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Replacement of *Artemia* Naupli with *Panagrellus redivivus* during Early Larval Stages of *Penaeus indicus*

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ABSTRACT

The present study focused on replacement of *Artemia* nauplii using nematodes (*P. redivivus*) during early larval stages (Mysis 3) of Indian White Shrimp (*P. indicus). P. redivivus* survive wide range of salinity (0 to 60ppt) and can be cultured in mass scale using dry yeast attain a maximum density of 1.26lakh/ml with a size range from 341 to 1693µm within 2 d of culture period makes them suitable live feed during shrimp larval rearing. Experiment was conducted in 20L plastic tubs using *P. indicus* larvae (Mysis 3) stocked at a density of 100 numbers. *Artemia* nauplii (AN) and *P. redivivus* (PR) fed thrice in a day at the rate of 10 numbers/larvae as monospecific and mixed diet combination (ratio of 1:1, 3:1 and 1:3). A significant difference in survival was observed in the treatment fed with *Artemia* nauplii and combination of AN+ PR (1:1 and 3:1) compared to other treatments. Maximum survival was obtained in the treatment fed with *Artemia* nauplii alone (75±1.2%) followed by combination of AN+ PR, 3:1 (73.3±0.9%) and AN+PR, 1:1 (70±1.7). Overall results suggest that *P. redivivus* can be effectively used along with *Artemia* nauplii upto 50 per cent during early larval stage of *P. indicus* to reduce the cost of production.

Key Words: *Artemia* nauplii, Larval stage, Live feed, Mixed diet, Nematodes, Survival.

INTRODUCTION

One of the major bottlenecks in aquaculture production is the larval rearing of fin fish and shell fishes. *P.indicus* is one of the commercially important Penaeid shrimp cultured on extensive farm in South-East Asian countries. Artemia are generally used in shrimp hatchery for feeding larval and post larval stages of Penaeid shrimp. Diets of larval decapods crustaceans commonly consist of *Artemia* nauplii is an excellent feed for crustacean larvae. Due to higher market price, variability in hatching rate, quicker growth and varying nutritional quality, other live food organisms includes moina, daphnia, copepods and nematodes have been used in Penaeid shrimp larviculture. *P. redivivus* is a free living nematodes considered as one of the promising potential live feed for aquatic animals especially during the larval stages due to their small size, high reproduction rate and option to charge the worm prior to preservation with a favorable fatty acid profile (Bruggemann, 2012). Harvested biomass of nematodes can be store in a desiccated mode up to 10 wk similar to *Artemia* sp. (Honnens *et al*, 2013). *P. redivivus* can be enriched through their whole body composition to improve their nutritional value. The present study evaluated the efficacy of *P. redivivus* as mono specific and mixed larval feed diet along with *artemia* nauplii during the early larval stages of *P. indicus*.

MATERIALS AND METHODS

The experiment was performed at Shrimp hatchery, Muttukadu Experimental Station of ICAR-Central Institute of Brackishwater Aquaculture, Chennai, India. Wild caught *P. indicus* broodstock screened for WSSV, IHHNV and other OIE listed pathogens and negative animals with stage IV bought in our hatchery for larval production. The

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female ready to spawn were transferred to 100L individual FRP spawning tanks filled with filtered seawater at 30ppt salinity and water temperature maintained at 30-32°C. Spawning occurs at night time and eggs were collected using fine mesh net washed properly and treated with 100ppm formalin for 30sec followed by Treflan dip (0.05-0.1ppm) to remove heavy metals and fungal infections and finally Povidon iodine (50ppm) dip for 1min and gently rinsed in running water for 5min. The eggs are then transferred into 500L FRP tanks treated with EDTA (5ppm). Once hatching occurred after 14hr, the nauplii were collected using light source provided at the top and treated with formalin and Povidon iodine dipped and released into larval rearing tanks for further rearing process. Microalgal species, *Cheatoceros* sp. was used as major diet during zoea stage.

P. redivivus (Fig. 1) stock culture collected from live feed section of Muttukadu shrimp hatchery and produced in mass scale using dry yeast for feeding during shrimp larval stages. A preliminary study was conducted to find the size, density of nematodes using yeast media and range of salinity (0 to 80ppt) at which nematodes can survive prior feeding to the shrimp larvae. One day old *Artemia* naupli produced from the imported cyst in an artemia hatching unit before the commencement of experiment. All the live feeds used for the study have produced in sufficient number on daily basis at 30ppt.

The experiment was conducted in plastic tubs (50L) in triplicate filled with filtered UV treated seawater (30psu) stocked with 100 number of mysis larvae (Mysis 3) fed with nematode and *Artemia* nauplii at the rate of 5-10nos/ larvae as monospecific and in combination till the larvae attain PL12. Daily water exchange was accomplished by using suitable mesh sieve to remove excess unutilized feed. *Chaetoceros* sp. added in each treatment at cell density of 1×10^5 cells/ml. Growth and survival rate was estimated at the end of the experiment to find out best feed diet during the larval stage.

Fig. 1: Microscopic view of *P.redivivus*

Statistical analysis

The growth and survival data of *P.indicus* larvae were analysed using one-way ANOVA by SPSS 16.0 to find out significant difference ($p<0.05$) among the treatments. All value represented as Mean± Standard Error (SE) of the mean.

