

GROWTH RESPONSE OF *CATLA CATLA* (ACTINOPTERYGII: CYPRINIFORMES: CYPRINIDAE) TO SOYA AND MAIZE SUPPLEMENTED TRADITIONAL FEED MIXTURE

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Background. In Indian carp culture, a 1 : 1 mixture of rice bran and oil cake is traditionally used as feed in powder form; it is nutritionally inadequate and also a large part of it goes unutilized as the fish cannot feed on the powder effectively. With a view to improve its nutritional quality and reduce the wastage, the mixture was supplemented with soya and maize flours and converted into pellet form. The performance of the supplemented diets was evaluated by feeding them to catla, *Catla catla*, one of the popular Indian major carps.

Materials and methods. Two test diets were formulated supplementing the feed mixture with soya flour (10% and 20%) and maize flour (39%), replacing equal quantities of rice bran and oilcake. These diets along with the control diet were fed to triplicate groups of catla fry maintained in outdoor cement tanks for 120 days.

Results. The test diets enhanced growth and net fish production significantly ($P < 0.05$). This resulted in higher economic returns under the two test treatments. Survival of fish ranged from 74% to 76%. Carcass of fish receiving the test diets had significantly ($P < 0.05$) higher lipid and lower moisture levels. Digestive enzyme activity was affected positively ($P < 0.05$) by the test diets.

Conclusion. Replacing rice bran and oilcake from the traditional feed mixture with soya and maize flours proved economically viable.

Keywords: *Catla catla*, feed mixture, growth, carcass composition, digestive enzyme activity

INTRODUCTION

Fertilization and artificial feeding are the two important management measures adopted in fish culture to enhance the production of ponds. Feed accounts for 30% to 70% of the production cost in culture fisheries, depending on the type of culture and the intensity of feeding (De Silva and Anderson 1995). Culture of carps, which contribute largely to Indian inland fish production, traditionally involves supplemental feeding with a 1 : 1 mixture of rice bran and oilcake which is employed in powder form. This feed mixture is nutritionally inadequate and also a large part of it goes unutilized as it gets to the pond bottom quickly. Improving the nutritional quality of the traditional feed mixture and providing it in pellet form would lead to better utilization of feed.

In recent years, plant proteins are being extensively used in fish and prawn feeds, mainly to replace the fish meal component, in order to reduce the feed cost.

Soybean, *Glycine max* (L.), meal is probably the most promising among the plant protein ingredients because of its high protein content and excellent amino acid profile (El-Sayed 1999), lower cost, and the ease of incorporation into feeds. Maize, *Zea mays* L., a cheap feed ingredient which is a good source of carbohydrate, is suitable for inclusion in catla diet (Manjappa et al. 2009). Inclusion of feedstuff with relatively high level of carbohydrate in formulated fish feeds is preferred in view of its protein sparing action, which makes the diet cost-effective (Hidalgo et al. 1993).

Catla, *Catla catla* (Hamilton, 1822), a popular species for polyculture in India, was used as the test species in the presently reported study. It is a surface feeder, mainly feeding on zooplankton (Jhingran 1991), but it also accepts artificial diets. The traditional feed mixture was supplemented with soya and maize flour and fed to catla fry in pellet form over a culture period of 120 days.

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The performance of fish and the economic implications of supplementing the traditional feed mixture are reported in this communication.

This study has been carried out in accordance with the country's regulations on animal experimentation.

MATERIALS AND METHODS

Feed ingredients and diets. Rice bran, groundnut cake, soybean, maize, tapioca flour, and vitamin-mineral mixture were procured locally. Maize flour was obtained by milling the shade-dried maize, while soya flour was obtained by powdering soybean kept soaked in water overnight and dried under shade. All the ingredients were sieved using ISI standard mesh No. 1 (Anonymous 1975). Proximate composition of the ingredients used is given in Table 1.

