Full Length Research Paper

Growth, survival and proximate body composition of Labeo rohita larvae fed artificial food and natural food organisms under laboratory condition

Yahya Bakhtiyar^{1*}, Seema Langer¹, S.K. Karlopia¹ and Imtiaz Ahmed²

¹Department of Zoology, University of Jammu, Jammu-1800 06, India. ²Department of Zoology, University of Kashmir, Srinagar-1900 06, India.

Accepted 11 April, 2011

This investigation was carried out to study the effect of different live feed and dry feed on growth and survival of *Labeo rohita* at a stocking density of 20 larvae per trough having 10 L of water. The increase in weights and lengths were used as measures of growth. The duration of the experiment was 30 days. Seven dietary treatments were tested viz. Artificial diet with 45% protein (LFr1), rotifers like *Asplanchna* and *Brachionus* (LFr2), wild zooplankton (LFr3), bioenriched zooplankton (raised on *Chlorella*, cod liver oil and vitamin C) (LFr4), Artemia (LFr5), Chironomous (LFr6) and Oligochaetes (LFr7). Among the different treatments, larvae fed with diet LFr4 resulted in the maximum body weight gain (BWG) (13.18 \pm 0.13 mg) which was significantly (P < 0.05) higher than all the treatments which followed the order of preference as: LFr5 (12.31 \pm 0.10 mg), LFr3 (9.75 \pm 0.08 mg), LFr2 (7.85 \pm 0.10 mg), LFr7 (6.49 \pm 0.15 mg), LFr6 (5.66 \pm 0.13 mg) and LFr1 (4.99 \pm 0.14 mg). The diet LFr4 recorded maximum survivability (92.0 \pm 2.0%). All the values recorded for the BWG and survival were found to be significantly different (P < 0.05) among others. Protein and lipid content of the prawn after the treatment was found to be highest for LFr5 and LFr4, respectively.

Key words: *Labeo rohita*, food organisms, growth, body composition.

INTRODUCTION

In intensive rearing of larvae of fishes and prawns, feeding constitutes a major factor since the fish obtain their entire nutritional requirement (except part of mineral requirement) through the food they consume (Pillay, 1990). The normal behaviour and growth of larvae mainly depends upon the quality of the diet provided to them. However, some formulated feeds have been observed to be richer in protein than live feeds but appraisal of literature revealed that larvae prefer live feed to formulated feed. Moreover. the minerals micronutrients needed by early stages of fish, prawn and shrimp are not yet fully understood and hence cannot be incorporated in formulated feeds. On the other hand, the natural live food organisms supply all these micronutrients which otherwise are not known to us. Since live feed is rich in proteins, carbohydrates and fats along with various types of vitamins and minerals, it is always preferable to have a regular supply of live feed (Singh et al., 1994). Thus "live feed" serves as "living capsules" of nutrition (Tiwari, 1986) and their nutritional status can be further enhanced by using technique known as "bioenrichment" so that nutritional status of the fishes, prawns and shrimps feeding on them could be increased. This investigation is also a small contribution in this direction where zooplankton fed chlorella enriched with cod liver oil and vitamin C has been used as one of the dietary treatment, comparing its performance with non-enriched live and artificial feeds.

MATERIALS AND METHODS

Rearing units and procedure

The study was conducted at the Wet Laboratory, Department of

^{*}Corresponding author. Email: yahya.bakhtiyar@gmail.com.

Table 1. Growth and survival of *Labeo rohita* larvae fed different feed.

