

LIVE FEEDS IN MARICULTURE - AN OVERVIEW

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Live food organisms include all plants (phytoplankton) and animal (zooplankton) lives grazed upon by economically important fishes. Phytoplanktons are generally eaten by zooplankton. Thus, phytoplankton forms the basis of the food chain. Live foods are able to swim in water column and are constantly available to fish and shellfish larvae are likely to stimulate larval feeding response (David, 2003). In an aquatic ecosystem, these live food organisms constitute the most valuable resource for aquaculture. Most of the fish and shellfish larvae in nature feed on small phytoplanktonic and zooplanktonic organisms. However, natural fish food organisms are usually not abundant in clear pond water, but are abundant in ponds having greenish water. The green color indicates the presence of phytoplankton and other natural food organisms. In the natural food web, zooplankton constitutes a major part of the diet for marine fish larvae and it is generally believed that copepods can meet the nutritional requirements of fish larvae (Evjemo et al., 2003).

Breeding, seed production and culture practice for different marine finfish and shellfish species has been initiated several years ago in different parts of the world. But, it has been well established only for limited number of marine finfish and shellfish species with variable success. The major impediment to commercial and successful production of larvae for marine candidate species is the utilization of an appropriate live feed during the first feeding phase of the larval cycle. This period is extremely crucial for the optimal development of marine fish larvae. A live feed with the proper nutritional composition, constituting a suitable size range, and stimulating a feeding response is necessary for large scale production of larvae, and thereby to establish, and expand marine fish culture. At present, the seed production of commercial hatcheries relies on successful supply of live zooplankton species such as rotifer, copepods and *Artemia* nauplii during larval stages. It has been studied in different places that the availability of large quantities of live food

organisms such as marine rotifers (*Brachionus plicatilis* and *Brachionus rotundiformis*), different species of copepods and *Artemia* nauplii to meet the feed requirements of different stages of fry production has contributed to the successful fry production of at least 60 marine finfish species and 18 species of crustaceans. However, to ensure sufficient live food supply, fish hatcheries need to establish a food chain supply from algae (phytoplankton) to zooplankton.

Nutritional requirement

Marine fish larvae require live feeds that contain essential nutrients at appropriate concentrations for survival, development and proper growth. A marine fish larva requires different nutrients including free amino acids, fatty acids, minerals and vitamins, etc. Among all these, fatty acids are considered as important for early development stage, and it is present in animal and vegetable fats and oils. The n-3 highly unsaturated fatty acids (HUFAs) including docosahexaenoic acid (DHA, 22:6n-3) and eicosapentaenoic acid (EPA, 20:5n-3) are essential for marine fish larvae and the ratio of DHA to EPA significantly affects the survival of marine fish larvae. The yolk of many wild marine fish eggs contains a DHA: EPA ratio of about 2.0, and it is suggested that at least 2:1 ratio of DHA: EPA is essential for first-feeding.

Selection of live feed

The cultivation of larvae is generally carried out under controlled hatchery conditions and it requires specific culture techniques especially with respect to husbandry techniques, feeding strategies, and microbial control, because the developing larvae are usually very small, extremely fragile, and generally not physiologically fully developed. Incomplete development of larval “perception” organs (ie. eyes, chemoreceptors) and digestive system, are limiting factors in proper feed selection and use during early first-feeding. Therefore the feed that is used for the larvae should match the mouth size of the fish larvae, and should elucidate the feeding response and eventually should be nutritionally rich.

i. Size at first feeding

The mouth size of first-feeding larvae usually restricts the size of the food

particles which can be ingested by the larvae. In general, mouth size is correlated with body size, which in turn is influenced by egg diameter and the period of endogenous feeding (ie. yolk sac consumption period). Therefore, in hatchery, while selecting first exogenous feed, it is important to look for the live feed with size lesser than mouth size of fish larvae.

ii. Feeding response

Eliciting feeding response of fish larvae is an important criterion for proper feeding of the cultured larvae. Fish larvae have originally evolved to feed on groups of zooplankton in nature, so the stimuli produced by the movement of live feed organisms is needed for many marine fish larvae to elicit a feeding response. The swimming activity of live food organisms generally assures a good distribution of food items in the water column, this in turn facilitating more frequent encounters with the developing fish, which has low mobility in most cases.

