long and straight. The long axis of the shell is at right angle to the hinge and the left valve is slightly deeper than the right. There is a byssal notch on each valve at the base of the anterior ear. In *Pteria* spp. (*penguin* & *colymbus*) the shell width is much longer than the height and the hinge angle is prominent and pronounced. Classification of important pearl oysters is given below in table.

Kingdom	Phylum	Class	Order	Family	Genus	Species
Animilia	Mollusca	Bivalvia	Dysodonta	Pteriidae	<i>Pinctada</i>	<i>maxima</i> <i>margaritifera</i> <i>fucata</i>
					<i>Pteria</i>	<i>penguin</i> <i>colymbus</i>

Pinctada margaritifera



Pinctada margaritifera

The pearl oysters occur in almost all the seas of the tropical and subtropical regions of the world. Although 28 species of pearl oysters have been identified, only three species have been found to produce pearls of gem quality having commercial value. They are *P. maxima* (Jameson), *P. margaritifera* (Linnaeus) and *P. fucata* (Gould). *P. maxima*, commonly known as the ‘white lip’ or ‘silver lip’ or ‘gold-lip’ pearl oyster and is the largest of all the pearl oysters. It is prized for both its shells and the gold coloured and the white pearls it produces. It occurs in the Southern seas, Australia, Burma, Thailand, Indonesia, Philippines and Papua New Guinea at depths ranging from low tide level to 80m. Along the Indian coast, six species of pearl oysters, viz: *Pinctada fucata* (Gould), *P. margaritifera* (Linnaeus), *P. chemnitzii* (Philippi), *P. sugillata* (Reeve), *P. anomoides* (Reeve) and *P. atropurpurea* (Dunker) have been reported to occur.

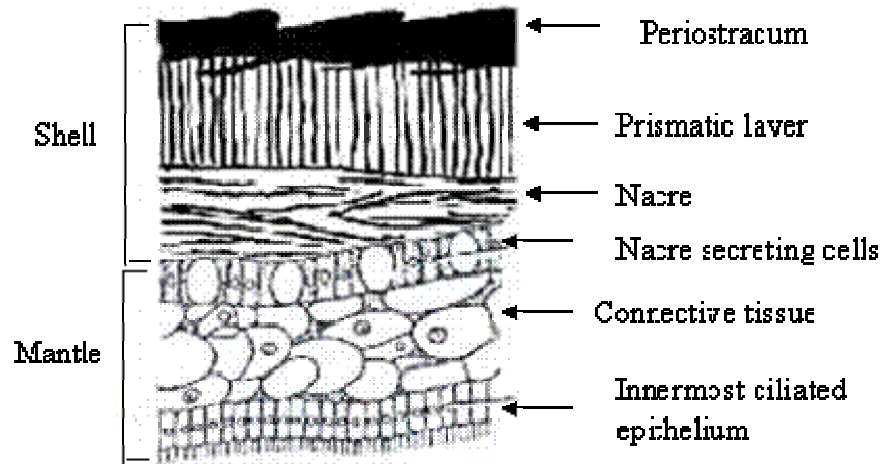
The freshwater pearl producing mussel belongs to genus *Lamellidens* (Family Unionidae) and *Parreysia* (Family Amblemidae). The *Lamellidens marginalis*, *L. corrianus* and *Parreysia corrugata* are the most important species, which possess good ‘mother of pearl’ in the shell.

3. BIVALVE SHELL STRUCTURE AND NACRE SECRETION:

Shell is composed of three layers: the first outer most is a thin horny layer of organic matter conchiolin, is known as ‘periostracum’, the second middle later is made of column of crystalline calcium carbonate separated by conchiolin and is called as ‘prismatic layer’, and the third innermost layer is iridescent nacre, which is formed of many thin and alternating layers of

calcium carbonate and conchiolin. The first two layers are secreted by edge of the mantle, whereas the innermost layer by its whole surface.

T.S. of oyster Shell Periostracum Prismatic layer Nacre Nacre secreting cells Connective tissue
Innermost ciliated epithelium Mantle Shell



T.S of oyster Shell

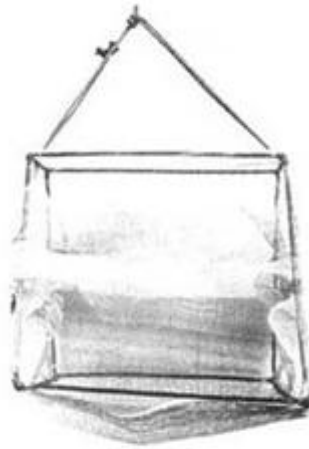
4. MARINE PEARL PRODUCTION:

