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Use of Copepods as Live Feed for Larviculture of Damselfishes

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Abstract

It is well established that small-sized live feed with sufficient DHA, EPA and ARA as starter feed is the key factor for the success in larviculture of marine fishes with altricial larvae having limited yolk, which are in an undeveloped state at hatching. Copepods form a major component of the natural diet of many fish larvae and the wide range of body sizes of copepods both within and between species is extremely useful for employing the early stage nauplii and copepodites as starter feed for very small larvae with small mouth opening. Improved survival, growth and normal pigmentation have been documented in the larviculture of several marine finfishes reared with the early stage nauplii and copepodites. This is generally attributed to the levels of DHA, EPA and or arachidonic acid (ARA) in the diet and particularly to the DHA: EPA ratio in the diet. Two species of copepods viz. *Euterpina acutifrons*, a harpacticoid copepod and *Pseudodiaptomus serricaudatus*, a calanoid copepod were selected and cultured. They were employed as starter feed for the larviculture of three species of damsel fishes viz. the three spot damselfish, *Dascyllus trimaculatus*, the humbug damselfish, *Dascyllus aruanus* and the blue damselfish, *Pomacentrus caeruleus*. It was found that co-culturing of copepods in greenwater in the larviculture tank is the most effective method for initiating the exogenous feeding of the species studied. Initially greenwater was developed in the larval rearing tanks by adding sufficient quantity of the culture of microalgae *Nannochloropsis* sp. so as to get a cell count ranging from 1×10^5 cells / ml to 6×10^5 cells / ml. Adults of copepods *E. acutifrons* and *P. serricaudatus* were introduced into the greenwater. When the copepods started their growth phase, newly hatched larvae were introduced into the tanks. The number of egg bearing copepods and nauplii per 50 ml in the larviculture tanks upto 20-25 days of post hatch is presented. The larval survival in relation to abundance of nauplii and egg bearing copepods is also given. The utility of copepods in the larviculture of damselfishes is discussed in the light of the results of the study.

Introduction

Copepods constitute a first vital link in the marine food chain leading from primary producers to fish. The rapid expansion of hatchery production of seed for marine aquaculture and increasing interest in new species and the culture of ornamental species to replace wild fisheries, necessitate development of suitable larval feeds which could not be met by conventional species of live feeds such as rotifer and *Artemia*. Thus

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interest in copepods has been generated and the use of copepods as live feed in aquaculture gained momentum. They are employed mainly because they are the only prey of acceptable size for small larvae of ornamental fish species or the only type of live feed that will support the rearing of many species of marine finfish with altricial type of larvae. Marine copepods, the principal diet for most marine fish larvae in nature, contain high levels of DHA and other PUFA, either obtained through their phytoplankton diet or accumulated despite low PUFA levels in the diet. Copepods are also an important source of exogenous digestive enzymes and are thought to play an important role in fish larval digestion.

The newly hatched larvae of marine ornamental fishes of the family Pomacentridae (other than clownfishes) are very small and hence they cannot be reared by employing rotifers as live feed for first feeding. Three species of damselfishes viz. the humbug damsel *Dascyllus aruanus*, the three spot damsel *D. trimaculatus* and the blue damsel *Pomacentrus caeruleus* have been successfully reared by using copepods.

Materials and Methods

Two copepod species viz. *Euterpina acutifrons* and *Pseudodiaptomus serricaudatus* were selected based on their small size and isolated from wild plankton collections. Mixed culture experiments of the two species were done in 5 tonne FRP tanks. Initially greenwater was produced by the microalga *Nannochloropsis oculata* at a cell count range of 1×10^5 cells^{-ml} to 6×10^5 cells^{-ml}. The two species of copepods selected were isolated from the wild collection and inoculated to the culture tanks at a density of about 20-25 adults per 50ml. When the copepods started their growth phase, daily counts of adults, egg bearing ones, copepodites and nauplii were recorded. The microalgal cell count was maintained by adding fresh cultures throughout the period of experiment.

Freshly hatched larvae of three species of damselfishes viz. *Dascyllus aruanus*, *D. trimaculatus* and *Pomacentrus caeruleus* from the hatchery were employed for larviculture experiments. Larval rearing was carried out in 5 tonne FRP tanks of light blue colour. Initially green water was produced in the larviculture tanks using *Nannochloropsis oculata* at a cell count range of 1×10^5 cells^{-ml} to 6×10^5 cells^{-ml} and the same was maintained during the entire period of experiment. The mixed culture of the two selected species of copepods from the culture tanks was filtered and inoculated to the larviculture tanks at a density of 20-25 adults per 50 ml. When the copepods started their growth phase, as was observed by counting the number of egg bearing copepods and nauplii per 50 ml, about 2000 numbers of the newly hatched larvae of each species of fish were introduced into the respective tanks. The experiment was conducted upto 25th day of post hatch (dph) for *Dascyllus aruanus* and *D. trimaculatus*

and upto 20th (dph) for *Pomacentrus caeruleus*. Three replicates were carried out for each species and the average values were taken. The control experiments were done by employing the rotifer *Brachionus rotundiformis* as live feed instead of copepods.

