



Effect of dietary protein levels on growth, feed utilization and carcass composition of endangered bagrid catfish *Horabagrus brachysoma* (Gunther 1864) fingerlings

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Abstract

An 84-day feeding trial was conducted to study the effect of different levels of dietary protein, 250 (P25), 300 (P30), 350 (P35), 400 (P40) and 450 g (P45) kg⁻¹ dry matter (DM) on growth, feed intake, feed utilization and carcass composition of bagrid catfish *Horabagrus brachysoma* fingerlings. Triplicate groups of fingerlings with mean initial body weight of 2.2 g were fed the experimental diets twice daily, till satiation, in 150-L tanks supplied with flow-through freshwater. Daily dry matter intake by the fingerlings decreased significantly ($P < 0.05$) when fed P25 diet, containing 250 g protein kg⁻¹. The highest body weight gain, specific growth rate (SGR) and protein efficiency ratio (PER), and the lowest feed conversion ratio (FCR) were observed in fish fed 350 g protein kg⁻¹ diet. The fish fed with P45 diet had the lowest ($P < 0.05$) carcass lipid content. The polynomial regression analysis indicates that *H. brachysoma* fingerlings require 391 g dietary crude protein kg⁻¹ diet.

KEY WORDS: carcass composition, dry matter intake, growth, *Horabagrus brachysoma*, protein requirement, threatened catfish, yellow catfish

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Introduction

Horabagrus brachysoma, popularly known as yellow catfish, is an endangered (CAMP report, 1998) tropical bagrid freshwater catfish endemic to Indian subcontinent (Kurup *et al.* 2004). This moderately elongated bony catfish is very popular among consumers and fetches high market price

owing to its less intramuscular bones (unpublished data). The aquaculture of this species has recently been emphasized for its demand as a food fish as well as an ornamental fish (Ali *et al.* 2007). The unspecialized as well as flexible feeding habit and ability to feed at different trophic levels make this species a potential candidate for aquaculture (Sreeraj *et al.* 2006). The induced spawning of *H. brachysoma* is well practiced in Asian countries, India, Thailand and Vietnam, but its aquaculture is not getting momentum because of non-availability of adequate number of stocking materials. The success of availability of seed for aquaculture of any fish species depends on the availability of suitable feeds that are readily consumed, efficiently digested and utilized for growth and survival (Giri *et al.* 2002). The nutritional requirements of this important omnivorous catfish (Prasad & Ali 2008) have not been studied for any life history stages. This information is required for formulating feeds for optimum growth at the lowest cost.

The dietary protein requirement of a species is of prime importance in aquaculture, because feed protein influences growth of the fish and determines the cost of feeding. The quantity as well as the quality of dietary protein is the determinants of the level of protein utilization by the fish. Therefore, studies on protein requirement are usually one of the first nutrient requirement experiments conducted when a new fish species is introduced in to aquaculture. To quantify nutrient requirement, semi-purified or purified diets are generally used. Very often, practical diets are also formulated to quantify the protein requirement in different catfishes, *Ictalurus punctatus* (Li & Lovell 1992; Li *et al.* 2008), *Clarias isheriensis* (Fagbenro 2007), hybrid catfish (Adebayo & Alasoadura 2001; Giri *et al.* 2003) and *Mystus nemurus* (Khan *et al.* 1993, 1996).

Given this lack of information and considering the basic nutrient requirements of *Horabagrus brachysoma* fingerlings are unknown, an experiment was planned to study the effect

of feeding different levels of protein, in practical type of feeds, on growth, nutrient utilization and eviscerated body carcass composition of this species. Also, the dietary protein requirement for optimal growth of yellow catfish was determined.

Materials and methods

Diet preparation

Five iso-energetic (16.5 ± 0.20 MJ kg⁻¹) practical diets P25, P30, P35, P40 and P45 were formulated to contain 250, 300, 350, 400 and 450 g crude protein (CP) kg⁻¹, respectively (Table 1). Because digestible or metabolizable energy values for these feed ingredients have not been determined with *H. brachysoma*, standard mammalian physiological fuel values, 16.7, 37.6 and 16.7 kJ g⁻¹ for protein, lipid and carbohydrates, respectively, were utilized to calculate the energy content of diets. Earlier, these mammalian physiological fuel values of nutrients were also utilized to calculate the dietary energy value for channel catfish (Gatlin *et al.* 1986).