Parameter	Dietary treatment								
	LFr1	LFr2	LFr3	LFr4	LFr5	LFr6	LFr7		
IAW (mg)	0.37 ± 0.02	0.36 ± 0.01	0.36 ± 0.15	0.36 ± 0.10	0.37 ± 0.10	0.38 ± 0.00	0.35 ± 0.15		
FAW (mg)	5.36 ± 0.12^{9}	8.21 ± 0.09^{d}	10.11 ± 0.07 ^c	13.54 ± 0.12 ^a	12.68 ± 0.10 ^b	6.05 ± 0.13^{f}	6.84 ± 0.14^{e}		
BWG (mg)	4.99 ± 0.14^{9}	7.85 ± 0.10^{d}	9.75 ± 0.08^{c}	13.18 ± 0.13 ^a	12.31 ± 0.10 ^b	5.66 ± 0.13 ^f	6.49 ± 0.15^{e}		
SGR (%day ⁻¹)	8.87 ± 0.23^{f}	10.38 ± 0.10^{d}	11.05 ± 0.16 ^c	12.05 ± 0.11 ^a	11.74 ± 0.11 ^b	9.13 ± 0.08^{f}	9.84 ± 0.18^{e}		
Survival (%)	74.66 ± 2.30°	81.33 ± 2.30 ^b	87.33 ± 2.30 ^a	92.0 ± 2.0^{a}	89.33 ± 3.05 ^a	76.66 ± 3.85^{bc}	74.0 ± 3.46^{c}		

Values having the same super script do not differ significantly (P > 0.05).

Zoology, University of Jammu. Newly hatched larvae of *Labeo rohita* brought from the Gho-Manhasa fish farm were stocked at a density of 20 larvae per trough having 10 L of water. Water in the troughs was changed every alternate day. The increase in weights and lengths were used as measures of growth. The duration of the experiment was 30 days.

Feeds and feeding rates

The seven dietary treatments tested were: Artificial diet with 45% Protein (LFr1); rotifers like *Asplanchna* and *Brachionus* (LFr2); wild zooplankton (LFr3); bioenriched zooplanktons (raised on *Chlorella*, cod liver oil and vitamin C) (LFr4); artemia (LFr5); Chironomous (LFr6); Oligochaetes (LFr7). Three replicate troughs were assigned to each treatment. Artificial feed was provided at the rate of 10% body weight of larvae while live feed was provided ad-libitum. Larvae in each tank were weighed weekly to the nearest 0.1 mg and counted for determination of average weight and survival.

Growth parameters

The fish larvae were measured for initial average weight (IAW) at the start of the experiment and final average weight (FAW) at the end of the experiment to calculate the following parameters:

Specific growth rate (%SGR day⁻¹) =
$$\frac{\text{In (Final weight - Initial weight)}}{\text{Number of days}} \times 100$$

Proximate analysis

At the end of the experiment, the experimental animals were analysed for their nutritional status as per standard methods. The analyses were calculated using the following methods: protein by Lowry et al. (1951), lipid by Folch et al. (1956), moisture and ash by AOAC (1999).

Statistical analysis

The data were analysed to test the level of significance with the help of Microsoft Excel 2003 and the Statistical Package for the Social Sciences (SPSS) (12.0 Version, Chicago, USA). The level of significance was also tested by one way analysis of variance (ANOVA), Duncan Post Multiple comparisons (Duncan, 1955).

RESULTS AND DISCUSSION

The proximate composition of the food items used during the experiment was analyzed. During the experimental period, no disease, abnormality or deformity was noticed in the experimental larvae. The performance of larvae in terms of growth, survival and body composition was also put on record (Table 1).

Table 1 clearly revealed that diet LFr4 resulted in the maximum body weight gain (BWG) (13.18 \pm 0.13 mg) which was significantly (P < 0.05) higher than all the treatments followed by LFr5 (12.31 \pm 0.10 mg), LFr3 (9.75 \pm 0.08 mg), LFr2 (7.85 \pm 0.10 mg), LFr7 (6.49 \pm 0.15 mg), LFr6 (5.66 \pm 0.13 mg) and LFr1 (4.99 \pm 0.14 mg). All the values recorded for the BWG were significantly different (P < 0.05) among other treatments.

