*Pakistan J. Zool., vol. 46(1), pp. 61-66, 2014.*

# Evaluation of Locally Available Plant Meals as a Replacement of Soybean Meal as Dietary Protein Source for *Catla catla* Fingerlings

**Rafia Tayyab,[1] Noor Khan,[1]* Muhammad Sharif Mughal,[1] Naureen Aziz Qureshi,[2] Muhammad Naeem Khan[3] and Fayyaz Rasool[1]**

[1]*Department of Fisheries and Aquaculture, University of Veterinary and Animal Sciences, Lahore, P.O. Box. 54000, Pakistan*

[2]*Department of Wildlife and Fisheries, Govt. College University, Faisalabad*

[3]*Department of Zoology, University of the Punjab, Lahore.*

**Abstract.-** The possibility of locally available plant meals as a replacement of soybean meal as dietary protein source for *Catla catla* fingerlings was evaluated under intensive rearing condition in glass aquarium for 60 days. Three iso-nitrogenous (24% crude protein) diets (D1, D2 and D3) were prepared and fed to fish at 3% wet body weight per day. Each treatment group had two replicates. The data was analyzed by employing one way ANOVA. The survival rate of fish as observed in D1 and D2 was 75%, while it was 100% in D3. The study showed across the board poor fish growth in all the treatment groups with non-significant difference ($P \geq 0.05$) in final weight, gain in weight, SGR and FCR. However, a significant difference ($P < 0.05$) was observed for fish whole body crude protein, crude lipid, ash and crude fiber contents. Crude protein and ash contents significantly increased with the increased level of plant protein meals while crude lipid percentage decreased with increasing the plant meals. It can be concluded that this replacement of soybean meal with other local plant protein meals did not have significant effect on fish growth. However, inclusion of higher level of plant meals (Ipil ipil leaf meals, sunflower meals and rice polish) resulted in increased crude protein, fiber and ash contents of the fish.

**Key Words:** Plant source meals, *Catla catla*, growth, proximate composition.

## INTRODUCTION

Feed and nutrition is critical in fish farming because feed represent over 60% of the production costs (Li and Wang, 2004). Presently, aquaculture production is greatly constrained by undersupply, scarcity and high cost of conventional fish feed.

The ever growing cost and uncertainties about the quality and availability of fish and soybean meal have compelled the aquaculture nutritionists and feed manufactures to search and replace this with cheap and readily available plant protein sources. Besides cost, improvement in feed and nutrition of aquatic species is also necessary by enhancing the quantity and nutritional quality of fish flesh (Khan *et al.,* 2011). Identification of suitable alternate protein sources for inclusion in fish feeds becomes imperative to counter the scarcity of fish meal and soybean meal. A variety of plant protein sources including sunflower meal, ipil ipil leaf meal (Amisah *et al.,* 2009; Bairagi *et al.,* 2004), cotton seed meal, raw soybean meal (Patnaik *et al.,* 2005) and extract of Moringa leaves (Afuang *et al.,* 2003; Ritcher *et al.,* 2003) have been tested and included as alternatives to fish meal. Since sunflower meal contains about 38% crude protein (Shazadi *et al.,* 2006), rice polish 12-16% (Abid and Ahmed, 2009) and *Leucaena* leaf about 22.7%, these can be good candidate to replace as cheaper and quality ingredient in aqua feed (Atawodi *et al.,* 2008). The nutritive value and digestibility of Ipil ipil is higher than that of tropical grasses (Zamal *et al.,* 2009).

Pakistan, an agro-based country has the potential to sustain the production of various plants based feed ingredients (rice polish, sunflower meal and Ipil ipil, etc) that can be utilized to replace costly protein ingredients such as fish and soybean meals to produce cost effective aqua feeds. Recently few commercial aqua feed industries are utilizing costly imported soybean meal and incorporating them in the commercial fish feed. As already mentioned Pakistan has plenty of plant based feed ingredients that are sustainable and cost effective and can be utilized for our commercially important

* Corresponding Author: noorkhanuvas@yahoo.com

0030-9923/2014/0001-0061 $ 8.00/0

fish species. *Catla catla* is one of the three most popular commercially important culturable herbivorous fish in South Asia including Pakistan. It is a fast growing and surface feeding fish and can utilize efficiently mash feed from the water surface as well. The present study was therefore planned to study the effect of locally available plant based feed ingredients diet with the replacement of soybean meal diet for the survival, growth and body composition of *Catla catla* fingerlings.