RESULTS AND DISCUSSION

P. redivivus could be cultured in yeast media attained a maximum culture density of 1.26lakh larvae/ml within a period of 48hr. The size of the species varied from 341 to 1693µm. The salinity tolerance study revealed that *P. redivivus* can tolerate a wide range of salinity from 0 to 60ppt. Highest survival was observed at 0, 10, 20, 30 and 40ppt respectively. *P. redivivus* did not consume algae and can survive in saltwater for extended period of time. They can be cultured easily and never grew to a size too large to be consumed by shrimp larvae.

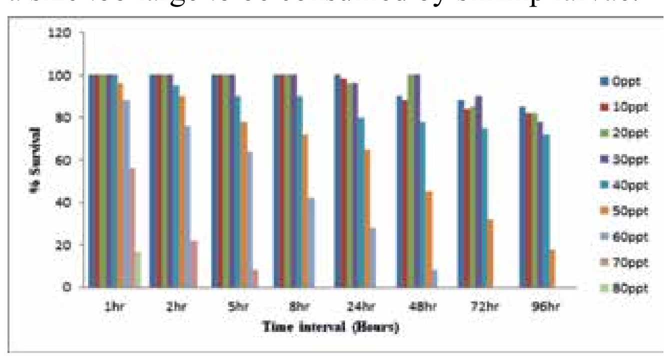

Fig. 2: Percentage survival of *P.redivivus* in different salinities at various time intervals

Despite a slower growth rate compared to leading Penaeid shrimp culture, *P. indicus* has great potential to the aquaculture industry as it was tolerant of recirculation system and gets matured easily in captivity. Microalgal concentration of 30-40 cells/ PL was adequate during the zoea stages of *P. indicus* (Galgani and Aquacop, 1988). Percentage survival of shrimp larvae using *P. redivivus* and *Artemia* naupli as monospecific or in combination observed that there was a significant difference in survival ($p<0.05$) in the treatment fed with *Artemia* naupli alone and combination of AN+PR (3:1 and 1:1) compared to other treatments (Fig. 3). All the live feeds were started taken by the shrimp larvae during the Mysis 3 stage. Treatment fed with *Artemia* nauplii with percentage survival of (75±1.2%) followed by combination of AN+ PR (73.3±0.9%) and AN+EB (68±1.7). *Moina micrura* can effectively substitute *Artemia* nauplii throughout the larval rearing period of *Macrobrachium rosembergii* with no difference in survival rate, mean stage of development or growth rate (Alam *et al*, 1995). It was not possible to determine to what extent the nematodes were actually utilized by the shrimp larvae fed with mixed animal diet. However, it seemed likely that even in situations where *Artemia* nauplii cannot be completely eliminated from the larval diet, the quantity required might be substantially reduced by introducing *P. redivivus* supplement without sacrificing production performance. Combination of *P. redivivus* and *Artemia* nauplii gave beneficial effect towards the mass production of *P. pelagicus* larvae (Affandi *et al*, 2019). Average wet weight and total length of *P. indicus* indicated that *Artemia* naupli can be effectively replaced up to 50 per cent with nematodes. *P. aztecus* and *P. setiferus* larvae fed with nematode showed equal survival, metamorphosis and dry weight growth than that fed with *Artemia* nauplii. *M. micrura* can partially replace *Artemia* nauplii in early postlarval stages of *L. schmitti* without affecting survival, growth rate, protease and a-amylase activities and resistance to osmotic stress (Martin *et al*, 2006).

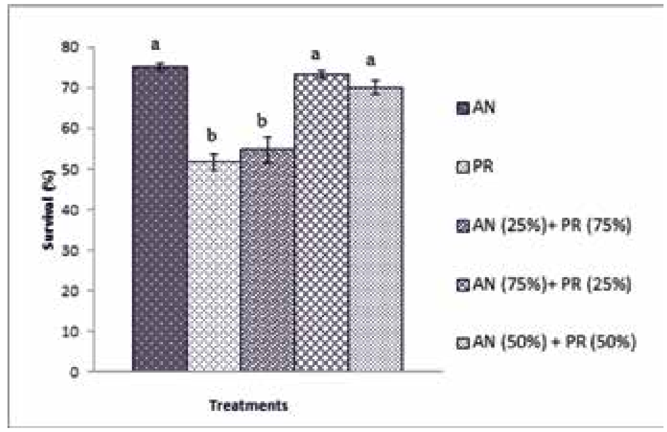

Fig. 3: Percentage survival of *P. indicus* Post larvae (PL12) at the end of experiment

CONCLUSION

The present study concluded that *P. redivivus* has significant potential in expanding the aquaculture industry in terms of larval production. The rising cost of Artemia and cost effective productive practice of nematodes should be the best choice to be implemented in order to improve the output of larval production. It has been found that *P. redivivus* was an effective live feed which can be used along with *Artemia* nauplii in different combination during the early larval stages of *P. indicus* to reduce the cost of production significantly. In addition, further research on nutritional profile need to carry out to determine the level of acceptance and live stability towards the use of different enrichment component such as protein, lipid, vitamins and minerals.

Table 1: Average total length and wet weight of *P. indicus* (PL 12) fed with different feed diets

	AN	PR	AN(75%)+PR(25%)	AN(50%)+PR(50%)	AN(25%)+PR(75%)
Wet weight (mg)	14.2^{a}± 2.1	10.1^{b}± 1.8	16.3^{ac}± 2.5	15.3^{a}± 1.9	11.2^{b}± 1.5
Total length (cm)	1.3^{a}±0.2	0.8^{b}±0.1	1.5^{a}±0.2	1.3^{a}± 0.3	1^{c}±0.1

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