Specific growth rate (SGR) was also recorded to be significantly higher (P < 0.05) for LFr4 (12.05% day 1) followed by LFr5 (11.74 \pm 0.11% day 1), LFr3 (11.05 \pm 0.16% day 1), LFr2 (10.38 \pm 0.10% day 1), LFr7 (9.84 \pm 0.18% day 1), LFr6 (9.13 \pm 0.08% day 1) and LFr1 (8.87 \pm 0.23% day 1). However, the values recorded for diets LFr6 and LFr1 were insignificantly different (P > 0.05). Higher growth was supported by the increment of both BWG and SGR.

Poor survival rate of the larvae was recorded with the diet LFr1 (74.66 \pm 2.30%), whereas, maximum surviving ability was recorded with the LFr4 diet (92.0 \pm 2.0%) and was not found to be significantly different (P > 0.05) from LFr5 (89.33 \pm 3.05%) and LFr3 (87.33 \pm 2.30%).

A significant variation was also observed in the main body constituents of the larvae after the experimental trial was over (Table 2).

Moisture content was found to be significantly higher (P < 0.05) with diet LFr1 (81.34 \pm 0.25) which was not found to differ significantly with diet LFr4 (79.51 \pm 0.31%), and LFr3 (79.65 \pm 0.31%), LFr2 (79.99 \pm 0.39%) and LFr5 (80.08 \pm 0.83) diets.

The protein content was recorded to be maximum for LFr5 (15.49 \pm 0.13%) and LFr4 (15.46 \pm 0.17%). The values recorded for LFr5 and LFr4 were significantly higher (P < 0.05) than all the treatments but no significant variation (P > 0.05) was noticed among all the treatment. The lowest value of protein was recorded for LFr1

Table 2. Effect of different feed on the body composition of <i>Labeo rohita</i>

Parameter	Dietary Treatment								
	LFr1	LFr2	LFr3	LFr4	LFr5	LFr6	LFr7		
Moisture (%)	81.34 ± 0.25 ^a	79.99 ± 0.39 ^{bc}	79.65 ± 0.31 ^c	79.51 ± 0.31°	80.08 ± 0.83^{bc}	80.69 ± 0.40^{ab}	80.68 ± 0.61 ^{ab}		
Protein (%)	14.61 ± 0.10°	15.12 ± 0.22^{b}	15.34 ± 0.16 ^{ab}	15.46 ± 0.17 ^a	15.49 ± 0.13 ^a	15.32 ± 0.12 ^{ab}	15.05 ± 0.21 ^b		
Lipid (%)	1.34 ± 0.12^{d}	1.48 ± 0.11 ^{cd}	1.71 ± 0.08 ^b	2.17 ± 0.11 ^a	2.06 ± 0.10^{a}	1.51 ± 0.15 ^{bcd}	1.65 ± 0.10 ^{bc}		
Ash (%)	2.01 ± 0.18^{a}	2.08 ± 0.16^{a}	1.54 ± 0.11 ^{bc}	$1.33 \pm 0.07c$	1.52 ± 0.06^{bc}	1.44 ± 0.09^{bc}	1.60 ± 0.08^{b}		

Values having the same super script do not differ significantly (P > 0.05).

 $(14.61 \pm 0.10\%)$ which was found to differ significantly (P < 0.05) from all other treatments.

Lipid content was noticed to be higher in LFr4 (2.17 \pm 0.11%) and was found to differ significantly (P < 0.05) with all the treatments except LFr5 (2.06 \pm 0.10%) in which the variation was found to be not significant (P > 0.05). The lower values of lipid content were found with LFr1 (1.34 \pm 0.12%) which was not observed to differ significantly with LFr2 (1.48 \pm 0.11%) and LFr6 (1.51 \pm 0.15%), respectively.

Similarly, ash content was recorded to be higher in LFr2 (2.08 \pm 0.16%) which was not found to differ significantly (P > 0.05) from LFr1 (2.01 \pm 0.18%). The lowest value for ash was recorded with LFr4 diet (1.33 \pm 0.07%) which did not significantly differ from LFr6 (1.44 \pm 0.09%), LFr5 (1.52 \pm 0.06%) and LFr3 (1.54 \pm 0.11%) diets.

