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Influence of certain environmental parameters on mass production of rotifers: A review

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Original Article

Abstract

Larviculture of many finfishes and crustaceans in aquaculture depends mainly on the live feed and its unavailability in sufficient quantity is hampering its expansion and culture progress all around the world. The most suitable feed for marine finfish larvae is the commonly available zooplankton species such as rotifer, copepods and *Artemia* nauplii. Among all, the live feed that has been demonstrated more successfully as the first feed for most of the marine finfish species is rotifer. Optimum conditions are required for better growth, reproduction and increased productivity of rotifers. The major factors that influence the population size of rotifer are temperature and salinity. Hence, the impact of environmental parameters with special focus on the salinity and temperature on the increase in biomass and smaller rotifer production is of utmost importance in the present scenario.

Keywords: *Live feed, larviculture, zooplankton*

Introduction

The key of any aquaculture operation is larviculture of fishes and it depends mainly on the timely availability of appropriate size live feeds. In the advent of expanding culture, the progress of successful larviculture of marine finfishes is the major bottleneck. The use of artificial diets as the first feed on larvae does not yield the desired results due to the poor enzymatic activity of larvae and the absence of fully functional stomach (Pedersen and Hjelmeland, 1988). Efforts are being taken by aquaculturists to develop a formulated diet, which is easily digestible and non-polluting nature. In this context, hatchery operators are mainly depending on live feed to carry forward the larval rearing operations. Among the live feeds, the most commonly used starter feeds for larviculture of marine finfishes are *Brachionus plicatilis* and *B. rotundiformis* (Lubzens *et al*., 1989). The major problems persisting in mass production are the unpredictability of desired quantities, size groups and the quality of rotifer which hinders the progress of its expansion. In recent years, by manipulating environmental parameters various efforts are being undertaken to mass produce and to improve its nutritional quality. In this view, this paper insight the influence of major environmental parameters on the mass production of rotifers.

Why Rotifers?

The rotifers are preferred as an essential live food source because of their shape, size and slow moving nature which make the small larvae to easily prey on these organisms. Besides, its

nutritional composition can be either enhanced or manipulated to pass on the required nutrition to the cultured larvae. The experimental results of Markridis *et al.* (2000) showed that rotifers can be used for transferring probiotic bacteria to fish larvae. Lubzens *et al.* (2001) opined that it can be used for transferring medicinal and therapeutic agents to the fish larvae.

Mass culture techniques

The need for rotifers are rapidly increasing and hence several mass culture techniques have been developed for multiplication of rotifers (Fukusho, 1989). The most common type of rotifer production system in marine fish hatcheries is the batch culture method (Snell, 1991) due to its simplified and ease in the methodology. The batch culture technique involves using a part of the harvest for feeding fish larvae and the rest is used as inoculums for continuing the next batch of culture (Lubzens, 1987; Dhert, 1996; Odo *et al.*, 2015). Lubzens (1987) maintained a low density rotifer culture with an inoculation density between 50–200 individual's ml^{-1} to reach a final density of 300 to over 1000 individual's ml^{-1} in 3 -7 days, using microalgae and/or baker's yeast as food. However, studies by Suantika *et al.*(2000) revealed that feeding an artificial diet (Culture Selco®) with an initial density starting from 200 to 250 rotifer ml^{-1} achieved a final harvest of about 600 rotifers ml^{-1} in 4 days culture. Kailasam *et al.* (2015) has also described a high density culture methodology, in which they used the same artificial diet and harvested up to 2,000–5,000 rotifers ml^{-1} with an initial inoculum of 300–500 rotifers ml^{-1} in a culture period of 8 - 10 days.