The control diet (T_0) and the two test diets (T_1 and T_2) were formulated as per the composition given in Table 2. Tapioca flour was used as the binder in the control diet, while the maize flour included uniformly at 39% in the test diets, served as the binder. Soya flour was incorporated at 10% (T_1) and 20% (T_2) levels. The diets were prepared by the method of Jayaram and Shetty (1981) as follows. Ingredients were finely ground and mixed thoroughly with water to make dough. The dough was then transferred to an aluminium container and steam cooked in a pressure cooker at 103.42 kPa for 15 min. Vitamin-mineral mixture was mixed after cooling the dough. Pellets (2 mm diameter size) were prepared by a hand pelletizer and were air dried in an oven at 40°C. After drying, they were packed in airtight polythene bags and labelled.

Experimental setup. The experiment was carried out in triplicate in 9 outdoor cement tanks of 25 m³ (5 × 5 × 1 m) each, without a soil base, over a period of 120 days. Ground water was used to fill the tanks, maintaining a depth of 90 ± 5 cm throughout the experimental period. Catla fry (average weight 0.80–0.84 g) were stocked at a density of 40 per tank. They were fed every day once in the morning at 5% body weight. Fish were sampled every fortnight to measure body weight and length and the quantity of feed given was re-adjusted after each sampling, based on the weight recorded.

Water quality. Water samples from the experimental tanks were collected fortnightly between 0900 h and 1000 h for measuring temperature, dissolved oxygen, pH, free carbon dioxide, total alkalinity, and ammonia. Water temperature was recorded using a digital thermometer and pH was measured with a digital pH meter (LI-120, ELICO, India). Dissolved oxygen, free carbon dioxide, total alka-

linity, and ammonia were determined following standard procedures of APHA (Anonymous 1998). Plankton samples were also collected on fish sampling days, using a net made of No. 30 bolting silk cloth having 60 mm mesh size, by filtering 100 L of water from different locations of each experimental tank. Dry weight of plankton was determined by drying the filtrate in a hot-air oven at 80°C, till a constant weight was obtained.

Proximate composition. Proximate composition of the ingredients, diets and fish carcass was analyzed according to AOAC standards (Anonymous 1995) procedures as follows: moisture content by oven drying at 105°C for 24 h; crude protein (N × 6.25) by micro-Kjeldahl digestion, and distillation after acid digestion (Tecator-1002); crude lipid by Soxtec (Tecator-1043) and fibre by Fibretec (Tecator-1017) systems; ash by incineration at 550°C in a Muffle furnace to constant weight. Nitrogen-free extract (NFE) was computed by the difference method (Hastings 1976). The energy content of the feed ingredients and diets was calculated using values of 22.6 kJ · g⁻¹ for protein, 38.9 kJ · g⁻¹ for fat and 17.2 kJ · g⁻¹ for carbohydrate as NFE (Mayes 1990).

Carcass of five fish taken from each of the 3 replicate tanks at the termination of the feeding experiment was dried, ground and pooled for determining the proximate composition.

Enzyme assay. On termination of the experiment, analysis of protease, amylase, and lipase activity in the hepatopancreas and intestine was carried out using representative samples of five fish from each treatment, following the methods of Kunitz (1947), Bernfeld (1955), and Bier (1962), respectively.

Data processing and statistical analysis. Fish performance in terms of specific growth rate (SGR), feed conversion ratio (FCR), protein efficiency ratio (PER), and yield was calculated using the following formulae:

- Specific growth rate (SGR) = [(ln final weight – ln initial weight)/experimental duration in days] × 100.
- Feed conversion ratio (FCR) = Feed consumed [g] / Weight gain [g].
- Protein efficiency ratio (PER) = Weight gain [g] / Protein intake [g].
- Yield [g] = Mean body weight [g] × Total number of viable fish at harvest.

Comparison among treatments for various parameters was done by one-way analysis of variance (ANOVA), followed by Duncan's multiple range test at $P < 0.05$ (Duncan 1955, Snedecor and Cochran 1968).