The powdered major feed ingredients were mixed thoroughly for each diet, pressure-cooked at 2.7 kg pressure cm⁻² for 15 min, cooled, fortified with a vitamin–mineral mixture and oil and blended. The dough, so obtained, was extruded through a 1.0-mm-diameter die in a feed pelletizer. The

resultant pellets were dried overnight at 45 °C, crumbled and stored frozen (–20 °C) in airtight plastic jars.

Fish and feeding

The catfish fingerlings were obtained from the fish farm, at this institute. The fingerlings were acclimatized to the laboratory condition in 200-L capacity circular cement tanks that were attached with flow-through water system. Groups of 20 fingerlings of average 2.20 ± 0.01 g body weight were stocked in 15 cement tanks (150 L), and triplicate tanks were allotted for each dietary treatment. All the tanks were plumbed with flow-through system, and the flow rate of water was maintained at 1 L min⁻¹, during experiment. To ensure oxygen saturation, the water in each tank was continuously aerated by air stones. Daily water quality parameters, temperature, pH, dissolved oxygen, NH₄⁺ ranged from 29–30 °C, 7.8–7.9, 6.0–6.3 and 0.20–0.22 g L⁻¹, respectively, and there was no significant influence of dietary treatments on these parameters. The water quality parameters were within the acceptable range, reported for other catfish rearing and culture (Viveen *et al.* 1986; Rao *et al.* 1994; Giri *et al.* 2003, 2009).

For 84 days, fish were fed to satiation twice daily, at 9 a.m. and 4.30 p.m. For feeding, the known quantity of feed was offered in each tank, and 45 min after each offering, the remaining of the feeds was siphoned out and dried over night at 105 °C in a laboratory hot air oven. Daily intake of dry matter (DM) was determined by subtracting the residue from the offered feeds. At the end of the experiment, individual body weight of all the fish as well as the final biomass per tank was recorded. All the fish in each group were sacrificed, and viscera were quickly removed. The eviscerated fish were dried in the hot air oven, ground and stored in plastic jars for proximate analysis.

Growth performance and feed utilization efficiency were assessed by recording final body weight, determining net body weight gain, specific growth rate (SGR), feed intake, feed conversion ratio (FCR) and protein efficiency ratio (PER) as follows:

$$\text{Net weight gain (g)} = W_f - W_i$$

$$\text{Specific growth rate} = 100 (\ln(W_f) - \ln(W_i)) T^{-1}$$

$$\text{Feed conversion ratio} = W_{\text{TFC}} (W_f - W_i)^{-1}$$

$$\text{Protein efficiency ratio} = (W_f - W_i) W_{\text{prot},f}^{-1}$$

where W_i and W_f are the initial and final body weight (g), W_{TFC} is the weight of feed consumed (g), T is duration of the experiment (days) and $W_{\text{prot},f}$ is the weight of intake of dietary crude protein.

Table 1 Dietary formulation (g kg⁻¹) and chemical analysis (g kg⁻¹ dry matter) of the experimental diets

Ingredients	P25	P30	P35	P40	P45
Fishmeal	150	200	250	300	450
Groundnut oil meal	60	120	190	250	190
Soybean meal	100	100	100	100	100
Maize (ground, yellow)	510	400	280	180	90
Yeast (dry)	40	40	40	40	40
Vegetable oil ¹	80	80	80	70	70
Mineral and vitamin mix ²	30	30	30	30	30
Carboxy methyl cellulose (binder)	30	30	30	30	30
Chemical composition ³					
Organic matter	893	883	874	864	835
Crude protein (estimated)	250	300	350	400	450
Crude protein (analysed)	253	301	353	403	454
Crude lipid	113	116	119	112	112
Total ash	107	117	126	136	165
Total carbohydrate	504	441	374	318	246
Energy (MJ kg ⁻¹)	16.9	16.7	16.6	16.3	16.0
Protein: energy ratio (mg kJ ⁻¹)	15.0	18.0	21.3	24.7	28.4

¹ Soybean oil : groundnut oil : mustard oil = 1 : 1 : 1.

² See Giri *et al.* (2000b).

³ Values are mean of triplicate analysis.