4.1. Oyster Culture

To produce cultured pearls, the first basic need is the availability of good quality healthy marine oysters or freshwater mussels for nucleus grafting. They are obtained either from their natural habitat or raised from juvenile oyster (called as 'spat') collected from nature or hatchery. Following grafting, oysters are required to be reared in farms or in natural habitat under controlled conditions. Therefore, a good knowledge of oyster farming is important. Farming of pearl oysters can be done either in a bay or in a lagoon or in coastal waters or in onshore tanks, where the environmental conditions are conducive for their growth. In pearl culture establishment, farming activities go round the year. Therefore, the establishment must have a good laboratory of oyster surgery, pearl collection center, pearl grading and processing unit, farm stores, cage cleaning yard, mechanical workshop and boat *etc.* for transport of workers, oysters and farm materials.

Pearl oysters are collected either from the marine bed or raised from their juveniles, spat, obtained from hatchery or natural habitat. The pearl oysters occur in large number on the submerged rocky substrates known as 'paars' lying at depths of 12 to 25m. Skin and SCUBA (Self Contained Underwater Breathing Apparatus) divers collect pearl oysters from these beds. In the Gulf of Kutch, the pearl oysters are found sporadically on the inter-tidal reefs known as 'Khaddas'. Collection of oysters is done by hand picking. Since, the collection of wild oysters or their spats is highly unpredictable. Therefore, hatchery source is more dependable and ensures a sustained supply of oysters for pearl culture throughout the year. In India, the pearl oyster hatchery of Central Marine Fisheries Research Institute (CMFRI) has produced several million seeds of oysters, spat, over the last several years.

The juvenile oysters are reared in net-cages of velon screen bags whose sides are stretched with a steel rod in the form of a prism. The mesh size of the screen depends on the size of juveniles to be reared. The mouth of the bag is tied with a synthetic twine, which facilitates opening or closing when required. To provide further protection from predators, the bags are placed in old nylon fish net bags.

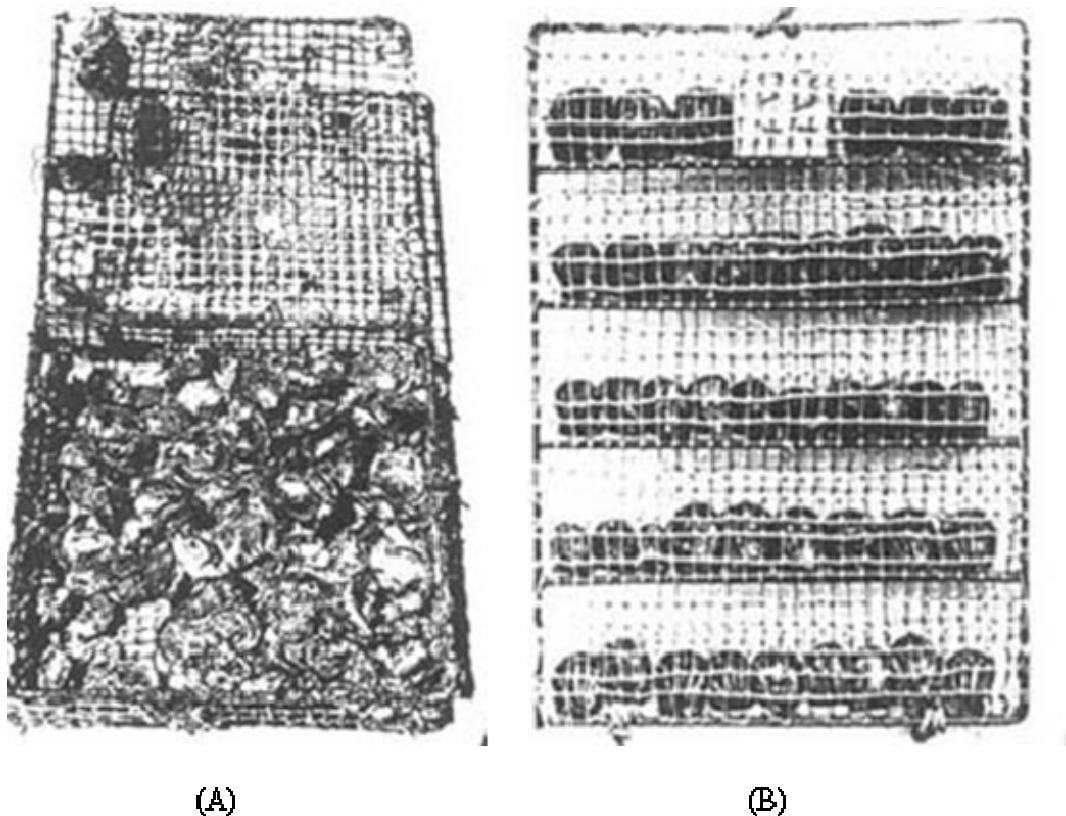


Velon Net-bag for Spat Rearing (adapted from FAO bulletin)

The spat are reared in the hatchery for about two months, by this time they attain a size of 3 mm or more. They are then transferred to the farm in velon screen net-cages with a mesh size of 400 μ m. Mortality may occur if spat measuring less than 3 mm are transported. Spat growth is monitored carefully and the net-cages are cleaned or changed whenever necessary. The oyster spat attains an average size of 40–45mm and 25g body weight in 12 months, which is the most suitable size for nucleus implantation. The survival of transplanted spat in the farm is about 30% by the end of 12 months.

