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Effects of Dietary Protein Levels on Growth, Feed Utilization, Protein Retention Efficiency of *Tor tor* (Hamilton, 1822)

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Abstract: During the present experiment, *Tor tor* fry with an average length and weight of 20.00 mm and 0.338 g, respectively were subjected to three experimental feeds containing 25% CP (T_1), 30% CP (T_2) and 40% CP (T_3) with a control group fed on feed containing 20% CP. The results revealed highest increment in overall length and weight in case of T_3, with as high as 86.3±2.2 mm and 5.329±1.11 g, over a period of 12 months (365 days). Similarly, highest values for SGR (0.328±0.03) and PER (0.846±0.02) were recorded in case of T_3. The best feed composition as T_3 was documented finally by the lowest FCR value of 0.82±0.02.

Key words: *Tor tor*, crude protein, SGR, FCR, PER

INTRODUCTION

Tor tor is the most common Himalayan mahseer and a very attractive sport fish with excellent food value. *Tor tor* is a highly nutritious fish with good economic value. It shows a steady decline in abundance in reservoirs in India. It inhabits riverine pools and lakes and also in streams with good flows and a rocky bottom where they attain the best growth. They are benthopelagic, potamodromous and occur in tropical freshwaters (15-30°C) at depths of upto 15 m. Adults have an omnivorous feeding habits and feed on small fish, insects, molluscs, zooplankton, debris, sand, mud, fish scales and bones, fruits, chironomid larvae, water beetles, crustaceans, filamentous algae and macrophytes. Juveniles mainly consume insects (Desai, 2003). Among Indian mahseers, *Tor tor* (Hami Hon, 1822) is the most important food and game fish of India after *Tor putitora* (Hamilton, 1822).

Studies on the nutritional aspects from culture viewpoint related to conservation and propagation, though important are very limited particularly for Narmada mahseer. Attempts have been made to raise fry of putitora mahseer on formulated diets at NRCCWF while in case of khudree mahseer more systematic studies have been conducted to evaluate the optimum protein requirement (Murthy and Keshavanth, 1986) protein sources (Keshavanath *et al*., 1986) and protein sparing effect of sardine oil (Bazaz and Keshavanath, 1993). During different stages of mahseer development, the protein requirement by this species needs to be understood for modifying/formulating the feed in order to make it more balanced and nutritive.

The present study has aimed to generate baseline data on nutrient requirements so as to develop appropriate feeds to enhance growth of mahseer in captivity under aquaculture conditions so as to increase its production. At present there is only limited knowledge on the nutritional requirement of mahseer *Tor tor* when cultured in captivity in tropical waters. Information on the nutritional requirement of fish and its availability from different sources is essential for formulation of complete feed. The present research describes, the nutritional requirement for the development of fisheries and aquaculture of *Tor tor*. The main aim of the study is to find out which formulated feed with low cost is more beneficial to obtain fast and better growth rate of this particular fish species when cultured in captivity in tropical climatic conditions. It will help to evaluate dietary requirement of fry to adult stage of *Tor tor*, so as to formulate nutritionally balanced diets.

MATERIALS AND METHODS

Growth is a useful integrated index of physiological status of an organism. The present study was carried out from July, 2005 to June, 2006 in two experimental trials to evaluate the change in growth of *Tor tor* during its rearing on various feeds. The experiments were conducted in order to identify suitable formulated feed having different protein contents for rearing mahseer (*Tor tor*) ranging from fry to advanced fingerling stage in captivity in tropical climatic conditions and the results were compared with the natural conditions.

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Treatments:

T_1 : Fishes fed with feed containing 25% protein level
T_2 : Fishes fed with feed containing 35% protein level
T_3 : Fishes fed with feed containing 40% protein level
T_4 : Fishes fed with the Available Commercial Feed (ACF)

For evaluating the growth performances, the following methodology was used.

