Hatchery protocols for seed production of cobia and pompano

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Introduction

Many Southeast Asian countries, European nations and Western countries like USA have developed commercial marine finfish hatchery technologies for many commercially important species namely groupers, salmon, tilapia, sea bass, sea bream, snappers, mullets, Chanos, etc. They have developed capacity to produce large and dependable quality of fish seeds which is the key for establishing reliable and sustainable marine finfish aquaculture sector. In India, technology for production of marine finfish seeds is in primitive stage except for sea bass. The Mandapam Regional Centre of the Central Marine Fisheries Research Institute (CMFRI) has developed hatchery technology for cobia, *Rachycentron canadum* during March 2010 for the first time in the country. Again the Centre has developed hatchery seed production technology for the silver pompano, *Trachinotus blochii* for the first time in the country. Both the technologies are standardised and hence the CMFRI has entered into agreements with interested entrepreneurs and farmers for dissemination of technologies for development of cobia and silver pompano aquaculture sector in our country.

The cost-effective hatchery technologies developed by the Mandapam Regional Centre of the CMFRI comprise

1. Induced spawning protocols
2. Appropriate live feeds for larval rearing,
3. Commercial-scale protocols for larval rearing,
4. Nursery and grow-out culture protocols

The larval rearing procedures of cobia and pompano are described below

Egg harvest

The fertilized eggs of cobia and pompano float and are scooped gently using 500 μm net. To minimise collection of poor-quality eggs, which usually float deeper in the water, it is advisable to collect only the eggs

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which float at the water surface. Therefore, aeration can be switched off allowing the unfertilized / dead eggs to settle at the bottom of the tank. The floating layer of eggs thicker than one cm should be avoided. A thicker layer may reduce oxygen supply to the eggs, leading to possible anoxia after a short time. The eggs should be sampled and examined for their quality, number and developmental stages. The embryonic development inside the eggs could be studied using a microscope.

Check for the following egg characteristics:

- Presence of opaque, whitish eggs which are unfertilised. Similarly, eggs in the sample with transparent, but without evidence of cell divisions
- Regular round shape and size (diameter 900-1000 μm in cobia; 800-900 μm in pompano), regular cell division that can be observed only in the first blastomers; regular shape of yolk (it should occupy the egg volume entirely, without perivitelline space),
- Absence of parasites or associated micro-organisms on the chorion surface.

Incubation of eggs

Incubation of eggs is done in incubation tanks of 3-5 tonne capacity. After hatching, only the hatched fish larvae have to be moved to the larval rearing tanks filled with filtered seawater. Prior to this, the aeration should be stopped briefly to enable the debris and exuviae to settle at the bottom which can be removed by siphoning. Aeration needs to be adjusted suitably, not too strong to avoid excessive physical collision among eggs, but should be sufficient only to keep the eggs suspended in water column. The main purpose of aeration is to prevent clumping and settling down of eggs. Air bubbles should not be too small, as seen while using air diffusers instead of stones, as it results in clumped eggs and damage of the eggs. Stocking density can be maintained at a moderate level of 200 to 500 eggs per litre. The hatching of eggs takes place from 18 to 24 hours. As the fecundity is normally high in cobia, we may require more incubation tanks, whereas pompano requires only one tank/ female.

The embryonic developmental stages of cobia and pompano normally look alike except for the duration of development and size of the larvae. The photos of embryonic development and newly hatched larvae are provided below;

Larviculture

The marine fish larvae are generally classified into altricial and precocial type. The altricial type of larvae is having very less yolk reserves at hatching and hence, the larvae are in an undeveloped stage when the yolk sac is completely resorbed. The development of digestive system is also very primitive in these types of larvae. Many of the marine fin fishes which are suitable for aquaculture are having the altricial type of larvae which poses challenges in their larviculture. When the yolk reserves are fully exhausted, the larval size and mouth gape are very small and the perceptive powers for searching and taking external feed is also very less. The period when the yolk reserves are fully exhausted and larvae need to resort to exogenous feeding is a critical period in the larviculture of most marine fin fishes. Unless proper live feeds of required size is provided in sufficient densities in larviculture media and its nutritional requirements especially in terms of PUFA are met, large scale mortality is bound to happen at this stage and hence it is evident that the larviculture of marine finfish having altricial larvae is really challenging and proper management of live feed is the most vital pre-requisite for the success in terms of survival and growth of the larvae.

In addition, since most of the larvae are visual feeders providing the required light also affect the larval survival. During the critical period, the density of the live feed and its nutritional qualities determines the percentage of the survival of the larvae. The density of the larvae of the concerned species should also be regulated in the larviculture tanks for getting good survival. The marine fish larvae exhibit highly differential growth even from very early stages (in the case of cobia, starting from the first week) and hence grading from an early stage is also very much needed for increasing the survival. In addition, variety of other factors such as tank colour, size of the tank, water temperature, water quality, etc., affect the larval survival and growth. From the foregoing, it is clear that the larviculture of marine finfish is highly complicated, unless each and every factor is taken care of, the survival and growth of the larvae will be very meagre.

