

## Growth and survival of *Clarias batrachus* (Lin.) larvae fed on formulated diets

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### Abstract

An experiment was conducted for a period of 28 days using 7-day old *C. batrachus* larvae of initial total length ( $\pm$ S.D.) of  $7.4 \pm 0.49$  mm and weight ( $\pm$ S.D.) of  $2.8 \pm 0.75$  mg. Five test diets viz. diets I, II, III, IV and V were prepared. Diet I was formulated using 30% fish meal (FM), 20% soybean meal (SM) and 20% Baker's yeast (BY), diet III using 15% FM, 20% SM, 15% cotton seed meal and 30% BY and diet IV using 20% FM, 30% BY and 30% powdered milk. The remaining two diets (diets II and V) were prepared using minced clam meat (96%) and BY (90%) respectively as the sole source of protein. Three replications were used for each treatment. The larvae fed on diet II exhibited significantly ( $P < 0.05$ ) better length gain and specific growth rate (SGR) than those of the larvae fed on diet III and there was no significant difference among the length gain and SGR of the larvae fed on diet I, II, IV and V. The larvae fed on diet II also showed significantly better weight gain and survival than those of the larvae fed on other diets. There was no significant difference between the condition factor of the larvae fed on diet II and IV. However, the larvae fed on diet II showed better condition factor than those of the larvae fed on other diets.

**Key words :** *C. batrachus* larvae, Feed, Rearing

### Introduction

In order to mitigate the protein deficiency, semi-intensive and intensive fish culture are necessary in Bangladesh. Intensive culture depends on regular supply of fish fry. This calls for the development of appropriate larval feed. *Clarias batrachus* locally called 'magur' is a suitable species for intensive culture because it grows in haors, baors, paddy fields and exhibits remarkable growth performance within short time even in waste water. Though the fry of *C. batrachus* are produced successfully through induced breeding yet larval feed for successful rearing of fry exists as one of the main impediments for its intensive culture. Mollah (1987) and Mollah and Nurullah (1988) reported that *C. batrachus* larvae were successfully reared with live feed (*Tubifex* sp.). But there are several unavoidable problems associated with natural live food organisms. Availability of live food organisms depends on environmental factor, as a result

they have not been found round the year and their collection from natural habitat is laborious, unpredictable as well as time consuming. Artificial diets, appropriate for this purpose, on the other hand, can be manufactured on a large scale and can be distributed easily to ensure regular supply.

Works of Alam and Mollah (1988, 1989) on larval feed development and that of Sanaullah et al. (1986) on fingerling rearing of *C. batrachus* need mentioning. But the result were not conclusive enough compared to the works of (Hect 1981 & 1982, Hogendoorn et al. 1983, Uys and Hecht 1985, Verreth et al. 1987) on *Clarias gariepinus*. Therefore, the present experiment was designed to observe the growth and survival of *C. batrachus* larvae fed on formulated diets.

## Materials and methods

The study was conducted in the indoor laboratory of the department of Fisheries Biology and Limnology, Bangladesh Agricultural University, Mymensingh. Larvae of *C. batrachus* used in the present study were produced by induced breeding with the use of human chorionic gonadotropin (HCG) at a dose of 3 and 2 IU/g body weight of female and male respectively. When the yolk sac was completely absorbed, the larvae were fed with hard-boiled chicken egg yolk. The experiment was conducted using 7-day old larvae having an initial total length of 7.4 mm and weight of 2.8 mg.

The experiment was carried out in glass aquarium of size 44 X 24 X 22 cm<sup>3</sup>. Each aquarium was partially filled with 10 litre of water. Each of the aquaria was stocked with 100 *C. batrachus* larvae at a rate of 10 larvae/l of water. The aquaria were divided into five treatment groups (groups A, B, C, D and E) each having three replicates. Fishes of treatments A, B, C, D and E were fed respectively with diets I, II, III, IV and V.

The larvae of group A, C and D were fed with formulated diets containing fish meal, soybean meal, cotton seed meal, powdered milk, Baker's yeast, wheat flour, cod liver oil, attractant, vitamin and mineral premix (Table 1). The solid dietary ingredients were dried in the sun and ground to powder form. Fish protein concentrate of "lata fish" (*Channa striatus*) was used for this study. The required amount of powdered ingredients and oil were then measured and mixed thoroughly. The water was added to the mixture in such an amount that the feed was in paste form. The larvae of group B and E were fed with minced meat of freshwater clam (*Unio* sp.) and Baker's yeast respectively (Table 2). After removing the shells of clam, the meat was minced by using meat chopper and then supplemented with vitamin and mineral premixes. Attractant, vitamin and mineral premixes were diluted in water to make the Baker's yeast into paste form which was used as feed for group E (Table 2). All the feeds were in wet condition and were stored in deep freeze.

