

Effect of protein restriction with subsequent realimentation on performance and carcass composition of Indian major carp (*Labeo rohita* H.)

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ABSTRACT

The aim of the present study was to determine the protein restriction followed by the realimentation suitability for culture of rohu. Acclimatized rohu fingerlings (150), 20.74 ± 0.13 g/f, were distributed into 5 experimental groups (30 fingerlings in each group with three replications of 10 fingerlings each) and reared in cleaned, disinfected and electrically operated aerators fitted plastic tubs of 50 liters capacity, using completely randomized design. Our results indicated that fish fed alternatively low protein and high protein diet at 3 or 7 days interval showed compensatory growth with the mechanism of hyperphagia, better feed conversion ratio (FCR), specific growth rate (SGR), absolute growth rate (AGR), protein efficiency ratio (PER) and performance index (PI) but fish fed alternatively low protein and high protein diet at 15 or 25 days interval was unable to growth compensation during 90 days trial period. Chemical composition of fish muscle indicated that the higher growth efficiency during compensatory growth was probably due to protein growth rather than fat deposition in gut. Thus, it is suggested that for profitable carp culture, farmers can use mixed feeding schedule of alternative feeding of low protein and high protein as a means of reducing feed cost.

Key words: Compensatory growth, Protein restriction, Muscle composition, Re-alimentation, Rohu (*Labeo rohita*)

The applicability of mixed feeding schedules with a high-protein diet was alternated with a low-protein diet in reducing feed costs and improving nutrient utilization has been demonstrated in Indian carp, *Catla catla* (Nandeesha *et al.* 1994), common carp, *Cyprinus carpio* (Nandeesha *et al.* 2002), Nile tilapia, *Oreochromis niloticus* (Patel and Yakupitiyage, 2003), sutchi catfish, *Pangasius hypophthalmus*; and silver carp, *Hypophthalmichthys molitrix* (Ali *et al.* 2005), rainbow trout, *Oncorhynchus mykiss* (Sevgili *et al.* 2006), hybrid striped bass, *Morone chrysops* $\times$ *Morone saxatilis* (Turano *et al.* 2007). Ali *et al.* (2005) suggested that feeding fish continuously with high protein diet is not economical. Therefore, present experiment was conducted to observe the influence of a limited crude protein supply followed by an adequate supply as compared to continuous feeding on performance and carcass

composition of Indian major carp, rohu (*Labeo rohita* H.).

MATERIALS AND METHODS

Acclimatized rohu fingerlings (150), $(20.74 \pm 0.13$ g/f), were distributed into 5 experimental groups (30 fingerlings in each group with 3 replications of 10 fingerlings each) and reared in cleaned, disinfected and electrically operated aerators fitted plastic tubes of 50 liters capacity, using completely randomized design. Feeding was done using 1 mm diameter pellet (D₁) with 30% crude protein (Table 1, NRC 1993) or 20% crude protein (D₂, Table 2) over 90 days trial period at 3% of body weight. Control group (T₀) was fed diet having 30% crude protein. Experimental groups T₁, T₂, T₃ and T₄ were fed on 20% and 30% CP diets alternatively for 3, 7, 15 25 days respectively. Daily rations were divided into 2 equal meals and fed at 9.00 and 16.00 h. After 1h of feeding, the left over feed was collected by siphoning to determine the weight of residues used to calculate actual feed consumption on dry matter basis after oven drying ($100 \pm 2^\circ\text{C}$ for 12 h) (AOAC 1995). Proximate principles (dry matter DM%; crude protein CP%; crude fibre CF%; ether extract EE%; total ash TA%; organic matter OM%; and nitrogen free extract NFE%) in feed were analyzed as per AOAC (1995). Calcium (Ca) of feed was determined by AAS method (AOAC 1995) and the

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Table 1. Ingredients and nutrient composition of experimental diet D₁

Ingredient composition	Quantity (kg/100 kg)
Maize	28
Rice bran	5
Soyabean meal	50
Fish meal	10
Wheat gluten meal	2
Wheat flour	2
Vegetable oil	1
DCP	0.5
Iodized salt	0.924
Trace mineral premix ^a	0.028
Vitamin premix ^b	0.423
Lysine	0.025
Butylated hydroxyl anisole (BHA)	0.05
Methionine	0.05
Total	100.00
Nutrient composition	
Moisture (% estimated)	13.37
CP (% estimated)	30.75
EE (% estimated)	4.50
TA (% estimated)	2.51
CF (% estimated)	5.78
NFE (% estimated)	43.09
ME (KCal/kg, calculated)	2562.70
Ca (% estimated)	0.28
Inorganic P (% estimated)	1.29
Available P (% calculated)	0.77
Lysine (% calculated)	1.56
Methionine (% calculated)	0.83
P/E ratio (mg/KCal, calculated)	119.99