Results

Measurements of copepod species employed

***Pseudodiaptomus serricaudatus*:** Freshly hatched nauplii : length 65 - 70 μ , width 45 - 50 μ ; Size range of different naupliar stages: 65 - 190 μ ; size range of copepodites: 200 - 700 μ ; Size range of adults: 700 - 850 μ .

***Euterpina acutifrons*:** Freshly hatched nauplii: length 50 - 60 μ , width 40 - 45 μ ; size range of different naupliar stages: 50 - 180 μ ; size range of copepodites: 190 - 500 μ ; size range of adults: 500 - 600 μ

Mixed culture of *P.serricaudatus* and *E.acutifrons*

The culture could be maintained in healthy condition for nearly 25 days. The daily counts of adults (non-egg bearing), egg bearing copepods, nauplii and copepodites in 50ml of culture is presented in Figs. 1- 4. The culture was in the productive phase for about 12 days from 5th day to 16th day.

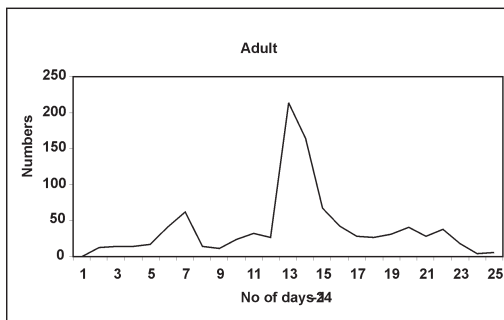


Figure 1. Daily count of adult (non-egg bearing copepods)

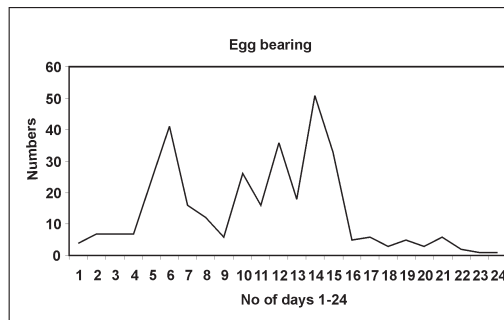


Figure 2. Daily count of egg bearing copepods

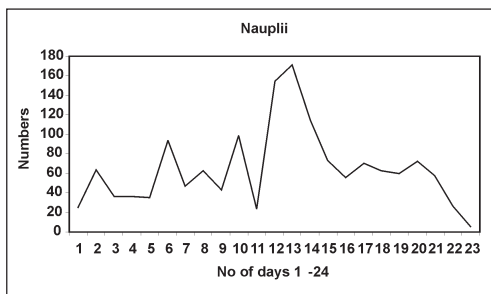


Figure 3. Daily count of nauplii

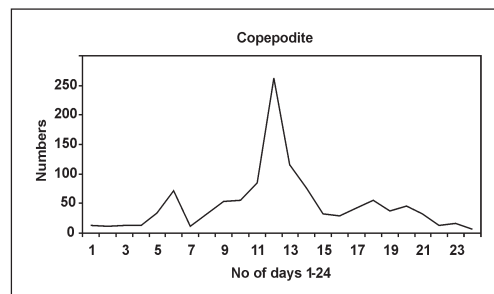


Figure 4. Daily count of copepodites

Larviculture of *D.aruanus*

The daily counts of egg bearing copepods and nauplii per 50 ml of the larviculture tank are given in Fig.5. The egg bearing copepods and nauplii in 50 ml ranged from 1-109 and 3-273 respectively. The larval survival on 25 dph ranged from 3-8%. In the control tanks total mortality was noted on 4 dph.

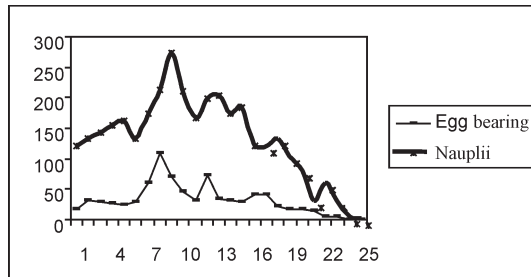


Figure 5. Daily counts of egg bearing copepods and nauplii per 50 ml in larviculture experiments of *Dascyllus aruanus* employing mixed culture of copepods.

Larviculture of *D.trimaculatus*

The daily counts of egg bearing copepods and nauplii per 50 ml of the larviculture tank are given in Fig.6. The egg bearing copepods and nauplii in 50 ml ranged from 7-97 and 35-203 respectively. The larval survival on 25 dph ranged from 3-4%. In the control tanks total mortality was noted on 4 dph.