**Table 1.** Percentage composition of ingredients of the test diets and their proximate composition (% dry weight)

Ingredients <sup>1</sup>	Diets				
	I	II	III	IV	V
Fish meal	30.0	-	15.0	20.0	-
Soybean meal	20.0	-	20.0	-	-
Cotton seed meal	-	-	15.0	-	-
Baker's yeast	20.0	-	30.0	30.0	90.0
Powdered milk	-	-	-	30.0	-
Wheat flour	20.0	-	10.0	12.0	-
Cod liver oil	3.0	-	3.0	3.0	3.0
Attractant <sup>2</sup>	1.0	-	1.0	1.0	1.0
Vitamin premix <sup>3</sup>	3.0	2.0	3.0	2.0	2.0
Mineral premix <sup>4</sup>	3.0	2.0	3.0	2.0	4.0
Minced clam meat	-	96.0	-	-	-
<b>Proximate composition (%)</b>					
Crude protein	44.21	53.16	47.89	52.24	39.58
Lipid	9.33	4.82	15.30	13.04	7.52
Ash	7.40	38.03	14.75	9.50	9.50
NFE	39.06	3.99	22.06	25.22	43.40
Energy (Kcal/100g)	417.05	271.08	417.50	427.20	399.60

<sup>1</sup>g/100g dry diet<sup>2</sup>Sodium aspartate (salt of aspartic acid)<sup>3 and 4</sup>Composition has been given in Table 3 and 4 respectively.

Proximate composition of the dietary ingredients and test diets were determined by the method described by A.O.A.C. (1965) with slight modification (Table 2). The caloric content of the test diets were estimated by methods of Hastings (1979).

**Table 2.** Proximate composition of dietary ingredients used in five different experimental diets (% dry matter basis)

Ingredients	Dry matter	Crude protein	Lipid	Ash	Crude fibre	NFE*
Fish meal	93.00	78.91	10.56	10.53	-	-
Soybean meal	88.92	36.30	18.33	4.97	06.89	3.51
Cotton seed meal	86.60	25.20	16.86	6.50	26.25	25.19
Baker's yeast	94.60	37.87	6.59	3.21	0.08	2.25
Powdered milk	97.30	27.13	28.98	5.90	-	7.99
Wheat flour	92.14	12.48	1.32	2.11	02.14	81.95
Minced clam meat	53.16	4.82	38.03	0.43	3.56	22.80

\*NFE (Nitrogen free extract) = 100 - (crude protein + lipid + ash + crude fibre)

**Table 3.** Composition of the vitamin premix used in experimental diets.

Vitamin	g/kg of premix*	Vitamin	g/kg of premix*
Thiamin(B <sub>1</sub> )	2.50	Choline	200.00
Riboflavin (B <sub>2</sub> )	2.50	Niacin (Nicotinic acid, B <sub>3</sub> )	10.00
Pyridoxin (B <sub>6</sub> )	2.00	Cyanocobalamin (B <sub>12</sub> )	0.005
Pantothenic acid	5.00	Retinol palmitate (A)	100,000 IU
Inositol	100.00	α - tocopherol acetate (E)	20.10
Biotin	0.30	Ascorbic acid (C)	50.00
Folic acid	0.75	Menadione (K)	2.00
Para aminobenzoic acid	2.50	Cholecalciferol (D <sub>3</sub> )	500,000 IU

