A STUDY ON PROTEIN SPARING EFFECT OF DIETARY LIPIDS IN *LABEO ROHITA* (PISCES: CYPRINIDAE)



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Abstract: The present studies were conducted to study the effect of dietary protein and lipid levels on the growth of carp *Labeo rohita* (Hamilton). Four isoenergetic diets were prepared by using rice bran oiled, rice bran deoiled, mustard cake oiled, mustard cake deoiled and fish meal, having increasing level of lipids and decreasing level of proteins, i.e., D1 ($P_{30}L_3$), D2 ($P_{25}L_6$), D3 ($P_{22}L_8$) and D4 ($P_{17}L_{10}$) and feeding was done @ 2% fish biomass. Water quality parameters viz., temperature, pH, dissolved oxygen, phenolphthalein alkalinity, methyl orange alkalinity, total alkalinity, ammonia, nitrite, nitrate, and soluble phosphate, remained within the optimal ranges suggested for carps. The overall growth (both in terms of length and weight) of *L. rohita* fed on diet D3 ($P_{22}L_8$) was found to be maximum. The growth of the fish species improved with an increase of dietary lipid level (from 3% to 8%) and a simultaneous decrease in protein level (from 30% to 22%). Feed conversion ratio was found to be better in D3. The protein efficiency ratio as well as protein utilization increased with a decrease in dietary protein and increase in dietary lipids. A positive correlation between increase in dietary lipid content and protein utilization was recorded which suggested that the dietary proteins are spared for growth by inclusion of lipids in fish diet. Maximum yield and maximum net profit was obtained in fish fed with diet D3. Hence, it can be concluded that the dietary protein so to 22% by an increase in dietary lipids from 3 to 8% as the inclusion of dietary lipids spare proteins for growth.

Key words: Fish, Fish nutrition, Dietary lipids, Dietary protein, Protein sparing effect, Protein Utilization, Carps.

INTRODUCTION

Development of cost-effective nutritionally balanced diets for fish is the main factor affecting the intensive aquaculture due to its influence on growth, health and production costs. Nutrients and energy are needed in the diet for growth and maintenance. Since protein sources are among the most expensive feed ingredients, it is economically desirable to minimize the protein level in the diet whilst keeping it high enough to sustain good growth. Excess protein can be replaced by an energy-rich compound such as lipid.

Fish, like other animals, eat to satisfy their energy requirements (Lee and Putnam, 1973). If the protein content of the diet is too high, the excess is catabolized to provide energy for growth, lowering the protein conversion efficiency (Lee and Putnam, 1973; Adron *et al.*, 1976). However when fish are fed on excess of energy, feed consumption decreases and growth is reduced (Ringrose, 1971; Lovell, 1998). Besides the high

production costs associated with inadequate feed rations, deterioration of environmental waters due to wasted feed and excretion through fish gills and kidneys may also be a result of inadequate feeds.

It has been suggested that nitrogen excretion from fish can be reduced by increasing dietary protein retention. This can be accomplished by adjusting the total energy (protein:energy ratio) in the diet to an optimal balance (Watanabe *et al.*, 1987). Consequently the objective of the present study was to evaluate the effect of high lipid contents of the diet on growth performance, protein utilization and sparing in the carp *Labeo rohita*.

MATERIALS AND METHODS

Cemented tanks each with an area of 0.008 ha (80 m³, 1.2 m depth). Tanks were manured with mixture of cow dung and poultry dropping (1:3) @ 10,000 Kg/ha/yr (80 Kg/tank/yr). One tenth

(8 Kg/tank) of the manure was applied 15 days prior to the stocking of fish and rest in equal installments at weekly intervals (1.44 Kg/week/tank). Each tank was stocked @10,000 fingerlings/ha. Four different diets were prepared by using rice bran oiled, rice bran deoiled, mustard cake oiled, mustard cake deoiled and fish meal. On the basis of the proximate composition of ingredients, done by AOAC method (2000), the percentage composition of ingredients was adjusted to prepare four isocaloric diets having increasing lipid levels and decreasing protein levels, i.e. D1 ($P_{30}L_3$), D2 ($P_{25}L_6$), D3 ($P_{22}L_8$) and D4 ($P_{17}L_1$).

Feeding was done @ 2% fish biomass once a day in the morning hours between 9.00 and 10.00 A.M. Growth was assessed at monthly intervals in terms of total body length and body weight. From the above recorded data, percent gain in total body length (TLG),percent gain in total body weight (TWG), net weight gain (NWG), specific growth rate (SGR), protein efficiency ratio (PER), feed conversion ratios (FCR) and Apparent Net Protein Utilization (ANPU) were estimated. All analysis were made in triplicates. Muscle tissues were sampled between the lateral line and the dorsal fin and flesh quality in terms of protein content (Lowry *et al.*, 1951) was estimated.