^aTrace mineral premix supplied the following per kg of diet: Cu (CuSO₄, 5H₂O, 25% Cu), 2.70 mg; Fe (FeSO₄, 7H₂O, 20.09% Fe) 27.03 mg; Mn (MnSO₄, H₂O, 36.01% Mn), 11.71 mg; Zn (ZnSO₄, 40.17% Zn), 27.03 mg; I (KI, 76.01% I), 5 mg; Se (Na₂SeO₃, 53.02% Se), 0.27 mg.

^bVitamin premix supplied the following per kg of diet: Vitamin A, 20,000 IU; Vitamin D₃, 3,000 IU; Vitamin E, 16 mg; Vitamin K, 2 mg; Vitamin B₁, 1.6 mg; Vitamin B₂, 10mg; Vitamin B₆, 3.2 mg; Vitamin B₁₂, 0.041 mg; Niacin, 24 mg; Ca Pantothenate, 16 mg; Folic acid, 1.6 mg; Biotin, 1 mg; Ascorbic acid, 100 mg; Choline, 1,500 mg.

phosphorus (P) of feed was estimated by spectrophotometric method (AOAC 1975). Body weight of fish was recorded at

Table 2. Ingredients and nutrient compositions of experimental diet D₂

Ingredient composition	Quantity (kg/100 kg)
Maize	48.5
Rice bran	8
Soybean meal	35
Wheat gluten meal	2
Wheat flour	3.5
Vegetable oil	1
DCP	1
Iodized salt	0.424
Trace mineral premix ^a	0.028
Vitamin premix ^b	0.423
Lysine	0.025
Butylated hydroxy anisole (BHA)	0.05
Methionine	0.05
Total	100.00
Nutrient composition	
Moisture (% estimated)	14.76
CP (% estimated)	20.34
EE (% estimated)	5.36
TA (% estimated)	0.84
CF (% estimated)	5.92
NFE (% estimated)	52.78
ME (KCal/kg, calculated)	2531.50
Ca (% estimated)	0.36
Inorganic P (% estimated)	0.57
Available P (% calculated)	0.34
Lysine (% calculated)	1.15
Methionine (% calculated)	0.17
P/E ratio (mg/KCal, calculated)	80.35

^{a,b}See Table 1.

the end of protein restriction and re-alimentation period and live weight change were also calculated. Feed conversion ratio (FCR), feed efficiency ratio (FER) or gross growth efficiency (GGE), protein efficiency ratio (PER), performance index (PI), specific growth rate (SGR), absolute growth rate (AGR), survival rate (SR) and mortality (%) were calculated as per Castell and Tiews (1980).

Daily water exchange was done and water quality parameters were estimated as per standard methods (APHA

Table 3. Water quality parameters during 90 days feeding trial

Treatment	Temperature °C	pH	D O (ppm)	Total alkalinity (ppm)
T ₀	25.30 ± 0.53	7.02 ± 0.08	6.96 ± 0.15	129.12 ± 14.58
T ₁	25.27 ± 0.20	7.11 ± 0.11	7.65 ± 0.40	131.91 ± 11.24
T ₂	25.47 ± 0.41	7.26 ± 0.21	7.07 ± 0.15	135.30 ± 13.40
T ₃	25.46 ± 0.32	7.17 ± 0.22	7.28 ± 0.21	141.31 ± 11.21
T ₄	25.72 ± 0.29	7.55 ± 0.30	7.58 ± 0.51	136.23 ± 14.33

P>0.05

1989). Initially 10 fish and 5 fish from each replicate of each group were collected after 45 days and 90 days of trial period and chemical composition like DM%, OM%, CP%, EE%, TA% and NFE% of wet muscle were estimated as per AOAC (1995). The data were analyzed statistically (Snedecor and Cochran 1994) using Compare Means (SPSS 1997).

RESULTS AND DISCUSSION

Throughout the experiment water quality parameters (Table 3) of all experimental groups were within the optimum level for fish production (Banerjee 1976), which indicated that water quality did not produce any stress to the fish during the trial period. Fish mortality pattern (Table 4) during 0-45

Table 4. Performance parameters of rohu fingerlings reared on different feeding schedules.