Mother pearl oysters, collected either from natural bed or spat raised, are placed in box-cages with nylon-woven coverings. The mesh size varies with the size of the oysters to be reared. The frames of the cages are made up of mild steel rods, coated with anticorrosive paints or coaltar. Each box cage has vertical meshed shelves, generally five in number. The oysters are arranged in rows in the shelves.

(A) A box-cage containing pearl oysters and (B) A frame net-cage with oysters (adapted from FAO bulletin)



(A) A box-cage containing pearl oysters and (B) A frame net-cage with oysters
(adapted from FAO bulletin)

Such oyster carrying box-cages are cultured at farm using any of the following culture systems:

- a. *Raft System*: This method is most commonly used for oyster farming in sheltered bay or in open areas. The raft size of 6x5 m is most suitable. Rafts are usually constructed with teak, casuarinas or eucalyptus poles of chosen length with the base of the pole having 10 cm diameter tapering to 6 cm diameter at the tip. These poles are lashed with coir ropes. Four floats are attached to the four corners of the raft for buoyancy. Floats are generally empty diesel drums with fibreglass coating or steel barrels painted with anticorrosive paints or FRP styrofoam. Rafts are moored with two anchors at opposite sides with tested quality chains and their position is decided according to the prevalent wind direction at the site. The oyster carrying cages are suspended from the raft at 5m depths in the sea.

Raft with Steel Barrel Floats (adapted from FAO bulletin)

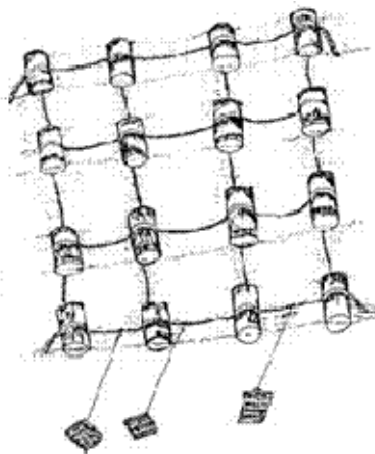


b. Rack system: This method is employed only in shallow and calm seas of 2 to 4 m depths. The general size of the rack is 10x10 m. Rack system is a fixed structure of teak or eucalyptus poles of chosen length, which are driven vertically into the sea bottom in rows at 1 m apart. Cross and horizontal poles are arranged as per requirements on the top of the poles and lashed with coir rope at a convenient height of 0.5m above the water level so that the rack thus erected remains always above the water. Generally a total of 400 box-cages holding oysters are suspended from such racks.

c. On bottom System: In the sea with hard or rocky bed, pearl oysters are cultured by placing them in box-type cages and arranged on the sea bottom in rows.

d. Onshore Tank System: Oysters are also reared in concrete tanks filled with clean seawater. The tanks are generally of 50t capacity. These tanks accommodate about 1250 mother oysters/seeded oysters. Tank culture has an added advantage over other open sea culture systems, as survival rate in this system is high due to the absence of predation and better control of environment. Also the oysters in tank system grow faster.

Long Line System (adapted from FAO bulletin)



Long Line System (adapted from FAO bulletin)

e. Long Lines System: In this method, a series of spherical or cylindrical floats are attached by synthetic rope or chain at uniform interval. The line is generally 20m long. The ropes are moored with anchors. The oyster cages are suspended from the ropes. This system is good for open sea conditions and is practiced in certain parts of the world.

g. Underwater Platform System: In this method, a hole is drilled near the hinge of the pearl oyster. A small thread is put through the hole, which is then tied to a straw rope coated with tar. The straw ropes are hanged from a raft. This method is practiced in French Polynesia for

farming black-lip pearl oyster in deeper lagoons. Oysters in strings are suspended from these platforms.