Methods for the study of fish growth

Net length/weight increment: Net length and weight of fish was calculated as under:

Calculations:

$$\text{Net increment in length} = L2 - L1$$

$$\text{Net increment in weight} = W2 - W1$$

Where:
L1 = Length of fish at time T_1 (initial time)
L2 = Length of fish at time T_2 (final time)
W1 = Weight of fish at time T_1 (initial time)
W2 = Weight of fish at time T_2 (final time)

Percent length/weight increment: Percent increment in the length and weight of fish was calculated by employing the following equation:

$$\text{Increment in length}\,(\%) = \frac{L2 - L1 \times 100}{L1}$$

$$\text{Increment in weight}\,(\%) = \frac{W2 - W1 \times 100}{W1}$$

Per day length/weight increment: Per day increment in length/weight of fish was calculating by employing the following equation:

$$\text{Per day increment in length} = \frac{L2 - L1}{T_2 - T_1}$$

$$\text{Per day increment in weight} = \frac{W2 - W1}{T_2 - T_1}$$

Data analysis for FCR, SGR and PER, the following formulae were employed:

$$\text{Feed conversion ratio} = \frac{\text{Amount of dry feed consumed}}{\text{Live weight gain}}$$

$$\text{Specific growth rate} = \frac{\begin{matrix}(\text{Log final body weight} - \\ \text{Log initial body weight}\end{matrix}}{\text{Number of days}} \times 100$$

Table 1: Formulation of three different diets and an available commercial feed (1 kg) for *Tor tor* having different protein levels

	Experimental diets (%, g)			
Ingredients	Feed I (25% P)	Feed II (35% P)	Feed III (40% P)	ACF (20%, g)
Fish meal	200	350	400	50
Soya cake	400	400	350	100
GNOC	200	50	-	-
Wheat bran	50	50	100	500
Rice bran	50	50	50	250
Soya oil	20	20	20	-
Molasses	60	60	60	80
Mineral mixture	20	20	20	20

Available Commercial Feed (ACF); GNOC = Groundnut Oil Cake; The formulated feeds were prepared at the Central Institute of Agricultural Engineering at Bhopal (CIAE)

$$\text{Protein efficiency ratio} = \frac{\text{Gain in body weight}}{\text{Protein intake}}$$

Length-weight relationship: The general parabolic equation of the form, $W = aL^3$ (Le Cren, 1951) which explains the length-weight relationship in fishes was used in the present study. This equation written in the linear form as:

$$Y = a + bx$$

Where:
Y = Log W
x = Log L

Hence, a and b were estimated empirically by the method of least square.

Preparation of formulated feeds: In the present study for preparing feed, the main ingredients used were, fishmeal, soyabean cake, ground nut oil cake and soya oil as protein source, wheat bran and rice bran as carbohydrate source, molasses and mineral mixture and topica as binder (Table 1).

Feed preparation: All the locally available ingredients were dried in a hot air oven at 60°C for 24 h. All the ingredients were ground into powdered form by electrically operated grinder. The binder (Tapioca) was also ground in an electrically operated grinder before preparation of pellets.

Feed formulation: Feed formulation was done basically by square method in order to initially balance the basal feed and protein supplement. Proportion of each ingredient required was calculated precisely providing allowance for the premix (Rath, 2000).

RESULTS

The results of the present findings revealed that *Tor tor* fry with an average length of 20.00 mm and