Table 1

Proximate composition (%) (mean ± standard error) of ingredients used in formulation of feed for *Catla catla*

Parameter	Rice bran	Groundnut oil cake	Maize	Soya flour	Tapioca flour
Moisture	13.76 ± 0.22	9.28 ± 0.18	10.78 ± 0.06	9.88 ± 0.21	9.80 ± 0.42
Crude protein	8.56 ± 0.55	32.85 ± 0.00	11.05 ± 0.27	34.00 ± 0.00	3.14 ± 0.00
Fat	2.23 ± 0.06	8.60 ± 0.10	3.70 ± 0.05	18.70 ± 0.10	0.40 ± 0.00
Ash	21.21 ± 0.03	6.16 ± 0.39	1.58 ± 0.03	4.43 ± 0.07	1.50 ± 0.03
Crude fibre	31.60 ± 0.15	24.80 ± 0.21	3.60 ± 0.03	6.60 ± 0.05	3.40 ± 0.01
Nitrogen-free extract	22.64	18.31	69.29	26.39	81.76
Gross energy [kJ · g ⁻¹]	6.69	13.92	15.85	19.50	14.93

RESULTS

Soya flour had the highest protein (34.0%) and fat (18.7%) contents, while maximum values for crude fibre (31.6%) and ash (21.21%) were recorded in rice bran. Carbohydrate (NFE) was the highest (81.76%) in tapioca flour (Table 1). Incorporation of soya and maize flour to the traditional feed mixture led to an increase in protein, fat and NFE levels and a decrease in fibre and ash contents in the test diets (Table 2).

The following values of the water quality parameters were recorded:

- water temperature = 27–31°C,
- pH = 7.4–8.6,
- dissolved oxygen = 8.46–12.09 ppm,
- free carbon dioxide = 0–1.2 ppm,

- total alkalinity = 56.24–76.94 ppm,
- ammonia = 0–14.69 $\mu\text{g} \cdot \text{L}^{-1}$.

The average plankton dry weight varied from 3.81 to 46.23 $\text{mg} \cdot 100 \text{L}^{-1}$, increasing with the progress of the experiment.

Data on the final weight and length of catla is given in Table 3, while the growth trend is depicted in Fig. 1. Catla showed significantly higher ($P < 0.05$) growth with diets T₁ and T₂. The final average weight was the highest (49.82 g) under T₂ treatment; however, there was no significant difference between the two treatments. SGR values followed the trend of fish weight at harvest with the highest value (3.42%) under T₂ treatment (Table 3).

FCR was better with the test diets, though not significantly different from that of the control. It was the lowest (1.91)

Table 2

Ingredient proportion and proximate composition of experimental feed for *Catla catla* (mean \pm standard error)

Ingredient / parameter	Diet		
	T ₀	T ₁	T ₂
Ingredient proportion [%]			
Rice bran	45	25	20
Groundnut cake	45	25	20
Maize	—	39	39
Soya flour	—	10	20
Tapioca flour	9	—	—
Vitamin-mineral mixture*	1	1	1
Cost of feed per kg [INR]	9.60	9.59	10.01
Proximate composition [%]			
Moisture	5.23 \pm 0.07	5.38 \pm 0.12	6.61 \pm 0.03
Crude protein	17.88 \pm 0.27	18.27 \pm 0.06	18.82 \pm 0.23
Fat	5.02 \pm 0.17	5.90 \pm 0.15	6.43 \pm 0.14
Ash	15.95 \pm 0.15	9.98 \pm 0.02	8.80 \pm 0.07
Crude fibre	21.60 \pm 0.34	16.60 \pm 0.13	14.40 \pm 0.11
Nitrogen-free extract	34.32	43.87	44.94
Gross energy [$\text{kJ} \cdot \text{g}^{-1}$]	11.90	13.97	14.48

*Supplevite M; Sarabhai Chemicals, Mumbai, India; INR = Indian rupee (USD 1 was worth approximately INR 45 in 2011).