The rotifer can fed with microalgae individually or in combination of two or more species or group. However, the nutritional quality of cultured rotifers depends on the transfer of dietary compounds from algae or yeast to the rotifers (Kalidoss *et al.*, 2017). Microalgal diets such as *Nannochloropsis*, *Isochrysis* and *Chlorella* reveals that there is an increase in neutral lipid and phospholipid content. Fernandez-Reiriz *et al.*(1989) opined that *Isochrysis galbana* was found to contain substantial amounts of DHA and a low EPA content, whereas Sukenik *et al.* (1993) found *Nannochloropsis gaditana* contained substantial amounts of EPA. Rotifers were fed with a locally isolated microalgal strain *Nannochloropsis* were capable of achieving a mean production of 186.71 x 10^6 individuals m^{-3} day^{-1} of L-type rotifers and 308.75 x 10^6 individual's m^{-3} day^{-1} of S-type rotifers in the fibreglass tanks culture system of 1000 litre capacity (James and Abu-Rezeq, 1989).

The present trend in mass production of rotifers for aquaculture is the use of high quality and high-density biomass input as a means to increase maximum production and hence a continuous system with more sophisticated methods such as mechanization with further emphasis on automation has been developed (Abu-Rezq *et al.*, 1997; Kailasam *et al.*, 2015). In these improved techniques all the physicochemical parameters are maintained in optimum condition with a steady supply of microalgae. An ultra-high density mass culture of rotifers on algae was the need of the hour; Yoshimura *et al.* (1996) done the preliminary work on it and mass culture of rotifers using artificial diets (Suantika *et al.*, 2000) was also carried out. With ultra high density rotifer production systems using algal paste, one can achieve mass production of rotifers with less space and manpower when compared to conventional production systems (Kailasam *et al.*, 2015).

Key physicochemical parameter's for mass culture of rotifers

Among the various major physicochemical parameters (temperature, salinity, pH, dissolved oxygen and ammonia) required for the lucrative growth of rotifer, the temperature and salinity have variable effects on the productivity of different strains of rotifers (Miracle and Serra, 1989; Serrania-Soto *et al.*, 2011). The optimum environmental conditions favour higher production and unfavourable conditions keep the specific growth rate of a rotifer constant. The growth rate under favourable conditions is the maximum and is characteristic of a particular population structure. In addition, the reproductive potential of the individual strain decides the mass production of rotifers. As most of the marine finfish larvae need rotifer of smaller size groups as a first feed (Gopakumar *et al.*, 2013), the mass production of the rotifer with special emphasis on its size groups is a major concern.

Influence of temperature

Among these environmental factors, temperature plays a vital role in the reproduction of rotifers. The marine rotifer, *B. plicatilis* is a euryhaline group and it is most productive at lower temperatures while *B. rotundiformis* adapts to high temperature condition (Fukasho, 1983). *B. plicatilis* and *B. calyciflorus* are the most studied species to determine the relationship between reproductive potential (r) and temperature under controlled conditions. The reproductive potential of different strains of these species has a maximum 'r' values at different temperatures (Ignacio and Martinez, 1998). Snell (1986) also studied and they concluded that temperature has a significant influence on the reproduction of *B. plicatilis*. Pascual and Yufera (1983) recorded a highest 'r' value of 1.35 at the temperature of 35°C followed by 1.10 at 30°C (Snell, 1986) and the lowest reported 'r' value of 0.12 was noted at 10°C (Hirayama and Kusano, 1972). Likewise, Hagiwara *et al.* (1995) studied the influence of temperature on the reproductive potential of *B. rotundiformis* 'S' type and *B. rotundiformis* 'SS' and recorded the highest and the lowest 'r'

values of 0.77 and 0.54 at 30°C and 25°C respectively. The highest 'r' value of 2.19 for *B. rotundiformis* 'ss' type was recorded at 30°C and the lowest value of 0.40 at 19°C (Su *et al*., 1997). Starkweather (1987) investigated the influence of temperature on the reproductive potential of *B. calyciflorus.* Similarly, for *B. dimidiatus, B. angularis* and *Keratella cochlearis* were studied by Pourriot and Rougier (1975) and Walz (1983) respectively for the relationship between the temperature and the reproductive potential. Kandasami *et al.* (1998) opined that high temperature reduces the length, width and DLS (distance between lateral spines) of the rotifer. The size of the rotifers is bigger (130–240 μm in length) at low temperature (below 20°C) and smaller (100–210 μm) at higher temperatures.