4.2. Requirements of Culture Conditions:

Production of pearl oysters depends primarily on the environment in which they are reared. The physico-chemical characteristics like temperature, salinity, pH and nutrients of the aquatic environment influence their life processes such as respiration, nutrition and growth, osmoregulation and reproduction greatly. The salinity of about 30ppt and pH ranging from 7-8 are optimum for oyster culture. The best temperature range is 25-31°C. A slightly higher temperature facilitates the growth of oysters. The depth of suspending box-cages from rafts or racks is also critical in providing optimum conditions. The depth of 10m in open sea and 2-4m in sheltered bay is most suitable. The proper depth is required to avoid strong sunlight exposure to oysters, as it induces nacre-producing cells to secrete calcite crystals that form the prismatic layers on nucleus resulting in poor quality pearls. Sea bottom should be rocky or gravelly not muddy or sandy, as the silt may choke gills of oysters. Mild water current in culture areas is essential for proper oxygenation of water and also to bring fresh plankton upon which oysters feed. It also helps in removing the metabolic wastes and faecal products.

Water must be rich in food items and nutrients. The pearl oysters are filter feeders and utilize the available micro - algae, diatoms and other planktons in the waters. Oysters derive 'conchiolin' from the nitrogen substance of the plankton. The organic matter and calcium are directly absorbed from the seawater through food. The optimum amount of trace elements are also essential in the water, as their presence influences the colour of nacre. Gold and cream coloured pearls contain more copper and silver, whereas, skin and pink coloured have more sodium and zinc. Therefore, oyster farms must be established in such an areas where such above-mentioned favourable water conditions are met. In case of adverse situations, culture system should be shifted to favourable site.

4.3. Maintenance of Oyster Farm:

Farm maintenance requires regular cleaning of cages and oysters from borers (polychaetes and sponges) and foulers (barnacles, amphipods and polychaetes). A gastropod, *Cymatium cingulatus* is a predator capable of causing heavy mortality of farm oysters, if not eradicated. Cages must be looked after every 100 days. The temperature, pH, salinity, dissolved oxygen, primary productions of food item are to be regularly monitored. Pearl oysters can tolerate adverse environmental conditions for a short period, but, if these continue, the rafts/ racks should be shifted to the areas of favourable conditions. In recent years, fairly good amount of industrial effluents are discharged by rivers into the seas, which adversely affect their survival and life processes, hence, oyster farms should be established away from such discharges.

4.4. Production of Pearl from Oysters:

The technology essentially involves the introduction of the secretory mantle tissue (taken from the donor oyster) along with an artificial bead (nucleus) into the gonads of recipient oyster in proper orientation through skillful surgery. This event is termed as implantation or grafting. The epithelium of the mantle piece undergoes cell division and spread over the implanted nucleus forming the pearl sac. The epithelial cells of the pearl sac secrete and deposit the nacre or mother of pearl over the nucleus in concentric layers one after the other. A pearl oyster like the *Pinctada mazatlanica* secretes 3 or 4 concentric layers of nacre each day. These are thin layers and measure an average about one micron. Therefore, pearl grows slowly and attains its 'gem' quality in 8-12 months.

Oysters are cultured for pearl production in two phases, termed as (i) mother oyster culture and (ii) post-operative culture. Mother oyster culture refers the farming of oysters from the time they are brought to the farm till they are used for nucleus implantation. The post-operative culture refers to rearing of oysters from the day of implantation upto the day of pearl harvesting.

The mother oyster culture phase includes:

1. Selection of healthy oysters of suitable sizes
2. Conditioning
3. Preparation of graft tissues
4. Surgical implantation
5. Convalescence

Post-operative phase involves:

1. Culture of implanted oysters
2. Pearl harvesting
3. Cleaning, grading and processing of pearls.

a. Selection of Oysters:

On the farm, oysters are selected based on their weight, size, age, reproductive stage and overall health. Oysters of 1.5-2 years age and attaining a weight of 25g and size of about 40-45 mm are the ideal for implantation, while 20g oysters can also be considered for implantation of smaller size nuclei, i.e. 2–3mm in diameter. Reproductively spent oysters are selected for implantation, because the fully-grown gonads block the visibility of the implantation site and, hence the proper orientation of the mantle piece and nucleus cannot be ensured. Therefore, oysters in the immediate post-spawning recovery phase or those in the early phase of gametogenesis are selected. Further, oysters should be free from ectoparasites such as polychaete blisters, sponge borings and trematodes infection. Selected oysters are thoroughly cleaned and all the fouling organisms are carefully removed. Such cleaned oysters are transferred from the farm to the laboratory for implantation.

b. Conditioning:

For surgery the gap of 1 to 1.5cm is required so that surgical appliances can be used easily and nucleus along with mantle piece could be easily placed at right place. However, oysters when taken out of water, they close the shell valves very tightly, and the forceful opening of their valves is fatal for them. Under such circumstances, oysters are subjected to narcotization that causes natural opening of valves. For this, oysters are arranged first in a plastic trough with their hinge pointing downward, and then sea-water having menthol is gently poured in to submerge them. A little amount of menthol crystals are also sprinkled over the seawater and then the trough is covered. In about 60–90 minutes, the oysters are narcotized and relax their adductor muscles resulting in opening of valves. Immediately after opening of valves a small wooden peg (speculum) is inserted to keep valves opened. This process is termed as ‘conditioning’. The water temperature influences the narcotizing response, however once narcotized the oysters become almost non-responsive to touch. The conditioned oysters are usually operated within the next 10 to 15 minutes as prolonged exposure to menthol may cause swelling of tissues, copious secretion of mucus and mortality may occur. Therefore, the pearl oysters should be conditioned batch-wise. The conditioned oysters are cleaned with fresh seawater individually, placed in separate plastic tubs, and are readily operated for implantation. The donor oysters, from which mantle pieces are prepared, are not subjected to any conditioning process.

c. Graft Tissue Preparation:

The mantle pieces are obtained from the pallial mantle tissue taken from the donor oysters. A healthy donor oyster is selected for this purpose from the stock. To obtain pallial mantle tissue, a sharp knife is inserted in between the valves of the donor oyster upto the adductor muscle and the latter is cut vertically starting from the posterior margin tracing up to the anterior margin without damaging the mantle tissue. A strip of 5cm long and 0.5cm wide mantle tissue is cut and lifted gently. The mantle tissue is then stretched and layered on a soft, clean, moist wooden block in such a way that the inner epithelium of the mantle faces upward. The mucus and dirt, if any, of the mantle is wiped out gently with the help of wet sponge or blunt end of the scalpel. With the graft-cutting knife, the marginal thickened ends of mantle are cut away and, thus a long ribbon of 5cm length is obtained from the pallial zone of the mantle. Holding one end, the

mantle ribbon is lifted to reverse the side (top to bottom) so that outer epithelium faces now upward. Mucus and dirt are removed softly without causing damage to the outer epithelial layer. About 20-25 pieces of about 2-3mm square are cut from a ribbon. The graft tissues should be kept wet with sterilized filtered water all the time during preparation. Application of a weak solution of Azumin/eosin over it helps to keep cells alive for a longer duration. The graft tissues should be used within 15 minutes of preparation.

d. Nucleus:

The nucleus is another major requirement in pearl production operation, because when only a piece of mantle is grafted into the gonad of an oyster, it results in the production of irregular pearl. So, in order to get larger and spherical pearl in a short period, a spherical shell known as 'nucleus' is inserted along with the graft tissue.

The nucleus (bead) is manufactured in Japan from the thick shell of freshwater mussel, pig-stone, washboard, and dove. Japanese has a close guard on the technology of the production fine quality nuclei to hold their monopoly on it. Pearl culturing nations have to import nuclei from Japan. However, in recent past, some attempts have been made by many other countries including India to manufacture the nucleus of good quality, but still Japanese have an upper hand. In India, chank shells have been cut and processed into beads and used as nucleus for pearl culture. The general size of nuclei ranges from 2-7mm in diameter. The hardness and specific gravity of the nuclei are nearly identical with that of the deposited nacre.

e. Surgery for Implantation:

A successful surgery is achieved with the help of special surgical appliances that include oyster stand, shell speculum, incising-cum-grafting needle, nucleus insertion needle, graft cutting knife, spatula, needle hook, forceps, knife and scissors *etc.* The other instruments needed are graft cutting wooden blocks, wooden pegs, camel hairbrush, trays, rubber sponge towels, glass beakers *etc.* All these appliances are well sterilized before use.

The conditioned oyster with the speculum is first mounted correctly between two plates of the oysters stand. The tip of the foot is hooked with the needle on the left hand so that the base of foot is slightly elevated. The needles are held in position until the operation is completed. A sharp incision is made at the base of the foot on the right side with the help of oval knife end of the incision-cum-grafting needle. Through this opening, a subcutaneous passage is made in the gonadal tissue upto the site of implantation. A piece of graft tissue is now picked with the tip of the needle and inserted gently in such a way that the outer epithelium of the graft tissue faces the passage. Following this, a nucleus is implanted through the same passage using nucleus-implanting needle. Nucleus should be in the direct contact of outer epithelium of the mantle piece grafted. With the nucleus-implanting needle two margins of the cut are brought in close contact and smoothened. Speculum from the operated oyster is, then, withdrawn, and oyster is transferred in clean seawater.