Table 3

Growth parameters of *Catla catla* fed experimental diets (mean \pm standard error)

	Diet		
	T ₀	T ₁	T ₂
Final length [cm]	14.70 ^b \pm 1.00	16.46 ^a \pm 0.24	15.85 ^a \pm 0.15
Final weight [g]	36.01 ^c \pm 2.87	46.36 ^b \pm 0.69	49.82 ^a \pm 0.39
Specific growth rate [%]	3.16 ^b \pm 0.08	3.38 ^a \pm 0.02	3.42 ^a \pm 0.12
Food conversion ratio	2.51 ^a \pm 0.24	2.09 ^a \pm 0.03	1.91 ^a \pm 0.25
Protein efficiency ratio	2.11 ^b \pm 0.20	2.71 ^a \pm 0.05	2.81 ^a \pm 0.12
Survival [%]	74 ^a \pm 1.20	76 ^a \pm 0.80	76 ^a \pm 0.60
Net production [g per 25 m ² per 120 days]	657.40 ^b \pm 20.79	864.80 ^a \pm 46.36	931.40 ^a \pm 49.82
Cost of feed [INR per 25 m ² per 120 days]	15.84	17.33	17.81
Income [INR 40 per kg]	26.30	34.59	37.26
Gross returns [INR per 25 m ² per 120 days]	10.46	17.26	19.45
Carcass proximate composition [%]			
Moisture	80.22 ^a \pm 0.64	77.19 ^b \pm 1.1	77.28 ^b \pm 0.36
Protein	13.40 ^a \pm 0.0	13.75 ^a \pm 0.25	13.81 ^a \pm 0.02
Fat	2.36 ^c \pm 0.01	3.02 ^b \pm 0.01	3.33 ^a \pm 0.08
Ash	2.89 ^a \pm 0.20	3.35 ^a \pm 0.27	2.95 ^a \pm 0.33

INR = Indian rupee (USD 1 was worth approximately INR 45 in 2011); Figures in the same row with the same superscript are not significantly different ($P > 0.05$).

in T₂ treatment, in which the growth reached maximum and was the highest in control (2.51). PER in the two treatments improved significantly ($P < 0.05$); it was the highest in T₂ (2.81) treatment (Table 3).

The survival rate of fish under different treatments is shown in Table 3. The overall survival ranged from 74% to 76%. Fish production was significantly higher ($P < 0.05$) in the two test treatments. As a result, gross returns were also higher under them (Table 3). Profitability from the control (T₀) and T₁ and T₂ treatments works out to 39.8%, 49.9%, and 52.2%, respectively.

Carcass composition was affected due to feed, mainly in terms of fat and moisture. Moisture decreased significantly in the two test treatments and showed an inverse relation with fat. A progressive increase in fat content was noticed with increasing dietary soya flour level. Protein and ash contents fluctuated within a narrow range between treatments and the control (Table 3).

Soya and maize flour supplementation positively affected ($P < 0.05$) the activity of all the 3 digestive enzymes. As compared to the intestine, hepatopancreas showed higher amylase and lipase activity in the control as well as the treated fish (Table 4).

DISCUSSION

Both the test diets (T₁ and T₂) significantly ($P < 0.05$) enhanced the growth of catla, diet T₂ with higher level of soya inducing better growth. Superior performance of fish with the test diets reflects on the ability of catla to utilize the two supplemented ingredients effectively. These ingredients improved the nutrient content of the test diets, with an increase in protein, fat and NFE levels and a decrease in crude fibre and ash levels (Table 2). Products derived from soybeans have been shown to be excellent feed ingredients for commercially important cultured fish and prawn species (Storebakken et al. 2000, Grisdale-Helland et al. 2002, Kaushik et al. 2004, Davis et al. 2005, Dias, 2005, Fafioye et al. 2005, Hasanuzzaman et al. 2009). Apart from the high level of protein containing a reasonably balanced amino acid profile, the digestibility of the protein fraction