Influence of salinity

Salinity plays an important role in the distribution of rotifer in nature (Mustahal *et al.,* 1991; Joshi, 1998). However, several species can tolerate a wide range of salinity (eg. *B. plicatilis* from 1 to 97 ppt); but its optimal growth and reproduction were observed at salinities below 35 ppt (Lubzens, 1987). Like temperature, salinity also plays a vital role in the reproductive potential of an individual rotifer. Salinity directly influences the osmotic regulation capacity of an individual species which in turn strongly relates to the genotype and the species. Nevertheless, studies on salinity effects on rotifer individuals or populations are extremely scarce (Miracle and Serra, 1989). However, a study by Kabay and Gilbert (1978) had concluded that several freshwater rotifers are also salt tolerant to a certain extent. Few species of rotifers belonging to the genus *Brachionus* have been studied to determine the relationship between the salinity and 'r' under controlled laboratory conditions. Experimental studies were carried out by various workers on the species such as *B. dimidiatus, B. plicatilis* and *B. rotundiformis* (Pourriot and Rougier, 1975; Snell, 1986; Pascual and Yufera, 1983; Lubzens *et al.,* 1985; Su *et al*., 1994; Hagiwara *et al.,* 1995; Gopakumar, 1998; Assavaaree *et al.,* 2003) which are halobiont species and can withstand a wide range of salinities. Studies on the reproductive potential of *B. plicatilis* have found that the highest 'r' value was recorded at the salinity of 10 ppt to 20 ppt and above or below this salinity the 'r' value had deviated from its mean value (Roa, 1992). The best salinity to obtain the maximum 'r' value for *B. plicatilis* was at 19 ppt and 17 ppt was reported by Ito (1960) and Lubzens (1987), respectively. However, according to Snell (1986), the optimum salinity for the highest 'r' value for *B. plicatilis* was noted at 30 ppt. All these studies had revealed that different clones of *B. plicatilis* have different r-max values at different salinities.

B. rotundiformis is best adapted to low saline conditions (Fukasho, 1983). The influence of salinity on the reproductive potential of *B. plicatilis, B. rotundiformis* 'S' type and *B. rotundiformis* 'SS' type was examined by Hagiwara *et al.* (1995). The best salinity for the maximum reproductive rate of *B. plicatilis, B. rotundiformis* 'S' and 'SS' type were at 11 ppt ($r = 0.49$), 11 ppt ($r = 1.37$) and 11 and 22 ppt ($r = 1.57$ and 1.37), respectively. Rotifer size was also found inversely proportional to increasing salinities in the culture system. Kandasami *et al.* (1998) opined that the distance between the median spines (DMS) changes with salinity. The relationship of the r-max of *B. rotundiformis* 'ss' type with five salinity levels, ranging from 5 ppt to 30 ppt was studied by Su *et al.* (1994). They found the optimum culture conditions for 'ss' type was at a salinity of 10 ppt to 20 ppt and temperature of 30 °C to 33 °C. Pourriot and Rougier (1975) studied the relationship between the 'r' and salinity in *B. dimidiatus.* According to them, the highest reproductive output of this rotifer was noted at the salinities of 2 ppt ($r = 0.429$) and 19 ppt ($r = 0.424$). Thus different species or strains will have different 'r' maxima at various temperatures and salinities mainly due to species / strain collection location and its environmental parameter adaptation.