Treatment/Parameter	T ₀	T ₁	T ₂	T ₃	T ₄	P
Initial body wt (g/f)	20.63±0.09	20.75±0.07	20.66±0.11	20.78±0.14	20.82±0.04	> 0.05
Initial body length (cm/f)	12.38±0.05	12.45±0.04	12.39±0.06	12.47±0.08	12.49±0.03	> 0.05
Final body wt (g/f)						
0-45 d	67.39 ^a ±5.77	54.76 ^{ab} ±6.87	47.57 ^{bc} ±6.31	41.78 ^{bc} ±5.25	32.82 ^c ±3.35	< 0.05
45-90 d	128.68 ^a ±11.53	125.55 ^a ±10.90	130.59 ^a ±14.38	85.50 ^b ±11.52	65.79 ^b ±8.76	< 0.05
Average wt gain (g/f)						
0-45 d	46.76 ^a ±5.68	34.01 ^{ab} ±6.82	26.91 ^{bc} ±6.40	21.00 ^{bc} ±5.38	11.99 ^c ±3.31	< 0.05
45-90 d	61.29 ^{bc} ±5.76	70.79 ^{ab} ±4.01	83.02 ^a ±8.06	43.71 ^{cd} ±6.27	32.97 ^d ±5.41	< 0.05
0-90 d	108.05 ^a ±11.44	104.80 ^a ±10.84	109.93 ^a ±14.47	64.71 ^b ±11.64	44.96 ^b ±9.72	< 0.05
Average% wt gain						
0-45 d	226.45 ^a ±26.61	163.74 ^{ab} ±32.34	130.56 ^{bc} ±31.48	101.40 ^{bc} ±26.57	57.55 ^c ±15.78	< 0.05
45-90 d	90.81 ^c ±0.78	131.61 ^b ±9.41	176.29 ^a ±6.81	104.09 ^c ±2.31	99.14 ^c ±6.42	< 0.05
0-90 d	523.30 ^a ±53.27	504.79 ^a ±50.73	532.82 ^a ±72.13	312.10 ^b ±58.07	215.76 ^b ±41.45	< 0.05
Final b. length (cm/f)						
0-45 d	19.26 ^a ±1.65	15.65 ^{ab} ±1.96	13.59 ^{bc} ±1.80	11.94 ^{bc} ±1.50	9.38 ^c ±0.96	< 0.05
45-90 d	21.45 ^a ±1.92	20.93 ^a ±1.82	21.77 ^a ±2.40	14.25 ^b ±1.92	10.96 ^b ±1.46	< 0.05
DMI (g/f)						
0-45 d	97.97±16.55	96.84±20.03	100.88±23.58	95.55±19.39	79.05±13.48	> 0.05
45-90 d	195.79±33.61	236.55±37.01	296.01±59.34	213.59±43.20	168.12±32.57	> 0.05
0-90 d	293.76±50.14	333.39±56.79	396.89±82.91	309.13±62.59	247.16±46.05	> 0.05
DMI (% of b. wt)						
0-45 d	3.18 ^c ±0.27	3.85 ^{bc} ±0.32	4.58 ^{ab} ±0.49	4.99 ^{ab} ±0.39	5.28 ^a ±0.37	< 0.05
45-90 d	3.33 ^c ±0.29	4.14 ^{bc} ±0.30	4.94 ^{ab} ±0.46	5.46 ^a ±0.38	5.59 ^a ±0.35	< 0.05
FCR						
0-45 d	2.07 ^d ±0.10	2.85 ^{cd} ±0.06	3.77 ^{bc} ±0.10	4.73 ^b ±0.41	7.13 ^a ±1.06	< 0.05
45-90 d	3.15 ^b ±0.26	3.31 ^b ±0.33	3.49 ^b ±0.37	4.81 ^a ±0.30	5.06 ^a ±0.18	< 0.05
0-90 d	2.68 ^d ±0.19	3.14 ^{cd} ±0.22	3.54 ^c ±0.28	4.75 ^b ±0.17	5.53 ^a ±0.20	< 0.05
FER						
0-45 d	0.48 ^a ±0.02	0.35 ^b ±0.01	0.27 ^c ±0.01	0.21 ^c ±0.01	0.15 ^d ±0.02	< 0.05
Or GGE						
45-90 d	0.32 ^a ±0.02	0.31 ^a ±0.03	0.29 ^a ±0.03	0.21 ^b ±0.01	0.19 ^b ±0.01	< 0.05
0-90 d	0.38 ^a ±0.03	0.32 ^{ab} ±0.02	0.29 ^b ±0.02	0.21 ^c ±0.01	0.18 ^c ±0.01	< 0.05
PI						
0-45 d	22.43 ^a ±1.65	11.96 ^b ±2.35	7.19 ^{bc} ±1.75	4.66 ^c ±1.42	1.87 ^c ±0.68	< 0.05
45-90 d	19.45 ^b ±0.37	21.60 ^{ab} ±1.09	23.80 ^a ±0.21	9.02 ^c ±0.88	6.48 ^d ±0.92	< 0.05
0-90 d	40.11 ^a ±1.55	33.27 ^b ±1.94	30.83 ^b ±1.69	13.58 ^c ±2.24	1.20 ^c ±1.68	< 0.05
SGR (%/d/f)						
0-45 d	2.62 ^a ±0.18	2.12 ^{ab} ±0.28	1.81 ^{abc} ±0.31	1.52 ^{bc} ±0.29	0.99 ^c ±0.23	< 0.05
45-90 d	1.43 ^c ±0.01	1.86 ^b ±0.09	2.26 ^a ±0.05	1.58 ^c ±0.02	1.53 ^c ±0.07	< 0.05
0-90 d	2.02 ^a ±0.09	1.99 ^a ±0.09	2.03 ^a ±0.13	1.55 ^b ±0.16	1.26 ^b ±0.15	< 0.05
0-45 d	1.04 ^a ±0.13	0.76 ^{ab} ±0.15	0.60 ^{bc} ±0.14	0.47 ^{bc} ±0.12	0.27 ^c ±0.07	< 0.05
AGR (g/d/f)						
45-90 d	1.36 ^{bc} ±0.12	1.57 ^{ab} ±0.08	1.84 ^a ±0.18	0.97 ^{cd} ±0.14	0.73 ^d ±0.12	< 0.05
0-90 d	1.20 ^a ±0.12	1.16 ^a ±0.12	1.22 ^a ±0.16	0.72 ^b ±0.13	0.50 ^b ±0.09	< 0.05
0-45 d	1.58 ^a ±0.07	1.40 ^a ±0.03	1.05 ^b ±0.03	0.90 ^b ±0.07	0.58 ^c ±0.07	< 0.05
PER						
45-90 d	1.05 ^{ab} ±0.09	1.19 ^a ±0.11	1.16 ^a ±0.12	0.77 ^b ±0.04	0.79 ^b ±0.03	< 0.05
0-90 d	1.23 ^a ±0.08	1.25 ^a ±0.08	1.14 ^a ±0.08	0.81 ^b ±0.03	0.73 ^b ±0.03	< 0.05
SR (%)						
0-45 d	96.67±3.33	100.00±0.00	93.33±3.33	93.33±3.33	90.00±5.77	> 0.05
45-90 d	100.00±0.00	100.00±0.00	96.67±3.33	96.67±3.33	96.67±3.33	> 0.05