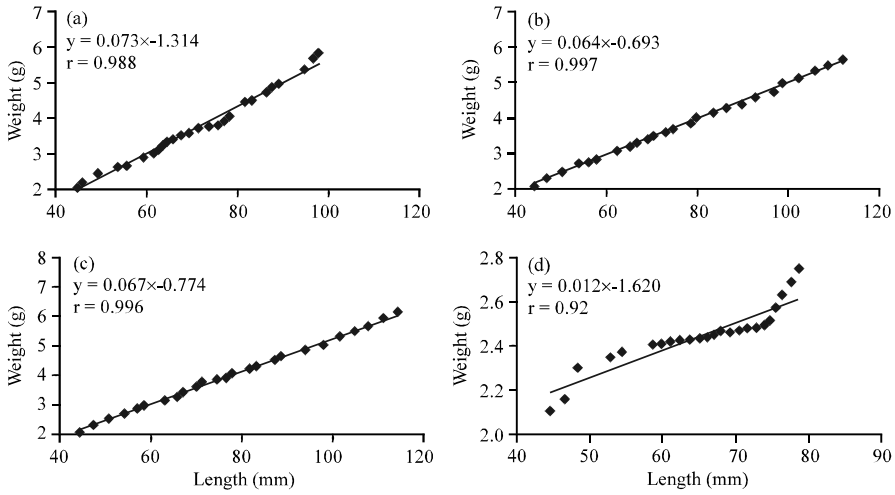

Fig. 1: Length-weight relation for different protein rations fed to *Tor tor* fry: a) T_1; CP = 25%; b) T_2; CP = 30%; c) T_3; CP = 40%; d) T_4; CP = 20%

Table 2: Mean growth performance, feed and nutrient efficiency of *Tor tor* fed various protein rations for 365 days

	Diet No.				
Parameters	T_1	T_2	T_3	T_4	Mean±SE
Initial length (mm)	20	20	20	20	0±0.0
Final length (mm)	68.9±2.2	68.6±1.9	86.3±2.2	47.2±1.6	8.00±1.20
Percent length	244.5±12.8	243±15.2	331.5±14.6	136±10.5	40.01±8.23
Initial weight (g)	0.338	0.338	0.338	0.338	0±0.0
Final weight (g)	3.34±0.52	3.72±0.92	5.329±1.11	1.928±0.26	0.699±0.03
Percent weight	888.16±12.6	1000.59±25.4	1476.62±29.2	470.41±14.8	206.8±12.8
Initial per day length (mm)	0.067	0.067	0.067	0.067	0±0.0
Final per day length (mm)	0.135±0.05	0.135±0.03	0.135±0.02	0.135±0.03	0±0.0
Initial per day weight (g)	0.0009	0.0009	0.0009	0.0009	0±0.0
Final per day weight (g)	0.0216±0.02	0.0216±0.01	0.0216±0.01	0.0216±0.01	0±0.0
Specific growth rate (g% day^{-1})	0.272±0.05	0.285±0.04	0.328±0.03	0.207±0.04	0.025±0.01
Food conversion ratio	1.3±0.02	1.12±0.03	0.82±0.02	1.8±0.03	0.21±0.01
Protein energy ratio	0.428±0.02	0.585±0.01	0.846±0.02	0.083±0.02	0.159±0.01

Values are means±SD of three replications (df 4, 35)

weight of 0.338 g, subjected to three formulated diets containing 25% protein (T_1), 35% protein (T_2) and 40% protein (T_3) showed considerable changes in growth parameters in contrast to T_4 in which the fishes were subjected to Available Commercial Feed (ACF) containing 20% of crude protein (Fig. 1). The other feed constituents were the same as that of the commercial feed and the main focus was to assess the feed preference of *Tor tor* in response to change in protein concentration.

Table 2 depicts the results for the growth of *Tor tor*. Out of the four treatments, T_3 showed the maximum increase in length (mm) (86.3±2.2) after the stipulated research tenure of 12 months (365 days). The lowest increase was shown by T_4 (ACF) with the maximum length (mm) of 47.2±1.6. The mean±SE for the treatment groups was calculated as 8.00±1.20. ANOVA (Table 3) for total length (df = 35) attainment among treatment groups showed no significant relation ($p>0.05$) among the treatment groups. More or less similar trend was observed in percent length among the treatment groups, calculated mathematically. In case of final weight, the highest weight (g) was achieved by fishes subjected to T_3 (5.329±1.11) in comparison to the ACF group (T_4) which showed a final weight of 1.928±0.26. The mean±SEM for the treatment set was 0.699±0.03. No significant relation ($p>0.05$) was recorded among the treatments groups (df = 35). Similar trend was observed for per day length and weight, corresponding to the attainment of final length and weight at the end of the experiment. ANOVA showed significant ($p<0.05$) relations among the treatment groups (df = 31) in case of per day length (Table 3). However, no significant differences ($p>0.05$) were observed in case of per day weight (df = 31).