RESULTS AND DISCUSSION

Growth performance of different fishes in terms of length and weight gain is shown in the figure 1 and 2. Among different treatments net weight gain and specific growth rate was maximum in D₃ ($P_{22}L_8$) and minimum in D₁($P_{30}L_3$). Feed conversion ratio was better in D₃ and poor in D₂ ($P_{25}L_6$). Protein efficiency ratio was maximum in D₄ ($P_{17}L_{10}$) and minimum in D₁. The flesh protein content was lowest in fishes fed on diet D₄ ($P_{17}L_{10}$). As the level of protein the diet decreased along with simultaneous increase in lipids, the flesh protein content showed an increasing trend. Apparent Net Protein Utilization was maximum in D₄ and Minimum in D₁. No mortality was observed in any treatment.

In the present investigations, the growth of fish improved with an increase in dietary lipid level (from 3% to 8% lipid) and a simultaneous decrease in protein level (from 30% to22% protein). However, it declined when fed on diet containing higher level of lipid (10%) and lower level of protein (17%). Hence, a level of 8% lipids could be considered optimum for *L. rohita* when the dietary protein content is around 22% and the gross energy of diet is 3.41 kcal/g the diet (D3) having a protein/energy ratio of 1: 0.15 gave the best results in terms of gain in body

Ingradiants	Diets					
Ingredients	Dı	D2	D3	D4		
Rice bran (oiled)		10	50	60		
Rice bran (deoiled)	40	40		10		
Mustard cake (oiled)		30	30	30		
Mustard cake (deoiled)	40					
Fish meal	20	20	20			

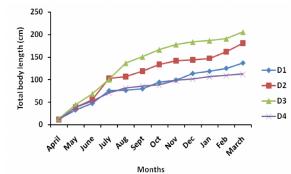
Table 1. Per cent composition of feed ingredients

Ingredients	Crude Protein	Ether Extract	Crude Fiber	Ash	Nitrogen Free Extract
Rice Bran (oiled)	7.22	12.32	18.34	7.61	39.32
Rice Bran (deoiled)	16.32	3.67	16.60	14.78	38.63
Mustard Cake (oiled)	34.50	7.53	7.63	4.10	33.86
Mustard Cake (deoiled)	40.25	1.15	8.34	6.32	36.24
Fish Meal	33.25	7.06	5.30	34.30	12.87

 Table 2. Proximate composition of different feed ingredients

Diets	Crude Protein	Ether Extract	Crude Fiber	Ash	Nitrogen Free Extract	Gross Energy (Kcal/g)	P:E (g/Kcal)
Dı	29.7	3.34	12.56	16.35	29.53	3.20	1:0.11
D2	25.4	6.33	12.01	15.32	31.09	3.29	1:0.13
D3	21.9	8.36	15.37	13.60	33.91	3.41	1:0.15
D4	17.5	9.82	17.17	6.56	36.6	3.42	1:0.19

Table 3. Proximate composition of different prepared supplementary diets



250 Total body weight (g) 200 150 D1 100 -D2 50 **D**3 0 -D4 March May June July Aug Sept oct Nov Dec Jan Feb April Months

Fig. 1. Effect of supplementary diets on total body length (cm) in *Labeo rohita* during the culture period

weight as well as body length. Gangadhar *et al* (1997) reported that the growth of fingerlings of *L. rohita* fed with diet containing 25% protein and 9% fat was comparable with the diet containing 30% protein and 6% fat, while Manjappa *et al.* (2002) reported a diet containing 24% protein and 6% lipid to be optimum for the growth of *Cyprinus carpio*.

The level of protein in the diet which supports maximum growth depends on the energy level in the diet (Lee and Putnam, 1973). It was observed that the growth of the fish increased

Fig. 2. Effect of supplementary diets on total body weight (g) in *Labeo rohita* during the culture period

relative to the increase in the protein/energy ratio from 1:0.11 to 1:0.15. This showed that the level of protein needed for maximum growth of these fish can be decreased by increasing the dietary lipid (non-protein energy) level. Page and Andrew (1973) suggested that fish eat to satisfy their energy requirement and hence the level of protein needed for the maximum growth in channel catfish (*Ictalurus punctatus*) can be decreased by increasing the dietary energy level. Reinitz *et al* (1978), while working on rainbow trout (*Oncorhynchus mykiss*), also found that in a diet with high energy content in relation to

	D1	D2	D3	D4
TW G (%)	1041.67	1487.72	1504.69	901.78
NWG	125.00	169.90	192.60	101.00
SG R	0.29	0.33	0.33	0.27
FCR	3.06	3.00	2.90	3.66
PER	1.10	1.32	1.43	1.57
Flesh protein	12.97 ^a	12.81 ^a	11.46 ^b	9.59 [°]
content	± 0.16	± 0.22	± 0.05	± 0.13
ANPU	40.43	46.93	48.54	50.69

Table 4. Comparison of growth and other parameters with reference to different diets

the percentage of dietary protein (high digestible energy/protein ratio), less protein is wasted as energy source.