Means bearing different superscripts (abcd) along rows differed significantly (P<0.05), whereas with same superscript did not differ significantly (P>0.05). DMI = Dry matter intake; FCR = Feed conversion ratio; FER = Feed efficiency ratio; SGR = Specific growth rate; AGR = Absolute growth rate; PI = Performance index; PER = Protein efficiency ratio; SR = Survival rate.

days and 45–90 days period of trial did not vary significantly between any of the treatments and in each case average survival rate was more than 90%. Thus mortality could be due to handling stress.

The final body weight (g/f), specific growth rate (%/d/f) and absolute growth rate (g/d/f) (Table 4) of fish at the end of 90 days of trial period indicated no significant difference of final body weight between control and the experimental groups restricted and re-alimented to proteins for alternative 3 or 7 days interval, but fish subjected to restricted and re-alimented to proteins for alternative 15 or 25 days interval, showed significantly lower final body weight. The experimental groups showed growth compensation with control might be due to increased per cent live weight gain with the advancement of experiment which was also confirmed in the present experiment (Table 4). These observations indicated that during deprivation growth retardation occurred and gradually these fishes tried to compensate reduced growth showing increased rate of growth, might be owing to increased per cent feed intake in relation to body weight, increased protein efficiency ratio which ultimately reflected by better performance index as confirmed in the present experiment (Table 4).