Other parameters

Apart from the key physicochemical parameters such as temperature and salinity, other factors which have a certain influence on mass production of rotifer are pH, dissolved oxygen and ammonia (Dhert, 1996). Aeration and dissolved oxygen are one of the prime factors in rotifer culture. Although rotifers can survive in dissolved oxygen levels of 2 mg L^{-1} (Table 1), for

Table 1. Optimal conditions for culture of rotifers (modified Dhert, 1996)

Parameters	Range	Optimum	
Temperature (°C)	26–34	28–30	Rotifers reared at higher temperatures within their optimum range had better growth and increased productivity.
Salinity (gL^{-1})	1 to 97 (depends on species)	25 to 30 (for marine species)	Better to rear them in salinity (± 5 ppt) closer to the larval rearing tanks.
Dissolved Oxygen (mgL^{-1})	2 to 5	>4	Mild aeration is enough for mixing. Strong aeration leads to physical damage to rotifers.
pH	7.5 to 8.5	8.0 to 8.3	The ammonia levels are influenced by the temperature and the pH of the water. High levels of un-ionized ammonia in the rearing conditions are toxic to rotifers.
Total Ammonia (mgL^{-1})	<1	<0.5	
Illumination	No need of separate light, natural day light enough		Avoid direct sunlight. It may promote filamentous algal growth.

optimum growth and multiplication the dissolved oxygen level should be maintained above 4 ppm (Fulks and Main, 1991). Aeration should be mild and enough for mixing the water; strong aeration may cause physical damage to rotifers. The pH of the culture system plays an important role as the pH, along with temperature and salinity highly influences ammonia levels and its toxicity (Bower and Bidwell, 1978). In culture conditions, better results are obtained with pH levels between 7.5 to 8.5. High levels of total ammonia in the rearing conditions are toxic to rotifers but optimal level of <1 mg L^{-1} of ammonia and the acceptable range 6 - 10 mg L^{-1} for ammonia and nitrate levels appear to be safe (Lubzens and Zmora, 2003). Use of ozone reduced ammonia, nitrite and nitrate levels by 67%, 85% and 67% respectively and the number of bacteria are found to be extremely useful in the mass culture of *B. plicatilis* (Suantika *et al*., 2001).

Organic waste accumulation by surplus food should be avoided in the rotifer culture tanks. It indirectly affects the physicochemical parameters and supports bacterial growth (Lubzens and Zmora, 2003). Not all bacteria are pathogenic to rotifers. Multiplication of harmful pathogenic bacteria should be avoided in the interest of rotifer and its feeding fish larvae (Dhert, 1996). An effective way to control the unwanted pathogenic bacteria counts is by feeding the rotifers with probiotics (Markridis *et al*., 2000). A probiotic combination containing beneficial bacteria such as *Bacillus, Thiobacillus, Acetobacter and Paracoccus* when supplemented with enzymes, was found to be more effective in the competitive elimination of Vibrios in rotifer mass culture tanks (Loka *et al*., 2016). This probiotic supplementation of beneficial bacteria has not only regulated the microfloral content but also enhanced the rotifer density and mass production. The occurrence of ciliates is generally considered as contamination, affects rotifers due to competition for food. The metabolic wastes of ciliates increase the NO_2–N level in the water. Simple screening and cleaning of the rotifers by passing through $<40\ \mu$m mesh reduces the number of ciliates and other contaminants (Dhert, 1996).

Conclusion

B. plicatilis and *B. rotundiformis* are more tolerant of temperature and salinity fluctuations (Kandasami *et al*., 1998). Such cultured live feed, especially rotifers, should satisfy the larval feeding purpose. The majority of the fish larvae with small mouth gape lack perceptive powers for searching external feed (Gopakumar *et al*., 2013). The size of the feed is important when the mouth size of the larvae is concerned (Santhosh and Anil, 2013). A suitable sized, nutritionally enriched live food organism is the major requirement for the mass rearing of many finfish larvae. To develop mass culture methods for the potential rotifer species, a controlled condition is necessary for a specific species. The environmental variables like temperature and salinity play a key role in developing a mass culture of rotifers. Thus, more understanding of the effect of these parameters on the growth and reproductive performance is very much essential to improve culture and nutritional quality of the rotifer species. Since seed production and culture practices of several marine finfishes such as Cobia, Silver pompano, Orange spotted grouper, Indian pompano, Tiger grouper and Pink ear emperor are taking up a major leap in the Indian aquaculture industry as an alternate species for mariculture; mass production of smaller rotifers is of utmost essential to take this technology forward.

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