## MATERIALS AND METHODS

The study was carried out at Research and Training Facilities, Department of Fisheries and Aquaculture, University of Veterinary and Animal Sciences, Ravi Campus, Pattoki, Pakistan under intensive rearing conditions. Fingerlings of *Catla catla* (average weight 8.0 g) were procured from Fisheries Research and Training Institute, Manawan, Lahore and kept in glass aquaria (60.96 × 55.88 × 40.64 cm) by using two replicates of each diet for 60 days. Fortnightly morphometric measurements (Total Body Weight and Total Body Length) of fish were conducted to monitor the growth and adjusting the daily ration requirement. Growth indices such as net weight gain (NWG), feed conversion ratio (FCR), and specific growth rate (SGR) were calculated by using following formulae:

NWG = Final body weight (g) – Initial body weight (g)
FCR =Feed intake (g) / Wet weight gain
SGR %/day = ln(W1 )- ln(W2 ) x100 /No. of days

*Artificial feed formulation*

Three types of experimental mash feeds (D1, D2 and D3) were prepared by using fish meal, soybean meal, ipil ipil leaf meal, rice polish, wheat bran, sunflower meal and Nutrimix as feed ingredients (Table I). Experimental feeds such as 0% (D1), 20% (D2) and 40% (D3) soybean meal was replaced with plant based ingredients meals like Ipil ipil leaf meal, sunflower meal and rice polish. The fish was fed twice a day at 3% fish wet body weight up for 60 days.

*Chemical analysis*

The formulated feeds, feed ingredients and fish samples were analyzed for dry matter, ash, crude protein, crude lipid and gross energy as per AOAC (2006).

*Water quality parameters*

Water quality parameters *viz*., temperature, dissolved oxygen (DO), total dissolved solids (TDS) and pH were monitored on daily basis, while nitrates were determined on fortnightly basis by using spectrophotometer (APHA, 1998).

*Statistical analysis*

The data collected after completion of trials were statistically analyzed using Analysis of Variance (ANOVA) (Steel *et al.,* 1997) using Minitab Statistical package 1.5. Comparison of means was done by using Tuky's t-test. The significance of probability was tested at $P<0.05$ level.

## RESULTS

Growth parameters of the current study showed non-significant differences ($p \geq 0.05$) in final weight gain and net weight gain in all the treatment groups (Table II). The SGR was 0.62%, 0.36% to 0.66%/day in D1, D2 and D3, respectively also revealed non-significantly poor growth performance. There were non-significantly ($p \geq 0.05$) higher values of feed conversion ratio (FCR) D1 4.71, D2 7.86 and D3 4.79 (Table II).

Proximate composition of whole body revealed significant differences ($p < 0.05$) among different treatment groups like; ash contents were significantly higher in D1 and D2 compared to D3 (Table III). Crude protein percentage was found significantly higher in D3 than D1 and D2. Crude lipid contents were found significantly higher in D1 followed by D2 and D3, respectively. Crude fiber was significantly higher in D2 followed by D1 and D3 (Table III).

The results of physico-chemical parameters such as temperature ranged from (28-30°C), dissolved oxygen (2.4–3.58 mg/l), pH (7.98–8.16), salinity (0.85–0.91ppt), total dissolved solids (1531.83-1646.99µs/cm) and nitrates (0.00017mg/l).

**Table I.- Ingredients composition and crude protein (CP) concentration of the experimental diets.**

| Feed ingredients | D1 (0% soybean meal) | CP% | D2 (20% soybean meal) | CP% | D3 (40% soybean meal) | CP% |
|---|---|---|---|---|---|---|
| Fish meal | 10 | 5.54 | 10 | 5.54 | 10 | 5.54 |
| Soybean meal | 20 | 8.05 | 10 | 4.03 | 0 | 0.0 |
| Ipil ipil leaf meal | 0 | 0 | 20 | 4.6 | 40 | 9.2 |
| Rice polish | 45 | 6.4 | 30 | 4.27 | 15 | 2.13 |
| Wheat bran | 25 | 4.15 | 23 | 3.81 | 21 | 3.48 |
| Sunflower meal | 0 | 0 | 7 | 1.86 | 14 | 3.71 |
| Total | 100 | 24.14 | 100 | 24.11 | 100 | 24.06 |

CP, crude protein % in diet.