\*This mixture was made up to 1 kg with α-cellulose

**Table 4.** Composition of mineral premix used in experimental diets (Jauncey and Ross1982)

Minerals	Chemical formula	g/kg
Calcium orthophosphate	CaHPO <sub>4</sub> · 2H <sub>2</sub> O	727.7775
Magnesium sulphate	MgSO <sub>4</sub> · 7H <sub>2</sub> O	127.5000
Sodium chloride	NaCl	60.0000
Potassium chloride	KCl	50.0000
Iron sulphate	FeSO <sub>4</sub> · H <sub>2</sub> O	25.0000
Zinc sulphate	ZnSO <sub>4</sub> · 4H <sub>2</sub> O	5.5000
Manganese sulphate	MnSO <sub>4</sub> · 4H <sub>2</sub> O	2.5375
Copper sulphate	CuSO <sub>4</sub> · 5H <sub>2</sub> O	0.7850
Cobalt sulphate	CoSO <sub>4</sub> · 7H <sub>2</sub> O	0.4775
Calcium iodate	CaIO <sub>3</sub> · 6H <sub>2</sub> O	0.2950
Chromic chloride	CrCl <sub>3</sub> · 6H <sub>2</sub> O	0.1275

The experiment was conducted for 28 days. The larvae were fed thrice daily between 0800 hr. and 2000 hr. The diets supplied in excess of satiation were settled at the bottom of each aquarium. The faeces in aquaria were removed by siphoning and the dead larvae (if any) were removed and counted in the morning and in the evening prior to each feeding. About two-third of the water was replaced after each cleaning. The water in the aquaria was aerated by laboratory air pump. Larval growth in length (mm) and weight (mg) were measured when the experiment was terminated after four weeks. The data expressed as percentage on survival rates were made arcsine transformation (Zaman *et al.* 1982) before statistical analysis. Statistical analysis of the data were done by one-way analysis of variance (ANOVA) and Duncan's multiple range test (Gomez and Gomez 1984) to determine differences in treatment means.

## Results

The growth and survival rates of *C. batrachus* larvae in response to different formulated diets are presented in Table 5. The results showed that the larvae fed on diet containing cotton seed meal (group C) exhibited significantly lower length gain ( $P < 0.05$ ) and specific growth rate than those of the larvae of other groups. However, no significant difference was observed among the length gain and specific growth rate of the larvae of groups A, B, D and E. The larvae of group B attained better length gain and specific growth rate than those of the rests. The larvae of group B showed significantly better weight gain ( $P < 0.05$ ) than those of the larvae of other groups. However, the larvae of groups A, D and E showed significantly better weight gain than those of the larvae of group C and no significant difference ( $P > 0.05$ ) was observed among the weight gain of the larvae of groups A, D and E. The larvae of group B showed better condition factor than those of the larvae of other groups. There was no significant difference between the condition factor of the larvae of group B and D and same was the case among groups A, C, D and E (Table 5).

**Table 5.** Growth parameters and survival rate of *C. batrachus* larvae fed on five different diets at the end of the 28 days experimental period

Treatment Parameters	Group A	Group B	Group C	Group D	Group E	± S.E.
Initial total length (mm)	7.40	7.40	7.40	7.40	7.40	-
Final total length (mm)	22.42 <sub>a</sub>	23.42 <sub>a</sub>	13.17 <sub>b</sub>	21.99 <sub>a</sub>	20.72 <sub>a</sub>	1.36
Length gain (mm)	15.02 <sub>a</sub>	16.02 <sub>a</sub>	5.77 <sub>b</sub>	14.59 <sub>a</sub>	13.32 <sub>a</sub>	1.36
Initial weight (mg)	2.80	2.80	2.80	2.80	2.80	-
Final weight (mg)	100.67 <sub>b</sub>	152.92 <sub>a</sub>	20.17 <sub>c</sub>	104.18 <sub>b</sub>	78.40 <sub>b</sub>	12.17
Weight gain (mg)	97.87 <sub>b</sub>	150.12 <sub>a</sub>	17.37 <sub>c</sub>	101.38 <sub>b</sub>	75.60 <sub>b</sub>	12.17
Specific growth rate (%/day/fish)	10.90 <sub>a</sub>	13.83 <sub>a</sub>	6.74 <sub>b</sub>	12.82 <sub>a</sub>	11.52 <sub>a</sub>	1.12
Condition factor	0.88 <sub>b</sub>	1.11 <sub>a</sub>	0.84 <sub>b</sub>	0.99 <sub>a</sub>	0.85 <sub>b</sub>	0.05
Survival (%)	11.38 <sub>b</sub>	39.23 <sub>a</sub>	10.07 <sub>b</sub>	22.66 <sub>b</sub>	15.33 <sub>b</sub>	4.60

Figures in the same row with same letters are not significantly different at  $P < 0.05$ .