The overall results revealed significantly higher (P < 0.05) BWG, %SGR and survival ability was recorded for LFr4 (bio-enriched zooplanktons) although the survival ability was not found to differ significantly with LFr5 and LFr3. Proximate body composition analysis although recorded a maximum protein with diet LFr5 (*Artemia*) was not found to differ significantly (P > 0.05) with LFr4 (bio-enriched zooplanktons) and the body lipid content was found to be higher with LFr4 but was not found to differ significantly from LFr5.

Correlation of the different body components (protein, lipid, moisture and ash) in *L. rohita* revealed that moisture showed significant but negative correlation with protein (r = -0.452, P < 0.05) and lipid (r = -0.523, P < 0.05), and a positive non significant relationship with ash (r = 0.335, P > 0.05). Protein was found to show highly significant positive relationship with lipid (r = 0.689, P < 0.01) and highly significant negative correlation with ash (r = -0.601, P < 0.05). Lipid showed highly significant negative correlation with ash (r = -0.601, P < 0.01).

Although body composition analysis revealed higher protein and lipid content for LFr4 and LFr5, the overall results of growth performance indicated LFr4 to be the best suited diet for the larvae of *L. rohita* as the diet LFr4 resulted in a higher BWG, SGR and survival ability than LFr5

The differences in the growth and survival of rohu larvae could be attributed to the quality preference,

ingestion and digestion of diets. In this study, diet LFr4 (bio-enriched zooplanktons) resulted in better growth, survival and biochemical composition of rohu larvae. These results were in conformity with the results obtained by Kaur and Bains (2005) where the authors also obtained better results with mixed zooplanktons and encapsulated egg diet for *L. rohita* larvae and recorded survival rate of 92.50% with SGR of 10.55% day⁻¹. Similarly, in their other experiment on frys of *L. rohita*, maximum weight gain and SGR were obtained when frys were fed mixed zooplankton.

The reason for the best growth and survival of rohu larvae with bio-enriched zooplankton may be due to the presence of variable size groups and a variety of prey organisms. The larvae have the option to select their prey according to their preference and choice. The variability in the size of zooplankton also helps to overcome gapesize limitation.

Copepods present in the LFr4 during this work were believed to meet the maximum nutritional requirements of the fish larvae. Scientists have reported that copepods have high contents of docosa-hexanoic acid (DHA) and eicosa-pentanoic acid (EPA), which results in better larval growth rate and survival (Evjemo et al., 1997; Shields et al., 1999). Cod liver oil (CLO) and vitamin C enriched zooplanktons fed to fish larvae improved their survival and growth. Many investigators further authenticated similar observations in the case of shrimp (Kyungmin, 2000; Immanuel et al., 2007). These results supported the hypothesis that, stress creates increased ascorbate requirements for larval fish and crustaceans. Vitamin C (ascorbic acid) is considered to be an essential component in the diet of fishes. In this respect, body vitamin C concentration may be reflected in the survival potential more accurately than variation in the growth rate (Dabrowski, 1992). The rapid growth rate observed during this study suggested that larvae of L. rohita have higher vitamin C requirements. Merchie et al. (1996) also reported improved survival, growth performance, skeleton development, stress resistance and immune response of larvae fed vitamin C enriched diet and premeditated the larvae to have higher vitamin C requirements than the adults.

However, in contrast to this study, Kaur and Dhawan (2004) reported that dry diet resulted in higher wet weight

gain and survival (90%) than live feed in case of rohu larvae. During the period of this investigation, although the growth and survival was obtained best with LFr4 diet, the diet LFr5 (*Artemia*) did not lag behind during the experiment and resulted in higher body protein composition which was not significantly different from the results obtained with diet LFr4. So the importance of *Artemia* as live feed was again confirmed by this investigation. Furthermore, Prinsloo and Schoonbee (1986) observed zooplankton as best larval food for a variety of fish larvae.

ACKNOWLEDGEMENTS

The authors are highly grateful to the Head, Department of Zoology, University of Jammu for providing necessary facilities and DST (Government of India) for providing financial assistance.