Analysis

The triplicate samples of diets and carcass were analysed using standard methods (AOAC 1990). Moisture was determined by drying at 105 °C for 24 h in a hot air laboratory oven; crude protein ($N \times 6.25$) by Kjeldahl method; crude lipid (CL) after extraction in petroleum ether by Soxhlet method; total ash by incinerating at 550 °C for 4 h in a muffle furnace. Organic matter (OM) was calculated by subtracting total ash value from DM. The nitrogen-free extract (NFE) of feeds was considered as total carbohydrates (TCHO) that is assimilated by the fish, and this is calculated by subtracting CP, CL and crude fibre (CF) from OM.

Data were presented as mean \pm standard error of triplicate groups. A one-way analysis of variance model was used to test differences between dietary treatments. When appropriate, means were evaluated for significance by the multiple range tests of Tukey. The second-order polynomial regression model was used to estimate minimum dietary protein level supporting optimum growth. All statistical tests were performed using GraphPad Prism version 3.00 for Windows, GraphPad Software, San Diego, CA, USA, 'http://www.graphpad.com'.

Results

Final body weight, net biomass gain, specific growth rate (SGR), daily feed intake, feed conversion ratio (FCR) and

Table 2 Growth and feed utilization of *Horabagrus brachysoma* fingerlings fed different levels of protein for 12 weeks

Attributes	P25	P30	P35	P40	P45
Initial weight (g fish ⁻¹)	2.17 ± 0.04	2.15 ± 0.03	2.25 ± 0.01	2.22 ± 0.02	2.20 ± 0.01
Final weight (g fish ⁻¹)	2.87 $\pm 0.09^a$	7.37 $\pm 0.12^b$	8.73 $\pm 0.13^c$	8.53 $\pm 0.13^c$	8.53 $\pm 0.22^c$
Net biomass gain (g fish ⁻¹)	0.70 $\pm 0.13^a$	5.22 $\pm 0.12^b$	6.48 $\pm 0.13^c$	6.32 $\pm 0.14^c$	6.55 $\pm 0.03^c$
Specific growth rate (SGR)	0.33 $\pm 0.06^a$	1.47 $\pm 0.02^b$	1.62 $\pm 0.02^c$	1.60 $\pm 0.03^c$	1.61 $\pm 0.03^c$
Feed intake (FI)	0.50 $\pm 0.09^a$	1.62 $\pm 0.01^b$	1.61 $\pm 0.04^b$	1.64 $\pm 0.03^b$	1.78 $\pm 0.06^b$
Feed conversion ratio (FCR)	6.47 $\pm 0.15^a$	2.81 $\pm 0.08^b$	2.35 $\pm 0.06^c$	2.42 $\pm 0.03^{b,c}$	2.52 $\pm 0.07^{b,c}$
Protein efficiency ratio (PER)	1.62 $\pm 0.04^a$	0.84 $\pm 0.02^d$	0.82 $\pm 0.02^d$	0.96 $\pm 0.01^c$	1.13 $\pm 0.03^b$

Values are the mean \pm SE of triplicate groups of 20 fishes.

^{a,b,c,d} Values in the same row with different letters differ significantly ($P < 0.05$).

FI = % Initial body weight day⁻¹.

SGR = $100(\ln(W_f) - \ln(W_i)) T^{-1}$.

FCR = $W_{TFC} (W_f - W_i)^{-1}$.

PER = $(W_f - W_i) W_{prot,f}^{-1}$.

protein efficiency ratio are presented in Table 2. The initial body weight of each group of fish was alike. In all the treatments, except P25, fish grew more than triple of their initial body weight during experiment. The final body weight, net biomass gain, daily weight gain and SGR were significantly influenced ($P < 0.05$) by the dietary treatments. For all these parameters, the value recorded in P25-fed fish was the lowest, followed by P30; however, both were lower than that of P35-, P40- and P45-fed fish. Best growth was obtained with diets containing protein levels 350 (P35), 400 (P40) and 450 (P45) g kg⁻¹ with no significant difference ($P > 0.05$) between them. Based on the second-order polynomial regression analysis of final body weight against dietary protein levels, an optimum protein requirement for best growth of yellow catfish fingerlings was calculated as 391 g kg⁻¹ diet (Fig. 1).