The larvae fed on minced clam meat (group B) showed significantly higher ( $P < 0.05$ ) survival rate than those of the rests and there was no significant difference among the survival rates of the larvae of group A, C, D and E. The growth and survival rate of the larvae which were fed with diet containing cotton seed meal was very low.

The mortality of the larvae of groups A, B, C, D and E were 3, 25, 29, 18 and 6% respectively during 1st 7 days. The mortality of the larvae of four groups (group A, C, D and E) ranged from 56-81% during 14 days whereas the larvae fed on minced clam meat (group B) showed only 9.67% mortality.

But mortality of the larvae of group B was highest (60.77%) on 28 days of observation. The larvae fed on diet containing cotton seed meal showed 81.67% mortality at day 14. The larvae of other three groups (group A, D and E) showed higher mortality. The larvae of groups A, C, D and E showed similar trend in mortality upto the end of the experiment.

## Discussion

Five diets were tested for a period of 28 days to develop a suitable artificial feed for *C. batrachus* larvae. Except the cotton seed meal based diet (group C), all the diets were readily accepted by the larvae. However, the minced clam meat was accepted more by the larvae than the other diets. The larvae fed on minced clam meat (group B) exhibited better growth response and survival rate followed by the larvae fed on diet containing yeast and powdered milk (group D). The larvae of group B showed an average weight gain, specific growth rate and condition factor of 150.12, 13.83 and 1.11mg respectively. This result contradicts with the findings of Alam and Mollah (1989). In their study they found that minced clam meat gave an average weight gain, specific growth rate and condition factor of 27.50, 7.45 and 0.86mg respectively in 20 days feeding trial (25 days after hatching). The causes of such better growth rates in the present study might be due to supplementation of clam meat with growth factors such as vitamins and minerals, better acceptability of the food to the larvae and also the duration of the feeding trial. However, the weight gain of the larvae of group B was comparable to the findings of Dabrowski *et al.* (1984). According to the authors, coregonid larvae fed on *Artemia* attained an individual weight of 161 mg within 35 days. But the weight gain of the larvae of group B was better than the findings of Verreth *et al.* (1987) and Bairage *et al.* (1988). The survival rate of the larvae fed on minced clam meat was higher than the survival rate reported by Alam and Mollah (1989). Coregonid larvae fed on single cell protein based dry diets showed individual weight of 98-107 mg (Dabrowski *et al.* 1984) which was comparable to weight gain of the larvae of group D of the present study. But the growth rates of the larvae of group D was better than the findings of Verreth *et al.* (1987) and Bairage *et al.* (1988).

The diet containing 30% yeast and 30% milk (group D) gave survival rate of 22.66% which was comparable to the result reported by Dabrowski *et al.* (1978) and Dabrowska *et al.* (1979) and inferior to the survival rate reported by Winfree and Stickney (1984), Dabrowski *et al.* (1984), Uys and Hecht (1985) and Alam and Mollah (1989). The larvae fed on diet containing cotton seed meal showed

very poor growth and higher mortality. The cause of such poor growth and higher mortality was perhaps due to lower acceptability of the diet to the larvae from the starting of the feeding trial. Dorsa *et al.* (1982) reported that diet containing more than 17.4% cotton seed meal inhibited the growth of channel catfish. The larvae of groups A, D and E exhibited higher mortality although their feeds were readily accepted by the larvae.

Madhury and Mollah (1990) assumed that lack of growth factor in yeast diet might be responsible for lower survival at the end of the feeding trial. But in the present study, yeast supplemented with vitamins and minerals gave better growth rates than the larvae fed on yeast only reported by Madhury and Mollah (1990). However, the survival showed a decreasing trend in this study.

The larvae fed on minced clam meat showed 22.33% mortality at day 21. Highest mortality was observed between day 15 and 20 may be due to deficiency of growth factors such as vitamins in the clam (Alam and Mollah 1989). Although the minced clam meat were supplemented with vitamins and minerals in the present study, the larvae showed highest mortality between day 21 and 28. It is, therefore, an indication that supplementation of growth factors although increased the growth of the larvae, it had no significant effect on the mortality of the larvae. It was also true for the larvae fed on yeast only.

During 28 days of experimental period, it was observed that the larvae fed on minced clam meat showed least mortality up to 14 days (9.67%). However, mortality in all treatments showed increasing from 14 days up to the end of the experiment. The cause of such higher mortality at the end of the experiment could not be ascertained. Future experiment may be carried out to investigate the cause of such higher mortality.

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