The Specific Growth Rate (SGR) analysis revealed the highest SGR value 0.328±0.03 in case of treatment 3, as compared to fishes subjected to ACF. The mean±SEM for the treatment set was 0.025±0.01. ANOVA in case of SGR revealed no significant differences ($p>0.05$) among the treatment

Table 3: ANOVA of different growth parameters of *Tor tor* fed with varying concentrations of protein

Parameters	df	SS	MS	F	F crit.	p-values	Variance
Length (mm)	35	11002.59000	5.7.7455 (WG) 296.2297 (BG)	1.714	2.901	0.183	T_1 = 257.7 T_2 = 296.0 T_3 = 535.6 T_4 = 95.50
Weight (g)	35	46.27428	2.143317 (WG) 1.245135 (BG)	1.721	2.901	0.182	T_1 = 0.917 T_2 = 1.213 T_3 = 2.583 T_4 = 0.265
Per day length (mm)	31	0.236495	0.01838 (WG) 0.00647 (BG)	2.837	2.946	0.05	T_1 = 0.004 T_2 = 0.004 T_3 = 0.014 T_4 = 0.002
Per day weight (g)	31	0.001509	0.0000074 (WG) 0.0000046 (BG)	1.614	2.946	0.208	T_1 = 0.000 T_2 = 0.000 T_3 = 0.000 T_4 = 0.000
SGR	31	1.047468	0.103155 (WG) 0.026357 (BG)	3.913	2.946	0.018	T_1 = 0.066 T_2 = 0.016 T_3 = 0.014 T_4 = 0.007
FCR	31	0.317066	0.092845 (WG) 0.001376 (BG)	67.4	0.000	2.94	T_1 = 0.001 T_2 = 0.000 T_3 = 0.001 T_4 = 0.001
PER	31	8.028233	0.638925 (WG) 0.218266 (BG)	2.92	0.05	2.94	T_1 = 0.134 T_2 = 0.354 T_3 = 0.384 T_4 = 0.000

SGR = Specific Growth Rate; FCR = Food Conversion Rate; PER = Protein Energy Ratio

groups (df = 31). The lowest FCR (0.82±0.02) was observed in T_3 (40% CP), as compared to other two high protein treatments. The highest FCR (1.8±0.03) was recorded for T_4 which received feed containing 20% CP. The mean±SEM for the treatment set was 0.056±0.01. ANOVA for FCR revealed significant differences ($p<0.05$) among the treatment groups (df = 31). In case of PER, the highest value (0.846±0.02) was recorded in T_3 (40% CP), as compared to ACF with PER as low as 0.083±0.02. The mean±SE for the treatment set was 0.159±0.01. ANOVA for SGR among treatments revealed significant difference ($p<0.05$) among treatment groups (df = 31). The variance among the treatment groups is illustrated in Table 3.

DISCUSSION

The present research work was an effort put with an intention to upgrade the aquaculture nutrition science with respect to commercially important fishes, such as *Tor tor* (Hamilton, 1822) by using different protein ratios for determination of a feed formulation which would reduce the FCR value and makes the fish growth economical. Although, there has been so many attempts to formulate the diets as per the requirement of the fishes, present research is no exception to that except it encompasses a featured research on all the aspects and feed formulation, feeding and its impact on various physio-biochemical aspects of the test species. The research can be used as an engender for the future researchers and will be a commendable contribution to aquaculture nutrition.