In different dietary groups, the daily feed intake varied between 0.50 and 1.78% of their initial body weight, with the lowest ($P < 0.05$) in P25-fed group. The FCR showed a significant improvement ($P < 0.05$) as dietary protein level increased from 250–300 g kg⁻¹ and reached a plateau thereafter (Table 2). Although not statistically significant, the fish fed with P35 diet showed better FCR in comparison with other protein levels. Protein efficiency ratio (PER) values ranged between 0.82 and 1.62, with the lowest ($P < 0.05$) in groups fed P30 and P35 diets.

The data of eviscerated body carcass analysis were presented in Table 3. The level of dietary protein did not significantly ($P > 0.05$) influence the carcass moisture and protein contents of fingerlings. The carcass lipid content of fish fed P25–P40 were similar to each other and also were significantly higher ($P < 0.05$) than that of fish fed the

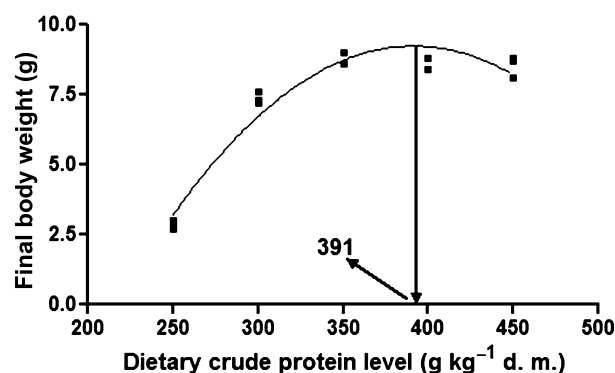


Figure 1 The effect of dietary crude protein level on final body weight of *Horabagrus brachysoma*. The dietary protein requirement for *H. brachysoma* fingerlings calculated by second-order polynomial regression is 391 g kg⁻¹ ($y = -37.02 + 0.2363x - 0.0003019x^2$; $R^2 = 0.95$).

Table 3 Body composition (g kg⁻¹ eviscerated wet weight) of *Horabagrus brachysoma* fingerlings fed experimental diets with increasing levels of dietary protein for 12 weeks

Parameters	P25	P30	P35	P40	P45
Moisture	731 ± 4	735 ± 2	737 ± 3	735 ± 2	744 ± 6
Protein	166 ± 2	162 ± 2	166 ± 2	167 ± 2	167 ± 3
Lipid	59 ± 4 ^a	56 ± 2 ^a	56 ± 2 ^a	51 ± 1 ^a	42 ± 1 ^b
Ash	40 ± 1 ^a	36 ± 1 ^b	37 ± 1 ^{a,b}	37 ± 0 ^{a,b}	40 ± 1 ^a

Values are the mean ± SE of triplicate groups of 20 fishes.

^{a,b} Values in the same row with different letters differ significantly ($P < 0.05$).

highest protein containing diet, P45. The carcass total ash content was significantly influenced by different dietary treatments and ranged between 36 and 40 g kg⁻¹.

Discussion

The protein concentration in the diets increased steadily from P25 to P45 diets, which was necessary to study the protein requirement of yellow catfish. Because of protein levels increased, to compensate this nutrient gaps, some other constituents must vary among the diets, and in the current study the total carbohydrate concentration decreased steadily from P25 (504 g kg⁻¹) to P45 (246 g kg⁻¹). This decrease in carbohydrate concentration was because of decreased levels of maize in the diets. The total ash content of diets increased from P25–P45, which was because of the presence of higher levels of fish meal as a major feed ingredient.

Although the initial body weight was similar among the treatments, the final body weight attained by P25 was the lowest, followed by P30, and both were lower than that of P35-, P40- and P45-fed fishes. One of the reasons for this inferior growth performance of P25 group of fish could be because of less daily dry matter consumption by the fish, which might be possibly because of poorer palatability of this low protein and less fish meal containing diet. Martinez-Palacios *et al.* (2007) in *Menidia ester* also observed similar decrease in daily feed intake in response to dietary protein level below 300 g kg⁻¹ diet. Daily dry matter intake by P30–P45-fed fishes did not vary significantly ($P > 0.05$). Therefore, another reason for inferior growth performance of fish fed on the P25 and P30 diets could be because of higher levels of carbohydrates in their diets, which supports to the observations of Jantrarotai *et al.* (1994) in hybrid catfish, Giri *et al.* (2000a) in *Clarias batrachus*, Giri *et al.* (2003) in hybrid catfish and Martinez-Palacios *et al.* (2007) in Mexican silverside, *Menidia ester*. The high-carbohydrate diet decreases enzyme activities and digestibility of carbohydrate