In oysters, most suitable site for the nucleus implantation is the gonad, particularly in its ventral portion. Single or double implantation is very common. In the single implantation, a nucleus is implanted close to the turn of intestinal loop; while in double implantation, the second nucleus is grafted close to the hepato-pancreas. In certain cases, however, multiple implantations are also carried out when large numbers of small pearls of about 2–3 mm are required. Large diameter nucleus, in the range of 6–7 mm, is generally used in single implantation. In double implantation, one large (6 mm) and one small (4 mm) nucleus are used for each oyster.

f. Convalescence:

Convalescence refers placing of freshly implanted oysters in the fresh seawater with mild circulation for two to three days in the laboratory under close observation. Within thirty minutes, the operated oysters slowly reopen their valves and resume the filtering activity. The mild water circulation is required to provide well-oxygenated water and also to remove the excreta rapidly from the trough. In 2-3 days, the incision wound heals completely. The implanted dead oysters are removed and healthy ones are shifted to natural environment.

However, in Japan, freshly operated oysters are hanged in deep and calm seawater for a period of 2–3 weeks, by which time they recuperate fully.

g. Post-operative Culture:

After convalescence, implanted pearl oysters are examined individually by fluoroscopy method to check the condition of the inserted nucleus. Only those with the nucleus in the proper position are cultured further for pearl production. They are placed in box-cages and are transferred to farms to be suspended from raft or racks or long line at the depth of 3-5m in the sea of high phytoplankton production. A box-cage measuring 40x40x15cm can accommodate 125 oysters of 30-35mm, 100 of 45-55mm, 75 of 55-60mm and about 50 of larger size, but to avoid the overcrowding, it is always recommended to place only 50-75 operated oysters of 40-45mm. Basically, there is no difference between pre-operative and post-operative culturing practices except the density of the oysters in the box-cages. During the post-operation rearing period, over-crowding may cause adverse effects such as production of low quality pearls, slow formation of the nacre layer, shell damage and physical stress, or even oyster mortality due to infectious diseases and parasites.

Once a month, cages and shells of the oysters are cleaned from predators and epifauna. They are cultured for four to eighteen months depending on the nucleus size inserted and desired final size of pearl. The deposition of nacre is faster in the tropical sea than in sub-tropical and temperate one.

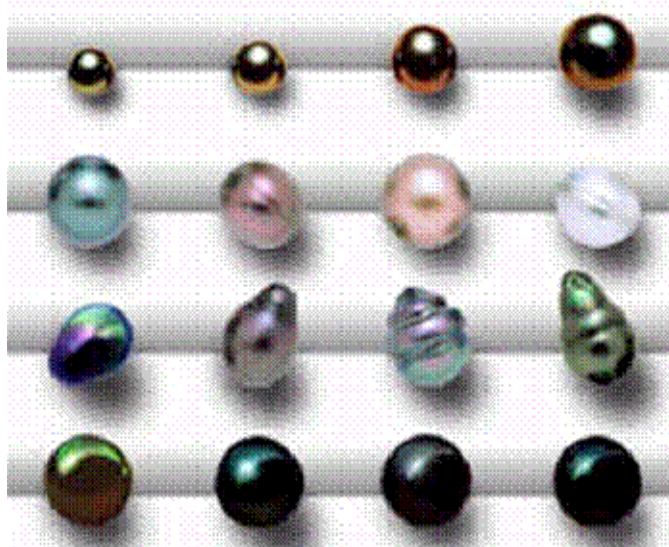
h. Harvesting of Pearls:

Harvesting of pearls from cultured oysters is carried out manually preferably in cold seasons. For procuring pearls, oysters are brought to the laboratory from farms. Then, their adductor muscle is cut, and pearls are removed by squeezing out the gonad. If the oyster is to be reused for implantation, adductor muscle is not cut and two valves are opened softly and pearl is extracted with the help of needle. The oysters are transferred to farm for re-implantation after recovery. In Japan, harvesting is also done with the help of instruments. The success of pearl production was initially 55.8%, but now it has been improved to 62.8% in case of single implantation and 68.3% in multiple implantations.

i. Cleaning, Grading and Processing of Pearls:

Harvested pearls are first washed with distilled water. Mucus on the surface of pearls are removed by placing them with powered salts mixed little water. After some time, pearls are taken out and washed with distilled water. The residual mucus on the surface of pearl, if any, is removed by rubbing with salts. Removal of complete mucus renders better luster. The pearls are, then sorted by size, shape, colour, luster, surface quality and thickness of nacre. A good quality cultured pearl should have at least 0.8mm of nacre. A pearl with nacre thickness of less than 0.8 mm is considered to be of poor quality.