A feeding trial was conducted to investigate the effects of dietary protein levels on growth, feed utilization, hepatosomatic index and liver lipid deposition of juvenile red snapper, Lutjanus argentimaculatus by Abbas *et al.* (2005). In their experiment, six fishmeal-based diets were formulated to contain various protein levels (20-45% in 5% increments). The fish at the end of the study had more than 10-fold (77.0 g) increase in weight compared to the initial (8.0 g). Fish fed diets of 40 and 45% protein produced significantly ($p<0.05$) higher weight gain of 77.2 and 76.5 g and Specific Growth Rate (SGR) of 2.65 and 2.62%. The studies of the above researchers correspond to the results which showed increase in weight (5.329±1.11) and SGR (0.328±0.03) in *Tor tor* fed on 40% CP diets.

Effects of the protein/carbohydrate ratio of extruded diets on protein synthesis, protein growth and body composition in juvenile brown trout (*Salmo trutta*) was studied by Viaplana-Marin *et al.* (2006). The researchers reported differences in specific growth rates (C: 0.88%; HC: 0.77%) associated more with the lower protein consumption rate and the lower level of digestible energy in fish fed HC than with the higher dietary carbohydrate content of the diet, documenting the impact of high protein in overall increase in SGR and PER. Tabassum and Mukhtar (2009) worked on the effects of dietary protein levels on growth, feed utilization, protein retention efficiency and body composition of young *Heteropneustes fossilis* (Bloch). The results of the research get complete support from the work of the present researchers.

An 8 weeks growth study was conducted to determine the effect of ration level, energy and protein maintenance requirement of catfish, *Heteropneustes fossilis* Bloch, fingerling (7.90±0.55 cm; 3.10±0.28 g) by feeding casein-gelatin-based purified diet (40% CP; 3.61 kcal g^{-1} GE) at six ration levels 1-6% of BW/day by Ahmed (2010). The researchers concluded that maximum live weight gain, best Feed Conversion Ratio (FCR), best Specific Growth Rate (SGR) and highest Protein Efficiency Ratio (PER) were evident for ration levels of 45% body weight (40-45% CP). This is in complete agreement with our findings. In 2011 Chaitanawisuti *et al*. (2011), worked on the effects of dietary protein and lipid levels and protein to energy ratios on growth performance and feed utilization of hatchery-reared juvenile spotted babylon (*Babylonia areolata*). A 120 day feeding trial was designed by researchers to determine the effects of different dietary protein and lipid levels and protein to energy ratio (P:E). Six diets were formulated to contain three protein levels (18, 28 and 36%). The results showed that the highest ($p<0.05$) values for growth and feed efficiency were observed for snails fed a diet containing 36% protein level and the same trend was observed for snails fed a diet with 10% lipid level. The higher protein levels substantiated by the appropriate energy contents increases the growth coefficient, as evidenced in the present study and documented by the mention earlier researchers.

Zehra and Khan (2012) enumerated the dietary protein requirement for fingerlings of *Channa punctatus* (Bloch), based on growth, feed conversion, protein retention and biochemical composition. The researchers reported that the maximum Absolute Weight Gain (AWG; 8.11 g/fish), Specific Growth Rate (SGR; 1.82%) and best Feed Conversion Ratio (FCR; 1.48) were recorded in fish fed diet containing 450 g kg^{-1} protein whereas Protein Efficiency Ratio (PER; 1.52), Protein Retention Efficiency (PRE; 25%), Energy Retention Efficiency (ERE; 78%) and RNA/DNA ratio (3.01) were maximum for the group fed dietary protein at 400 g kg^{-1}. The research of the mentioned earlier, researchers lends complete support to the findings, stating that with the appropriate incorporation of energy source, protein proportion could be utilized as the best convertible food substance by the mahaseer *Tor tor*.

CONCLUSION

The present study revealed that the inclusion of higher levels of protein (CP = 40%) in Mahaseer diets, containing appropriate P:E ratio is feasible for the better growth and food conversion efficiency of the given feed. As such research, in the field of Mahaseer nutrition needs some more information to make the dietary needs of *Tor tor* more understandable.

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