**Table II.- Comparison of growth parameters of *Catla catla* fingerlings fed 24% crude protein diets (means are of two replicates n = 2).**

| Parameters | D1 (0% soybean meal) | D2 (20% soybean meal) | D3 (40% soybean meal) | PSEM | *ANOVA (P value)* |
|---|---|---|---|---|---|
| No. of fish stocked | 8 | 8 | 8 | - | - |
| Survival % | 75 | 75 | 100 | - | - |
| Initial wt (g) | 8.03[a] | 8.22[a] | 9.46[a] | 3.52 | 0.910 |
| Final wt (g) | 11.48[a] | 11.24[a] | 13.58[a] | 4.66 | 0.865 |
| Net wt. gain (g) | 3.46[a] | 3.02[a] | 4.13[a] | 1.18 | 0.674 |
| SGR% | 0.62[a] | 0.36[a] | 0.66[a] | 0.12 | 0.476 |
| FCR | 4.71[a] | 7.86[a] | 4.79[a] | 1.8 | 0.283 |

*Figures with same letters are not significantly different (P > 0.05); PSEM, pooled standard error of mean.

**Table III.- Proximate composition of *Catla catla* fingerlings fed 24% crude protein diets**.

| Days | D1 (0% soybean meal) | D2 (20% soybean meal) | D3 (40% soybean meal) | PSEM | ANOVA (P value) |
|---|---|---|---|---|---|
| DM (%) | 99.0[a] | 98.75[a] | 99.39[a] | 0.495 | 0.510 |
| Ash (%) | 12.07[a] | 10.61[a] | 9.46[b] | 0.522 | 0.035 |
| CP (%) | 64.04[a] | 66.18[a] | 70.0[b] | 0.77 | 0.010 |
| CL (%) | 15.03[a] | 12.39[b] | 8.03[c] | 0.370 | 0.001 |
| NFE (%) | 1.86[a] | 2.58[b] | 1.25[c] | 0.12 | 0.003 |

*Figures with different superscripts are significantly different (P < 0.05); DM, dry matter; CP, crude protein; CL, crude lipid; NFE, nitrogen free extract; PSEM, Pooled standard error of mean.

Non-significant difference ($p \geq 0.05$) was observed for all the parameters among treatments except dissolved oxygen which revealed significant differences between D1 and D3 (Table IV).

## DISCUSSION

In present study growth parameters showed non-significant differences in final weight gain and net weight gain in all the treatment groups. Some mortality (25%) was observed in D1 and D2 while no mortality (0.0 %) was observed in D3 where soybean meal was replaced with 40% ipil-ipil leaf meal, 15% rice polish and 14% sunflower meal. The possible reason behind mortality and poor fish growth are not clear but can be related to low dissolved oxygen concentration in the aquarium which ranged from 2.4 to 3.5 mg/L. The SGR values during current study were 0.62%, 0.36% to 0.66%/day in D1, D2 and D3 revealed poor growth

**Table IV.- Physicochemical parameters of the experimental trial of *Catla catla* fingerlings fed 24% crude protein diets (means are of two replicates n = 2).**

| Parameters | D1 (0% soybean meal) | D2 (20% soybean meal) | D3 (40% soybean meal) | PSEM | *ANOVA (P value)* |
|---|---|---|---|---|---|
| Temp. (ºC) | 28.83[a] | 30.01[a] | 30.1[a] | 1.41 | 0.388 |
| DO (mg/l) | 3.58[a] | 2.72[ab] | 2.44[b] | 0.48 | 0.021 |
| pH | 8.16[a] | 7.98[a] | 8.01[a] | 0.10 | 0.076 |
| Salinity (ppt) | 0.85[a] | 0.91[a] | 0.88[a] | 0.12 | 0.800 |
| TDS (µs/cm) | 1646.997[a] | 1531.83[a] | 1563.224[a] | 160.1 | 0.594 |
| Nitrates (mg/l) | 0.00017[a] | 0.00017[a] | 0.00017[a] | 0.00 | 0.866 |

*Figures with different superscripts are significantly different ($P < 0.05$); DO, dissolved oxygen; TDS, total dissolved solids; PSEM, pooled standard error of mean.

performance. Similarly, the FCR values were also found poor and non-significantly different in D1 at 4.71, D2 at 7.86 and D3 at 4.79. Contrary to our results, Zamal *et al.* (2009) reported that 15% level of ipil-ipil in the diets have good nutritive values and have a significant effect on the growth, FCR and FCE of *Oreochromis niloticus***.** Their findings revealed that ipil-ipil leaf meal could be used as protein substitute up to 25% and optimum level 15% in diet of growing tilapia. Um-e-Kalsoom *et al.* (2009) reported feed conversion ratio (FCR) of blood meal (1.68 ± 0.96), followed by rice broken (1.64 ± 0.64) and wheat bran (1.59 ± 0.71) in hybrid fish (*Catla catla x Labeo rohita*) which are much lower and better than present study. Abid and Ahmed (2009) reported SGR 3.7% and FCR 1.5 for rohu fingerlings while feeding 100% rice polish. The possible reason of this contradiction in results of higher FCR in current study might be due to the differences in rearing conditions such as glass aquaria, low oxygen (stress), feed preparation, feed intake and inclusion of other plant based meals such as ipil ipil, sunflower meal, wheat bran etc. De Silva and Davy (1992) reported that feed digestibility plays an important role in lowering the FCR by efficient feed utilization.