Microalgae in Marine finfish and shellfish hatchery

Microalgae are indispensable in the commercial rearing of various species of marine fishes. Algae are used to produce mass quantities of zooplankton (rotifers, copepods, and brine shrimp) which serve in turn as food for early stage of fish larvae. Besides, based on the “green water technique” principle, algae are used directly in the larval tanks, where they are believed to play a role in stabilizing the water quality, nutrition of the larvae by maintaining the nutritional value of live prey organism in the tank, microbial control and increasing feeding incidence by enhancing visual contrast and light dispersion. Apart from finfish hatcheries, different species of marine microalgae are very much used in shell fish hatcheries to directly feed for nauplii and also to feed *Artemia* nauplii. The culture of algae seems to have its origins in the late 1800s and was enabled by the methods developed by bacteriologists. Afterwards, methods for marine algal culture continued to advance during the middle of the twentieth century with the development of artificial media and the development of ‘f₂’ medium for the enrichment of seawater. Later on the improved methods for mono-specific algal cultures allowed expansion of hatcheries for molluscan aquaculture and enabled

culture of live invertebrates as feed for larval fish and crustaceans. The following are the important species of algae commonly used in marine fish hatcheries.

i. *Nannochloropsis* sp.

This genus comprises approximately 6 species and is mostly known from the marine environment but also occurs in fresh and brackish water. The size of the cell is 2-4 μm in diameter, non-motile spheres and does not express any distinct morphological feature. This is the one of the major algae used extensively in the aquaculture industry for growing small zooplankton such as rotifers, copepods and *Artemia*. *Nannochloropsis* is well appreciated in aquaculture due to its nutritional value and the ability to produce valuable chemical compounds, pigments such as zeaxanthin, astaxanthin and polyunsaturated fatty acids (EPA). It is rich in omega-3 unsaturated fatty acids, especially EPA (Eicosa-pentaenoic acid), making up 30% of the total fatty acids. It also contains active complex polysaccharide, functioning as immune-enhancers that strengthen nonspecific immunity in fishes.

ii. *Isochrysis* sp.

The golden-brown flagellate *Isochrysis galbana* is a rich source of polyunsaturated fatty acids (PUFA), mainly Docosa hexaenoic acid (DHA), making it a useful feed for aquatic animals. It synthesizes important bioactive metabolites such as steroids, tocopherols, carotenoids and pharmaceuticals. Therefore, it is commonly cultivated in fish hatcheries as a feed for rotifers, copepods and *Artemia*.

iii. *Chaetoceros calcitrans*

It is a marine planktonic diatom belonging to the family Chaetocerotaceae. The cells (*diameter* 2 - 85 μm , *length* 2 - 45 μm) are elliptical in valve view. It is the widely used species in aquaculture, as it is composed of nutritional value suitable for marine filter feeders especially molluscs. It is used in shrimp hatcheries to increase the vitamin levels.

iv. *Thalassiosira weissflogii*

It is a large diatom (4-32 μm) cylindrical in shape belonging to family Thalassiosiraceae. It is the major species used in shrimp and shellfish larvi-culture.

It is the single best algae for shrimp larvae and also good feed for copepods, brine shrimp, and broodstock conditioning of oysters (post set), clams and mussels.

Zooplankton in marine finfish hatchery

Zooplankton is one of the primary foods of larval fish. Successful larval rearing primarily depends on the live feed and, zooplankton forms the most important component in the live feed in indoor culture phase. Larval feed should be smaller, easily digestible, rich in nutrients and allow autolysis. Thus, they are mostly preferred in the early stage of larval rearing in finfish hatcheries. Copepods, cladocerans, decapod larvae, rotifers, *Artemia* nauplii and ciliates are the important zooplankton organisms forming a mainstay in the food of fish larvae in the wild. However, the most popular zooplankton used for fish larvae culture are rotifers, *Artemia* and copepods. The intensive larval culture of most marine fish depends on a large supply of these zooplankton.