Present observations revealed that an increase in dietary lipid content from 3% to 8% can reduce the requirement of dietary protein from 30% to 22%. Lee and Putnam (1973) reported that high energy diets containing 24% herring oil promoted excellent growth in rainbow trout (Oncorhynchus mykiss). At this level of lipid, dietary protein was reduced to one-third with no loss in weight gain, while feed conversion, energy conversion and protein utilization markedly increased. Yigit et al. (2002) also reported that the protein content of practical trout feeds can be reduced from currently used 47% to around 44% without reduction in the growth rate and feed efficiency, if high quality protein is used and the gross energy is increased to about 20.34 kJ/g diet.

In the present study, increase in the fish growth with increase in dietary lipid level and simultaneous decrease in protein level of diets, may be attributed to protein sparing effect of lipid which has been demonstrated by many scientists in teleost fishes (De Silva et al., 1991, Gangadhar et al. 1997, Weatherup et al., 1997). Morais et al. (2001) demonstrated protein sparing effect of lipids in Atlantic cod (Gadus morhua L.) fed with four extruded diets differing in the levels of proteins (48% or 58%) and lipids (12% and 16%). The tested diets induced good performance results, with a protein sparing effect by lipid and a more efficient use of protein in diets with lower protein content. The $P_{48}L_{16}$ diet was reported to be the best compromise between growth, feed utilization and cost.

Satpathy *et al.* (2003) also reported that *L. rohita* could utilize lipid (50 to 150 g/kg) to spare protein but there is no significant advantage from this beyond the dietary protein level of 350-400 g/ kg in terms of growth and body composition. Meyer and Fracalossi (2004) demonstrated that jundia (*Rhamdia quelen*) presents dietary protein sparing effect when the energy concentration increases from 3200 to 3650 kcal/kg in an experiment where five protein concentrations [26%, 29%, 33%, 37%, and 41% crude protein

(CP)] were fed to jundia fingerlings, at two dietary energy concentrations [3200 and 3650 kcal metabolizable energy (ME)/kg diet]. A significant interaction between dietary protein and energy was observed for weight gain, SGR and PER.

In the present case, the inferior growth of fish fed on diet D₄ ($P_{r_7}L_{10}$) than that fed on diet D₃ ($P_{22}L_8$) reflected a negative impact of dietary lipid beyond the optimum level. This could be due to imbalance in the protein: lipid ratio and inability of fish to utilize lipid above a certain threshold level (De Silva *et al.*, 1991). Drastic decrease in the growth of fish fed on diet D₄ may also be attributed to lack of essential amino acids as it lacked the dietary ingredient fish meal.

PER increased with the increase in dietary lipid and decrease in dietary protein content. This shows that protein efficiency ratio is inversely proportional to dietary protein content as reported by Khan and Jafri (1990) in catfish (*Clarias batrachus*). Similar to the results obtained in the present study, Chan *et al.* (2002) reported that diets containing higher lipid levels exhibit improved protein efficiency ratio.

In the present study, the protein content of fish flesh decreased with a decrease in the dietary protein content. A drastic decrease in flesh protein content was observed in fishes fed on diet D4, having minimum protein (17%) and maximum lipids (10%). A decrease in flesh protein content with decrease in dietary protein content has also been reported by many scientists. Mohanty and Samantaray (1996) also reported a significant increase in carcass protein and a significant decrease in ash content with progressive dietary protein substitution in Channa striata. Al-Hafed (1999) reported that the body composition of Nile tilapia, Oreochromis niloticus, is influenced by dietary protein. Percentage body protein of the fish fed 40-45% protein was higher than that of fish fed 25-35% protein diets, whereas lipid content decreased with increasing dietary protein level. However, in the present case, the difference in the flesh protein content was insignificant in fishes fed on diet D1, D2 and D3. This shows that protein is retained in the fish flesh even when the fishes were fed with a diet having decreasing protein level thus depicting the protein sparing effect of lipids.

However, this protein sparing effect of lipids was found to be effective only till 8% dietary lipid. A drastic decrease in fish flesh protein content observed in fishes fed on diet D4 ($P_{17}L_{10}$) shows that when the protein decreased beyond 22%, the increase in lipids content was unable to compensate for the decrease in protein level.

In the present study, ANPU improved as the level of dietary lipid increased and dietary protein decreased. A positive correlation between increase in dietary lipid content and protein utilization was recorded which suggests that the dietary proteins are spared for growth by inclusion of lipids in the fish diet. Hassan et al. (1995) and Skalli et al. (2004) have also observed maximum utilization of proteins in Cirrhinus mrigala and common dentex, respectively, when fed with diets having same energy level but low protein level. They suggested that protein utilization can be enhanced by sparing effect of dietary lipids. De Silve et al. (2002) have also reported better utilization of protein on increasing the lipid content and decreasing the protein content of diet in juvenile murray cod, Maccullochella peelii peelii. The highest ANPU was observed in fish reared on diet P40L24 and the lowest in fish fed with diet $P_{50}L_{24}$.

From the present investigations, it can be concluded that the diet D₃ ($P_{22}L_8$) is the best in terms of growth as well as economics for *L. rohita* and the dietary protein of the carp can be decreased from 30 to 22% by an increase in dietary lipids from 3 to 8% as the inclusion of dietary lipids spare proteins for growth.

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