Newly hatched larvae have to be checked to assess their viability and condition prior to stocking in the larviculture tanks. At least 10 to 20 fish larvae have to be observed under the microscope for the following:-

- shape and dimensions
- deformities, erosions and abnormalities
- appearance of internal organs
- pigmentation
- absence of external parasites

The larvae hatched in the incubation tanks or larval rearing tanks need to be distributed in larviculture tanks to have minimal stocking density of 5 to 10 larvae/ litre for cobia and 10-20 larvae per litre for pompano. Care should be taken to avoid any mechanical stress or damage. Soon after hatching, the mouth is closed and the digestive tract is not fully developed. During this period the larvae survive on its reserves in the yolk sac.

Larviculture of cobia

Newly hatched larvae of cobia normally measures 3.4 mm size. Larval mouth opens at 3-5 days post hatch (dph). Metamorphosis starts from 18-21 dph. Newly hatched cobia larvae generally start feeding at 3 dph and they can be fed with the enriched rotifer (*Brachionus rotundiformis*) at the rate of 10-12 nos / ml, two times a day till 10 dph. From 8 dph, the larvae can be fed with enriched *Artemia* nauplii at the rate of 2-3 nos / ml, 2 times a day. During the rotifer and *Artemia* feeding stage, green water technique can be used in the larviculture system with the microalgae *Nannoc loropsis occulata* at the cell density of $1x10^{7}$ cells / ml. The weaning to artificial larval diets has to be started from 15- 18 dph. While weaning, formulated feed should be given 30 minutes prior to feeding with live feed. Size of the artificial feed has to be smaller than the mouth size of the fish. Continuous water exchange is required during weaning stage. Between 25-40 dph, the larvae are highly cannibalistic and hence size-grading has to be undertaken at every three days interval. During this stage, the fry could be weaned totally to artificial diets. Larval rearing can be practised both intensively in tanks and extensively in ponds. The major factors affecting the growth and survival of larvae are nutrition, environmental conditions and handling stress. Since there is high demand for essential fatty acids (EFAs), enrichment protocols are needed for live-feeds. The water exchange can be practically nil till 7dph and it can be gradually increased from 10-100 % from 8 to 12 dph. But, tank bottom siphoning should be carried out from day 1. The environmental conditions required during the larviculture period are DO_2: > 5mg/l, NH_3: < 0.1mg/l, pH: 7.8 – 8.4, Salinity: 25-35 ppt, water temperature : 27-33° C.

Green water has to be maintained in appropriate densities in the larval tanks. While weaning the fish larvae from rotifers to *Artemia* nauplii, co-feeding with rotifers has to be continued due to the presence of different size groups of larvae. The detail of weaning protocol is as follows.

Stage of Larvae (dph)	Size of Larvae (cm)	Size of Feed (μ)
18 – 19	2.3 – 2.6	100-200
20 – 23	2.5 – 3.5	300-500
23 – 30	3.5 – 8.0	500-800
31 onwards	> 8.0	800-1200

The juveniles measuring 10 cm length are ready for stocking in happas/ nursery tanks.

Nursery and grow-out rearing of cobia

Nursery phase of cobia can be carried out in happas or sea cages or indoor FRP / cement tanks. During nursery rearing, it is advisable to feed the juveniles with formulated feed of 1200 μ size which can be increased to 1800 μ size from 55 dph onwards. Once the juveniles reach a size of 15 gm, they are ready to stock in sea cages or land based ponds for grow-out farming.

Larvae and fingerlings are shown in the given plates (*dph = day post hatch*).

Larviculture of Pompano

The newly hatched larvae were stocked at a density of 15000 larvae in FRP tanks of 2 m^3 capacity filled with 1.5 m^3 filtered seawater. The tanks were provided with mild aeration and green water at a cell density of 1 x10^7/ml. The mouth of the larvae opens on 3dph and the mouth size was around 230 μ.

The larvae were fed from 3dph to 14 dph with enriched rotifers at a density of 6-8 nos. per ml in the larviculture tanks. Wherever possible, wild collected copepods could also be added as supplements. Co-feeding of rotifers with enriched *Artemia* nauplii has to be done during 12-14 dph, and thereafter upto 19 dph with enriched *Artemia* nauplii alone by maintaining a density of 3-5 nos. per ml in the larviculture tanks. Weaning to larval inert feeds has to be started from 15 dph and co-feeding with *Artemia* needs to be continued until 19 dph. From 20 dph feeding can be entirely on larval inert feeds. The metamorphosis of the larvae starts from 18 dph and all the larvae metamorphose into juveniles by 25 dph. Though cannibalism is not witnessed, grading has to be done during 20-25 dph to separate the shooters. Critical stage of mortality would occur during 3-5 dph and subsequent mortalities are negligible. The water exchange can be practically nil till 7dph and it can be gradually increased from 10-100 % from 8 to 14 dph.

Nursery Rearing of Pompano

Nursery rearing could be initiated from 25 to 30 dph. At this stage, artificial feed of 800 μ size could be provided. Thereafter, fingerlings were fed with progressively higher size range of floating extruded larval feeds. Daily water exchange of 100% is advisable. Water quality parameters like salinity, temperature, pH, Oxygen level and ammonia are closely monitored during the entire larviculture period. After 55dph, the fingerlings with size range from 1 to 1.5 inch size can be supplied to farmers for stocking in the happas / tanks for further nursery rearing and grow-out farming thereafter. The pompano fingerlings can be reared at salinities as low as 5 ppt. At lower salinities *i.e.* from 10 to 15 ppt, they grow faster than in pure seawater.