of soybean products has consistently been reported to be more than 90% for various fish species. The protein digestibility coefficients compare favourably with those of any other high-quality protein feedstuff such as various fish meals (Anonymous 1993). Lipid digestibility is also rather high in soybean meal (SBM). Experiments conducted with channel catfish in ponds have revealed that diets containing 28% to 32% crude protein primarily from SBM produce growth equivalent to diets containing some animal protein, such as fish meal and meat and bone meal (Robinson et al. 2000, Li et al. 2000). Excellent results have also been obtained using SBM-based diets with smaller sizes of mirror carp (Cremer et al. 2001).

Carp and tilapia are known to utilize high levels of carbohydrate (Anderson et al. 1984, Satoh 1991). Results reported by Manjappa et al. (2009) demonstrate the ability of catla to utilize diets with maize to the extent of 40%. In the present study, maize was included at 39% level. As a carbohydrate source, it possibly spared protein for growth (Hidalgo et al. 1993). Further, carbohydrates improve the pelleting quality and nutrient value of diets (Lovell 1989). Compared to rice bran and oil cake, soya and maize flours had low fibre levels (Table 1) and the addition of these two ingredients to the traditional feed mixture resulted in reduced crude fibre content in the test diets (Table 2). Fibre content of the diet affects feed digestibility and food retention in the gut, thereby influencing absorption of nutrients. Hilton et al. (1983) observed that dry matter digestion coefficients of the diets declined as the fibre level increased, whereas Meurer et al. (2003) reported a decrease in food retention time with increased crude fibre.

Water quality parameters monitored over the experimental duration were within the acceptable optimum range for catla, with no drastic variation between treatments and hence would not be a contributory factor for the difference in fish growth among treatments. Fish survival (74% to 76%) was not affected ($P > 0.05$) by soya and maize supplementation. The higher production ($P < 0.05$) obtained with the test diets was the result of higher fish growth.

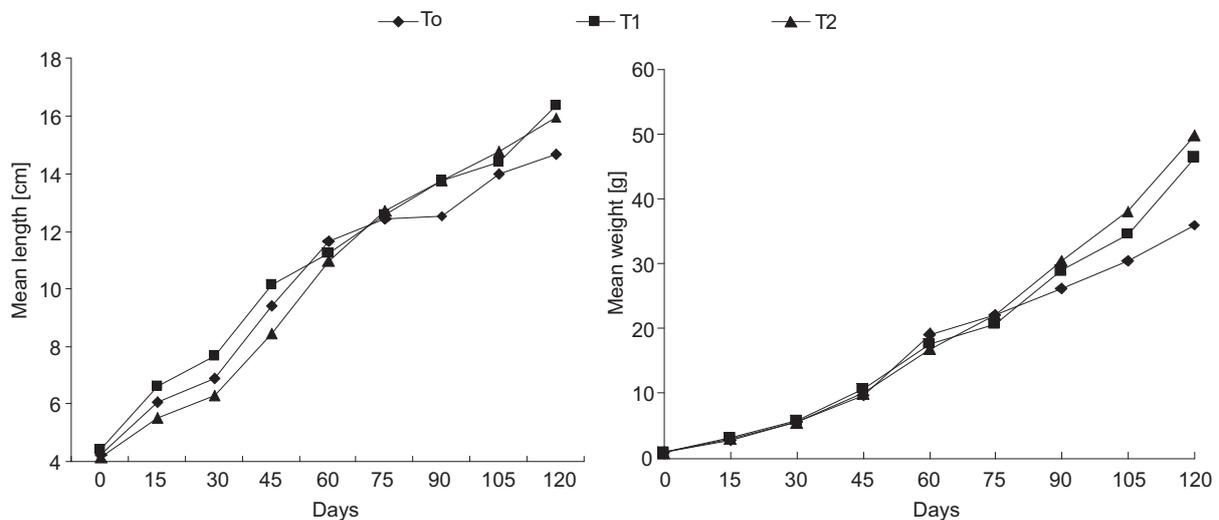


Fig. 1. Mean length and weight of *Catla catla* during the experiment

Digestive enzyme activity in the intestine and hepatopancreas of *Catla catla* fed experimental diets (mean \pm standard error)