Some of the pearls are perfectly round in shape and of outstanding colour and big size, while many are inferior, and some are valueless as gems. The pearls are classified into three grades: A, B and C. Approximately 37.6% of total pearl production in India is of 'A' grade, while grade B and C constitute 37.6 and 24.8% respectively. The color of the pearls is pink, white, black, yellow, cream, golden, blue and green. Black pearl is highly valued due to its rarity. Since, the pearls are biological product; it is rather difficult to find homogeneity or uniformity in size and quality. Some times, drilling is also done in pearls to make a hole with the help of a machine. Such drilled pearls are, and then bleached with hydrogen peroxide to improve its quality by removing organic impurities.



Pearls of different shape, size and colors

Collection and Selection of oysters

- ↓ Pre-operative conditioning
- ↓ Preparation of graft tissue (mantle pieces)
- ↓ Surgical implantation of nucleus and mantle piece
- ↓ Convalescence
- ↓ Rearing of implanted oyster
- ↓ Harvesting of pearl
- ↓ Cleaning, grading and processing of pearls

Flow chart for marine pearl oyster production



Lamellidens marginalis

5. FRESHWATER PEARL PRODUCTION

Large scale availability of wild stock of freshwater pearl mussels in easily accessible habitats, operational easiness in management of their farms, absence of natural fouling, boring and predatory organism in freshwater ponds and overall cost effectiveness of their culture make the production of pearls from freshwater mussel more advantageous than marine pearl culture. The technology of pearl production from oyster remains essentially same for the production of pearls from freshwater mussels as well. It differs only on certain points that are dealt at the appropriate places below. The freshwater pearl culture farming involves six major steps sequentially given below:

1. Collection of mussels
2. Pre-operative conditioning
3. Surgery
4. Convalescence
5. Culture of implanted mussels
6. Harvesting of pearls.

a. Collection of Mussels:

The healthy mussels are collected manually from the freshwater bodies and are transferred to the farm. Mussels generally live partly buried in the sand or mud in shallow marginal areas of the stagnant to slow flowing habitat like ponds, tanks, lakes, rivers and reservoirs. The collection of pearl mussels from the natural bed is not always dependable, owing to their irregular production and water pollution. To ensure the sustainable supply of pearl mussel, hatchery production of mussel's seed is much more reliable for culture throughout the year. Once the mussels are raised from such seed, they are selected for grafting by considering their age, weight, stage of sexual maturity and health. The mussels of 1.2 to 2 years in age and 25g or above in body weight are ideal for pearl culture. Some times, mussels of smaller size are also used for implantation. Mussels should be sexually spent and in resting phase.

b. Pre-operative Conditioning:

Like oysters, the collected mussels are also conditioned prior to implantation, but menthol is not used for conditioning of freshwater mussels. The healthy mussels are transferred to laboratory and are cleaned thoroughly first for 2 to 3 days with aged tap water. Then it is treated with limewater (7.5mg/l) for another 2-3 days, followed by 1% (v/v) sodium hypochlorite to make them completely free from any infection. In the last, mussels are again washed with aged tap water for another 2-3 days to ensure the removal of chemicals used in previous treatments. Finally, the mussels are given an immersion treatment in chloramphenicol (100 mg/l) for 24 h. Now, such treated mussels are kept in crowded condition in captivity at a stocking density of 1 mussel/ liter tap water. Such pre-operative overcrowding condition causes weakening of adductor muscles resulting in opening of valves. Speculum is placed immediately between valves after their opening.

c. Surgery

Procurement of mantle tissue from the donor mussel for preparation of grafts is achieved in a similar way as described in case of oysters. The beads or nuclei used are generally made-up from mollusc shell or other calcareous materials such as eggshell powder blended with suitable adhesive or stelon materials. Unlike oysters, implantations in the mussels are done very commonly at three sites viz., mantle cavity, mantle tissue and gonad.

i. Mantle Cavity Implantation:

Beads of 4-6 mm diameter are grafted into the mantle cavity region of mussel after opening their two valves (without causing injury to adductor muscle) and separating carefully the mantles of anterior sides from the shell using surgical appliances. Implantation can be achieved in mantle cavities of both the sides. After placing the beads at desired place, the gaps created for implantation are closed just by pushing the mantle onto the shell. This method is simple and

most successful. The pearl product is generally shell-attached. This is the reason that mussels are sacrificed for pearl harvesting in this method.

ii. Mantle Tissue Implantation:

In this method, implantations of graft and nuclei are done on recipient mussels by two ways viz., non-nucleated and nucleated. In the non-nucleated type, only the mantle piece is inserted into the pocket created at the inner side of posterior pallial mantle present at the ventral region of the mussel. In the nucleated method, a graft piece followed by a small nucleus (2mm diameter) is introduced in the pocket. In both the procedures care is taken so that graft or nucleus does not come out of the pocket. Implantations can be done at mantle of both valves. The pearl product on non-nucleated implantation method is irregular in shape, while it is small and round in case of nucleated method.

iii. Gonadal Implantation:

In this procedure, the labial palps and gills of the mussel are gently pushed up with spatula and then an incision is made at the edge of the gonad of the mussel, using specially made knife. Then, a graft is inserted into the gonad followed by nucleus (2-4mm diameter). Care is taken to ensure that nucleus is in close contact with the outer epithelial layer of the graft and the intestine is not cut during the surgery.

d. Convalescence:

Immediately after operation, the grafted mussels are placed in specially made nylon bags (two mussels per bag) with ventral side up in position. These bags are hanged at a depth of 0.2m in tanks made of ferro-cement or fiberglass reinforced plastics containing aged tap water for post operation care for ten days. During this period, immersion treatment with antibiotic like chloramphenicol enhances the survival of operated mussels and help in faster wound healing. Plankton and algae rich water should be added in tanks after 3-4 days. The operated mussels are examined daily for removal of dead mussels and the ones that reject the nucleus.

e. Culture of Implanted Mussels:

Following convalescence, the implanted mussels are cultured in the ponds for 12-18 months. The mussels are kept in nylon bags (two mussels per bag) and are suspended at one-meter depth from bamboo or PVC pipes in the ponds. The mussels are cultured at stocking density of 20,000-30,000/ha. Regular examination of mussels with removal of dead ones and cleaning of bags is required throughout the culture period. Submerged and floating plants are not allowed to grow, as they impede penetration of the light diminishing thereby the production of plankton in the pond water. Phytoplankton and zooplanktons are the important food items of mussels. They are strained together with other organic matters by the gills of the mussels and ingested as food. The plankton content of water can be increased through the application of organic or inorganic fertilizers. During dry months, water levels usually go down due to evaporation. Water loss should be made up by pumping water from the ground, river or other ponds.

Freshwater pearl mussel culture pond (adapted from CIFA, Bhubaneswar, bulletin)

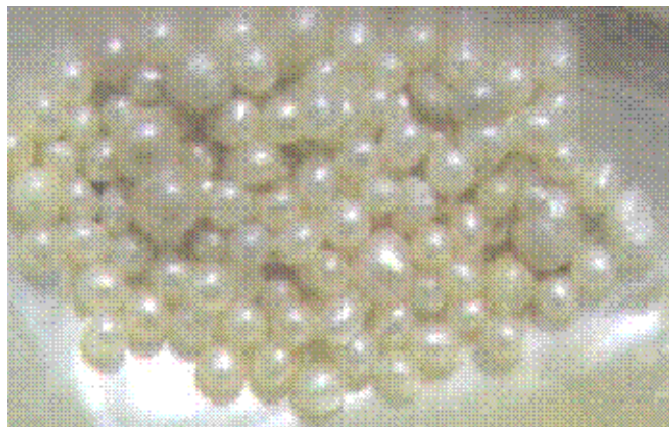


Freshwater pearl mussel culture pond (adapted from CIFA, Bhubaneswar, bulletin)

f. Pearl Harvesting and Processing

At the end of the culture period, the pearls are harvested surgically, for which mussels are brought to the laboratory. Each mussel is opened by cutting the adductor muscles, exposing the body to have clear visibility of gonads, mantle cavity or mantle tissue, and pearls are removed. The mussels are sacrificed in case of mantle cavity pearl production method. The pearls obtained are cleaned, graded and processed as described in case of oyster pearls.

Freshwater pearls



Freshwater pearls

POTENTIAL OF PEARL CULTURE IN INDIA:

India has great potential for pearl culture, as it is bestowed with along cost-line and of marine zone along south India and Andman and Nicobar islands. The seas around the Indian mainland are rich in *Pinctada fucata* in Gulf of Mannar, Mandapam, Tuticorin, Trivandrum etc. While sea of Andman and Nicobar islands has abundant *P. margaritifera*. Several sites at these islands have been identified as prospective pearl oyster beds.

The freshwater pearl mussels are also available abundantly in easily accessible natural water bodies like pond, river, lakes, reservoirs etc. India possesses rich biodiversity of freshwater mussel with over fifty species described all over the country out of which three species viz. *Lamellidens marginalis*, *L. corrianus* and *Parreysia corrugata* are found to possess high potential for pearl formation. The wider area of inland farming is also available in various regions of the country.

In India, Central Marine Fisheries Research Institute (CMFRI), Cochin and Central Institute of Freshwater Aquaculture (CIFA), Bhubaneswar, have developed and mastered the techniques for oyster and mussel pearl production successfully. Therefore, perfect indigenous techniques and abundant resources provide good scope for pearl culture in India. Though, pearl culture is a long term investment, a huge profit can be made in a successful culture operation, as pearls are still in high demand world over. India can join with private firms and technical experts to enhance the pearl production efficiency.