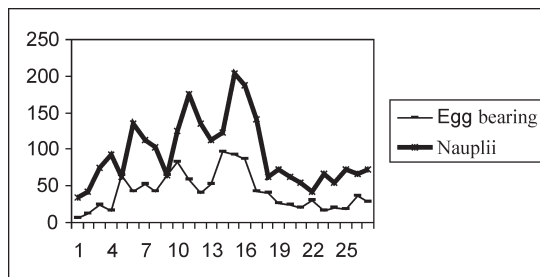


Figure 6. Daily counts of egg bearing copepods and nauplii per 50 ml in larviculture experiments of *Dascyllus trimaculatus* employing mixed culture of copepods.

Larviculture of *P.caeruleus*

The daily counts of egg bearing copepods and nauplii per 50 ml of the larviculture tank are given in Fig.7. The egg bearing copepods and nauplii in 50 ml ranged from 7-41 and 23-132 respectively. The larval survival on 20 dph ranged from 3-4%. In the control tanks total mortality was noted on 4 dph.

Discussion

Copepods are nutritionally suitable for marine fish larvae (Stottrup, 2000; Stottrup 2006; Stottrup and Norsker, 1997) and constitute a large percentage of the natural diet of fish larvae (Mc Kinnon et al., 2003). When compared to *Artemia* nauplii and rotifers,

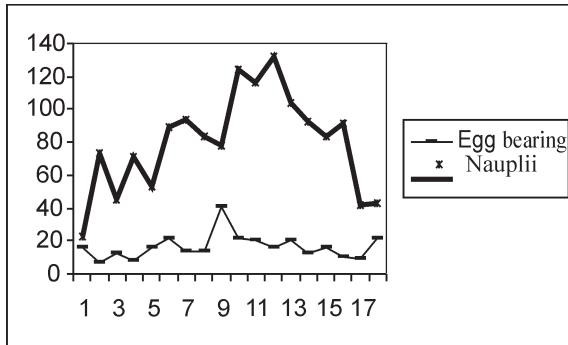


Figure 7. Daily counts of egg bearing copepods and nauplii per 50 ml in larviculture experiments of *Pomacentrus caeruleus* employing mixed culture of copepods

survival and rates of normal pigmentation have been documented for several marine fish species fed copepods alone or as supplement to the traditional diets of rotifers or *Artemia* nauplii. (Heath and Moore 1997; McEvoy et al., 1998; Naess and Lie 1998; Nanton and Castell 1999). The improvements in larval growth, survival and rates of normal pigmentation are generally attributed to levels of DHA, EPA and or arachidonic acid (ARA) in the diet (Castell et al; 1994; Reitan et al. 1994; Sargent et al. 1997; Nanton and Castell 1998).

Production of high-density cultures of copepods is the major bottleneck for the seed production of marine finfishes. It has been noted that feeding mixed culture of suitable sized copepods is advantageous for the survival of larvae since a variety of size ranges of nauplii will be available as larval feed. The copepods selected for mixed culture in the present study *viz.* the calanoid *P.serricaudatus* and the harpacticoid *E.acutifrons* have the required sized nauplii suited for the initial feeding of the larvae. The adult *P.serricaudatus* occupied the water column of the culture tank whereas the adult *E. acutifrons* were mostly at the bottom of the tank. But the naupliar stages of both the species were spread throughout the water column, which facilitated larval feeding. However maintaining a high-density mass culture of copepods similar to rotifer culture is not easy since the multiplication rate of copepods cannot be compared to rotifers. The low density of copepods in the mixed culture experiments indicates the same.

The larvae of the three species of damselfishes employed in the present study are very small larvae at the time of hatching. The mouth gape of the newly hatched larvae ranged from 150-200 μ . The newly hatched nauplius of the two species of copepods employed ranged from 50-70 μ and is suited for the first feeding of the larvae of the damselfish species studied. Co-culturing of the copepods in green water in the larviculture tanks is congenial, since regular availability of nauplii to the larvae can be

various species of copepods offer different size ranges of nauplii suitable for the first feeding of many marine finfish larvae, which have small mouth gape. Copepods are the only acceptably sized prey for small larvae of ornamental fish species or the only type of live feed that will support the rearing marine fish species with very small larvae (Doi et al, 1997a; Doi et al., 1997b; Naess and Lie 1998; Toledo et al., 1999; Payne and Rippingale 2000b; Payne et al., 2001). Improved growth,

assured by this method. The comparatively low percentage of larval survival can be attributed to the lower density of egg bearing copepods and newly hatched nauplii in the medium. It is felt that the larval survival can be increased if a higher density of egg bearing copepods and nauplii could be maintained in the larviculture system. Total mortality of the larvae in the control tanks with rotifers as live feed can be attributed to the larger size (around 150 μ lorica length) and poor nutritional quality of rotifers.

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