i. Rotifers

Use of rotifers for larval rearing had been initiated in 1960s, when Japanese researchers discovered that the rotifer, *Brachionus plicatilis*, previously considered a pest in culture ponds, could be used as a first food for larvae of both freshwater and marine fish species. After this important finding, significant development in marine finfish larval rearing was achieved. This allowed the culture of many more species whose larvae hatched with small mouth size which was insufficient for the ingestion of the larger *Artemia* prey. Later on large numbers of commercially important marine fish species have been brought into culture that rely on rotifers as first food in culture facilities

Rotifers are small sized metazoans with over 2000 described species present in both freshwater and seawater environment. Presently, two marine species, *Brachionus plicatilis* (large) with size ranges from 130 to 340 μm (average 239 μm) and *B. rotundiformis* (small) with the size ranges from 100 to 210 μm (average 160 μm) have been used to culture several marine finfishes and shell fishes. In tropical aquaculture two different types of small rotifer (*Brachionus rotundiformis*) are used. (a) Super-small or 'SS'-strain - Used for first feeding when larvae change from internal food source to external feed. It is mainly preferred for the first feeding

of fish larvae with small mouth openings of less than $\sim 200 \mu\text{m}$ at start of initial feeding (rabbitfish, groupers, and other fish. (b) Small or S-strain – It is slightly larger rotifers, used after the first few days of initial feeding in the larviculture. Rotifers are planktonic filter feeders, feeding on organic particles brought to their mouths by the movements of their coronas. Rotifers ingest many types of feed, including natural algae (*Nannochloropsis sp* and *Isochrysis sp*, etc), yeast, bacteria and other formulated feeds (algal paste, selco). Depending upon their food source, rotifers may have 52 to 59 % protein, up to 13 % fat, and 3.1 % n-3 HUFA in their body content. Rotifers propagate quickly under suitable conditions, with populations doubling over a few days. Cultures can become quite dense and commonly exceed 1000 rotifers/ml.

The disadvantage with rotifers is that they lack the proper nutritional profile required by marine fish larvae due to lack DHA, EPA, and ARA. Rotifers lack the ability to elongate shorter chain fatty acids and, therefore, must be enriched to satisfy the HUFA requirement before they are fed to marine fish.

ii. Copepods

Rotifers and brine shrimp nauplii are the two conventional common live food organisms for early life stages of marine finfish in hatcheries because of their size. However, some fish species produce larvae with mouth size that is too small to ingest even rotifers at first feeding. In this situation, an alternative live prey that is suitable to use is copepod nauplii. Copepods provide additional desirable characteristics such as size and nutritional value to finfish larvae and, play a supplemental role in larval rearing. Furthermore, it is well known that the nutritional value of copepods is better than that of the convenient live feeds such as rotifers and brine shrimp, so use of copepods is becoming increasingly popular in many places. Copepods are tiny crustaceans, and are one of the most ubiquitous marine organisms with over 21,000 species currently described. Marine fish larvae eat copepod nauplii, and juvenile fish consume adult copepods. The role of copepods in the marine trophic system is essential to the survival of many marine fish species.

iii. Brine shrimp

Brine shrimp (*Artemia*) is a small branchiopod crustacean and is one of the most important live feed for the larvae of farmed aquatic organisms and therefore is an important consumable in hatcheries. After rotifers, the most common live feed utilized during marine finfish fingerling production is *Artemia* spp. Size of *Artemia* generally range from 350 to 900 μm , and represent the transitional feed after which artificial dry feeds can be used. *Artemia* nauplii are used during the later stages of larval rearing and it is purchased from commercial suppliers and hatched in the tanks. Like rotifers, brine shrimp must be enriched to increase their nutritional value before they are fed to fish larvae.

Conclusion

Strategic use of microalgae and zooplankton in hatcheries during the very early stage of the larvae improves the success of first feeding, which is prerequisite for efficient survival, growth and quality in finfish and shellfish larviculture. Marine finfish and crustacean aquaculture has greatly expanded since early 1980s in different parts of the world, which could possible by the development of fairly standard hatchery protocols for live feed. Development of standard feeding protocol for larvae of different species is very much essential to reduce the effort needed for the same when new species are developed or new geographical regions opened up to aquaculture. Presently, use of live feeds is becoming more appealing for larval culture than the use of artificial feeds because of several inherent advantages like maintaining good water quality, optimum size and amenability to enrichment. However, use of optimum live feeds depends on the fish species, nutritional status of the larvae and facilities and funds available in the hatchery.

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