Final length of experimental fish changed at the similar trend of final body weight change indicated compensatory growth happened due to protein growth rather than due to fat deposition in the gut. From these observation it might be said that increased feed intake (hyperphagia), improved specific growth rate, absolute growth rate, feed efficiency or gross growth efficiency, protein efficiency ratio and superior feed conversion ratio were the important mechanism through which the experimental groups which were alternatively restricted and re-alimented to protein at 3 or 7 days

during 90 days trial period could able to compensate the growth retardation to catch up the final body weight of control but the experimental groups which were restricted and re-alimented at 15 or 25 days interval were unable to compensate the growth retardation during 90 days trial period.

Similar to present observation Li and Qin (2003), Jiang *et al.* (2002) and Wang *et al.* (2000) found that specific growth rate in deprived groups of *Lates calcarifer*, red drum and tilapia respectively was greater to achieve compensatory growth. Improved growth and feed efficiency had also been reported for some other fishes showing compensatory growth (Qian *et al.* 2000). Hyperphagia might be one of the important mechanisms for compensatory growth as conformed in the present findings was also corroborated with the observation of Johansen *et al.* (2001), Zhu *et al.* (2003), Nikki *et al.* (2004), Cho *et al.* (2006), Turano *et al.* (2007).

Chemical composition of fish muscle (Table 5) at the end of the 90 days trial indicated that protein deposition in muscle was highest in T₁ and T₂ and lowest in control (T₀) and other treatments (T₃ and T₄). However, fat deposition was more in fish reared on continuously high protein diet, which suggested that the higher growth efficiency during compensatory growth was probably due to protein growth rather than fat deposition. De Silva (1985) and Nandeeshia *et al.* (2002) also reported higher protein deposition in fish grown on mixed feeding schedules.

Among the mixed feeding schedules, the highest saving in protein and cost was recorded with feeding of fish by alternative low and high protein at 3 or 7 days intervals during culture period. Thus, it is suggested that for profitable carp culture, farmers can use mixed feeding schedule of alternative feeding of low protein and high protein as a means of

Table 5. Proximate composition of muscle of rohu reared on different feeding schedules.

Treatment/Parameter		T ₀	T ₁	T ₂	T ₃	T ₄	P
Initial moisture (%)		81.71±0.53	81.54±0.22	81.36±0.37	82.38±0.46	81.27±0.89	>0.05
Initial CP (%)		11.22±0.14	11.30±0.42	11.46±0.24	10.87±0.25	11.61±0.56	>0.05
Initial EE (%)		2.68±0.11	2.72±0.15	3.22±0.23	2.66±0.28	2.85±0.17	>0.05
Initial TA (%)		1.64 ^{ab} ±0.03	1.75 ^{ab} ±0.21	1.74 ^{ab} ±0.20	1.91 ^a ±0.11	1.40 ^b ±0.11	>0.05
Moisture (%)	0–45 ^d	75.04 ^c ±0.31	75.92 ^d ±0.30	77.10 ^c ±0.28	78.25 ^b ±0.33	79.43 ^a ±0.09	<0.05
	45–90 ^d	71.35 ^c ±	72.23 ^d ±0.30	73.41 ^c ±0.28	74.56 ^b ±0.33	75.74 ^a ±0.10	<0.05
CP (%)	0–45 ^d	15.13 ^b ±0.35	16.06 ^a ±0.17	15.38 ^{ab} ±0.43	14.76 ^b ±0.04	14.59 ^b ±0.13	<0.05
	45–90 ^d	18.15 ^b ±0.35	19.08 ^a ±0.17	18.40 ^{ab} ±0.43	17.78 ^b ±0.04	17.61 ^b ±0.13	<0.05
EE (%)	0–45 ^d	4.25 ^a ±0.09	3.95 ^b ±0.04	3.80 ^b ±0.04	3.61 ^c ±0.04	3.30 ^d ±0.06	<0.05
	45–90 ^d	4.86 ^a ±0.09	4.56 ^b ±0.04	4.41 ^b ±0.04	4.22 ^c ±0.04	3.91 ^d ±0.06	<0.05
TA (%)	0–45 ^d	1.31 ^d ±0.09	1.52 ^c ±0.09	1.60 ^{bc} ±0.02	1.69 ^{ab} ±0.02	1.79 ^a ±0.04	>0.05
	45–90 ^d	1.21 ^d ±0.09	1.41 ^c ±0.09	1.49 ^{bc} ±0.02	1.58 ^{ab} ±0.02	1.68 ^a ±0.04	>0.05

Means bearing different superscripts (^{abcde}) along rows differed significantly (P<0.05), whereas with same superscript did not differ significantly (P>0.05). CP = Crude protein; EE = Ether extret; TA = Total ash.

reducing feed cost.

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