Table 4

Diets	Protease		Amylase		Lipase	
	Intestine	Hepatopancreas	Intestine	Hepatopancreas	Intestine	Hepatopancreas
T ₀	3.72 ^c \pm 0.19	4.41 ^c \pm 0.36	4.35 ^b \pm 0.29	26.08 ^c \pm 0.67	1.68 ^b \pm 0.18	3.37 ^b \pm 0.01
T ₁	5.86 ^a \pm 0.90	5.68 ^b \pm 0.02	6.32 ^a \pm 0.42	38.06 ^a \pm 0.26	2.25 ^a \pm 0.61	6.50 ^a \pm 0.61
T ₂	4.26 ^b \pm 0.13	6.21 ^a \pm 0.19	6.47 ^a \pm 1.30	33.04 ^b \pm 0.94	2.87 ^a \pm 0.37	7.12 ^a \pm 0.75

Enzyme activity is expressed as μ moles of product liberated per min per mg tissue protein at 30°C; Values with the same superscript in each row are not significantly different ($P > 0.05$).

Soybean is known to contain antinutrients (Francis et al. 2001), particularly a trypsin inhibitor that affects fish growth; however, no negative effect of soya flour on fish growth was observed in this study. Soaking of soybean before milling and the heat treatment during feed preparation would have eliminated this antinutrient.

A decrease ($P < 0.05$) in carcass moisture content observed in catla fed the test diets reflects true growth. This is consistent with the findings of Chamundeswari Devi et al. (1999) who recorded lowest moisture in the muscle of rohu fed 20% soybean incorporated diet. The significant ($P < 0.05$) increase in fat level in fish receiving the test diets indicates enhanced lipid production which can be related to the fat and NFE levels of the diets. Fafioye et al. (2005) observed similar increase in carcass lipid of *Clarias gariepinus* fed soya-based diets. Fountoulaki et al. (2003) stated that higher carcass lipid was associated with increase in the efficiency of metabolism in gilthead bream fingerlings. Dietary lipid level is known to influence carcass lipid positively (Bazaz and Keshavanath 1993, Manjappa 2009). Further, unutilized dietary carbohydrate is also converted into fat.

Enzymes play an important role in the digestion and utilization of feed (Dabrowski and Glogowski 1977). The test diets employed in this study significantly ($P < 0.05$) influenced digestive enzyme activity. Higher digestive enzyme activity in fish receiving the test diets would have resulted in better diet utilization, leading to higher growth. A positive relationship was recorded between hepatopancreatic amylase activity and dietary carbohydrate level, a clear indication that dietary nutrients influence digestive enzyme activity (Gangadhara et al. 1997, Manjappa et al. 2009).

Partial replacement of rice bran and groundnut cake from the control diet with soya and maize marginally increased the cost of diet T₂, but not diet T₁. The cost (Indian rupee: INR) worked out to INR 10.01 per kg⁻¹ for diet T₂ as against INR 9.60 for the control (Table 2). (USD 1 was worth approximately INR 45 in 2011.) The superior fish growth and production obtained under T₂ treatment (38.35% and 41.68% respectively over control) more than compensates for the cost increment as seen from the higher returns under the two treatments (Table 3). Based on the present results, it may be concluded that supplementation of the traditional feed mixture with soya and maize, partly replacing rice bran and groundnut cake, is economically beneficial.

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