ABBREVIATIONS:

LFr1, Artificial diet with 45% Protein; LFr2, rotifers like Asplanchna and Brachionus; LFr3, wild zooplankton; LFr4, bioenriched zooplanktons; LFr5, artemia; LFr6, Chironomous; LFr7, Oligochaetes; IAW, initial average weight; FAW, final average weight; BWG, body weight gain; SGR, specific growth rate; DHA, docosa-hexanoic acid; EPA, eicosa-pentanoic acid; CLO, cod liver oil.

REFERENCES

- AOAC (1999). Official methods of the Association of Official Analytical Chemists, p.1298.
- Dabrowski K (1992). Ascorbate concentration in fish ontogeny. J. Fish Biol., 40: 273-279.
- Duncan DB (1955). Multiple range and multiple 'F' tests. Biometrics, 11: 1-42.
- Evjemo JO, Olsen Y (1997). Lipid and fatty acid content in cultivated live feed organisms compared to marine copepods. Hydrobiologia, 358: 159-162.

- Folch J, Less M, Stanley GHS (1956). A simple method for the isolation and purification of total lipids from animal tissues. J. Biol. Chem., 226: 497-509.
- Immanuel G, Citarasu T, Sivaram V, Shankar S, Palavesam A (2007). Bioencapsulation strategy and highly unsaturated fatty acids (HUFA) enrichment in *Artemia franciscana* nauplii by using marine trash fish *Odonus niger* liver oil. Afr. J. Biotechnol., 6(17): 2043-2053.
- Kaur VI, Dhawan A (2004). Efficacy of feed types and feeding rates on the growth and survival of *Labeo rohita* larvae. In: Abstracts of Third Indian Fisheries Science Congress, pp 98.
- Kour K, Bains T (2005). Efficacy of live and dry feeds in rearing of larvae and fry of Labeo rohita (Ham.). in: Abstract book workshop on Fisheries and Aquaculture in Indus River Region conservation, management and development of indigenous fish fauna, 21-22 December 2005, PAU Ludhiana, p. 41.
- Kyungmin H, Geurden I, Sorgeloos P (2000). Enrichment strategies for *Artemia* using emulsions providing different levels of n-3 highly unsaturated fatty acids. Aquaculture, 183(3-4): 335-347.
- Lowry OH, Rosenbrough N, Fair AC, Randall RJ (1951). Protein measurement with folin phenol reagent. J. Biol. Chem., 193: 265-275.
- Merchie G, Lavens P, Kontara EK, Ramos X, Leon-Hing Kujan A, Van Hauwaert A, Pedrazzoli A, Naessens-Foucquaert E, Nelis H, De Leenheer A, Sorgeloos P (1996). Supplementation of ascorbic acid 2–phosphate during the early postlarval stages of penaeid shrimp (*Penaeus monodon* and *Penaeus vannamei*), (1996a, submitted).
- Pillay TVR (1990). Aquaculture-Principles and Practices. Fishing News Books, London, U.K.
- Prinsloo JF, Schoonbee HJ (1986). Comparison of the early larval growth rates of the Chinese grass carp (*Ctenopharyngodon idella*) and Chinese silver carp (*Hypopthalmychthys molitrix*) using live and artificial feed. Water, 12: 229-234.
- Shields RJ, Bell JG, Luizi FS, Gara B, Bromage NR, Sargent JR (1999). Natural copepods are superior to enriched *Artemia nauplii* as feed for Halibut larvae (*Hippologssus hippoglossus*) in terms of survival, pigmentation and retinal morphology: relation to dietary essential fatty acids. J. Nutr., 129(6): 1186-1194.
- Singh DK. Kumar A, Reddy AK (1994). Role of live feed in fish and prawn seed production and their culture. Fishing Chimes, July, pp. 13-16.
- Tiwari VK (1986). Live feed culture, Silver jubilee celebrations. Hi tech Aquaculture, open house, May, p.15.