and protein, which culminated in retarded growth in fish (Ufodike & Matty 1983). In the present study, it was observed that yellow catfish were possibly capable of tolerating up to 374 g carbohydrate kg⁻¹ diet (P35) without decrease in their growth performance, and this value was higher than the values reported for channel catfish (280 g kg⁻¹) (Garling & Wilson 1977) but equal to the value reported for hybrid catfish of *C. batrachus* × *C. gariepinus* 373 g kg⁻¹ (Giri *et al.* 2003) and less than the values reported for Nile tilapia (400 g kg⁻¹) (Anderson *et al.* 1984), hybrid catfish of *C. macrocephalus* × *C. gariepinus* (500 g kg⁻¹) (Jantrarotai *et al.* 1994) and *C. batrachus* (511 g kg⁻¹) (Giri *et al.* 2000a). The second-order polynomial regression analysis of final body weight of fingerlings indicated the dietary protein requirement for *H. brachysoma* fingerlings was 391 g kg⁻¹ with corresponding energy value of about 16.6 MJ kg⁻¹ diet. This predicted optimum dietary protein requirement value of *H. brachysoma* is similar to the protein requirement of other tropical catfish, particularly *Clarias gariepinus*, 400 g kg⁻¹ (Degani *et al.* 1989), hybrid catfish of *C. macrocephalus* × *C. gariepinus*, 400 g kg⁻¹ (Jantrarotai *et al.* 1998) and bagrid catfish *Mystus nemurus* (400 g kg⁻¹) (Ng *et al.* 2001). However, the comparison of growth performance attained by yellow catfish in the present study is not possible because of non-availability of published data with this species.

The SGR values were the lowest in fish fed P25, followed by P30, and they both were lower than that of other three protein levels. The SGR value did not vary among P30–P45 treatments, and the values were similar to that reported for *Menidia ester* (Martinez-Palacios *et al.* 2007) and juvenile *Melanogrammus aeglefinus* (Kim *et al.* 2001).

Although fish fed to satiation, the daily DM intake by P25-fed fishes was the lowest ($P < 0.05$) in comparison with that of other treatments. This might be the reason that the FCR and PER values were varied significantly in response to dietary protein contents. This increased PER in P30–P45-fed fish with increased dietary protein level has also been observed in *Cyprinus carpio* (Ogino & Saito 1970), *Hypophthalmichthys molitrix* (Singh 1990) and *Sparus auratus* (Santinha *et al.* 1996).

The carcass protein content of *H. brachysoma* did not vary ($P > 0.05$) in response to dietary protein levels, which agrees to the similar observations in another bagrid catfish *M. nemurus* (Ng *et al.* 2001) when fed diets containing 200–500 g protein kg⁻¹. The carcass lipid content of yellow catfishes fed 250–400 g CP kg⁻¹ was similar to each other and were significantly higher than that of fish fed 450 g CP kg⁻¹ diet, which is consistence with the earlier observations in *Sarotherodon mossambicus* (Jauncey 1982), *Cichlasoma*

urophthalmus (Martinez-Palacios *et al.* 1996) and *Mystus nemurus* (Ng *et al.* 2001). Several studies have shown that body fat in catfish increases when dietary protein concentration decreases as protein/energy ratio decreases (Reis *et al.* 1989; Li & Lovell 1992). In the present study, no consistent trend of dependence of the carcass ash content was observed in response to dietary protein levels.

Data from the present study indicated that for maximum growth and best feed utilization, *H. brachysoma* fingerlings required 350 g crude protein at an energy level of 16.6 MJ kg⁻¹ DM and a protein to energy ratio of 21.3, when fish meal, groundnut oil meal and soybean meal were used as primary protein sources. The second-order polynomial regression graph suggests that 391 g CP kg⁻¹ diet to be optimum for best performance of catfish fingerlings.

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