In the present study, diet (D3) containing 40% Ipil ipil leaf meal, 15% rice polish and 14% sunflower meal comparatively showed slightly higher growth when compared with D2 and D1. 100% survival in D3 might be due to some growth promoting agents in plant source meals that may have enhanced the survival and growth of fingerlings in hypoxic condition. Hassan *et al.* (1994) has reported a trend of reduced growth performance of Indian major carps fed with diets containing 25% soaked *Leucaen*a diets, while Ghatnekar *et al*. (1983), by using 30-65*% Leucaena leucocephala* recorded no adverse effects on growth and even 15% level used in the diets have good nutritive values and played a significant role on the growth, FCR and FCE in *Oreochromis niloticus.*

The results of proximate composition of fish whole body revealed that increase in the level of both ipil-ipil leaf meal and sunflower meal increased the protein contents significantly. However, Crude lipid and ash contents during this study showed opposite trend. According to Zamal *et al.* (2009) Ipil ipil and leaf material of *Leucaena* is a good source of crude protein (CP), calcium (Ca) and phosphorus (P), and β-carotene that might enhanced the protein contents in the experimental fish.

The range of physico-chemical parameters were found within acceptable limit (Rahman *et al.*, 2009) except dissolved oxygen for *Catla catla* fingerlings. The reason of such differences is not clear but that might be associated with higher fish growth and metabolic rate in treatment group D3 where fish showed numerically higher growth compared to D1.

## CONCLUSIONS

It can be concluded from the present study that replacement of soybean meal with locally available plant meals (Ipil ipil leaf meal, sunflower meal and rice polish) did not significantly increase the growth of fish. However, higher level of

inclusion of Ipil ipil leaf meal and sunflower meal resulted in higher survival rate, increased crude protein and crude fiber contents. It can be suggested that ipil ipil whole plant meal may be incorporated instead of leaf meal with other local plant origin protein sources to prepare pelleted feed that binds the ingredients firmly and may be applied in field condition for long term basis (6-9 months). Additionally detailed chemical analysis of ipil ipil may also be done to explore the growth inhibiting factors for future research.

## ACKNOWLEDGEMENTS

The authors are extremely thankful to the Chairman, Department of Fisheries and Aquaculture, University of Veterinary and Animal Sciences, Lahore, Pakistan for providing facilities for conducting this study in Fish Hatchery aquaria.

## REFERENCES

ABID, M. AND AHMED, M.S., 2009. Growth response of *Labeo rohita* fingerlings fed with different feeding regimes under intensive rearing. *J. Anim. Pl. Sci.*, **19**: 45-49.

AFUANG, W., SIDDHURAJU, P. AND BECKER, K., 2003. Comparative nutritional evaluation of raw methanol extract residues and methanol extract of Moringa (*Moringa oleifera* Lam.) leaves on growth performance and feed utilization in Nile Tilapia. (*Oreochromis niloticus* L.). *Aquat. Res.*, **34**: 1147-1159.

AMISAH, S., OTENG, M. A. AND OFORI, J. K., 2009. Growth performance of the African catfish *(Clarias gariepinus)* fed varying inclusion levels of *Leucaena leucocephala* leaf meal. *J. appl. Sci. Environ. Managem.*, **13**:21-26.

AOAC (Association of Official Analytical Chemists) 2006. *Official methods of analysis*. Horwitz W. 18th edition 2006, Washington, DC, pp. 1018.

APHA, (American Public Health Association) 1998. *Standard methods for the examination of water and wastewater* (20th Ed.). American Public Health Association, the American Water Works Association and the Water Environment Federation, pp. 1220.

ATAWODI, S.E., MARI, D., ATAWODI, J.C. AND YAHAYA, Y., 2008. Assessment of *Leucaena leucocephala* leaves as feed supplement in laying hens. *Afr. J. Biotechnol.*, **7**:317-321.

BAIRAGI, A., GHOSH, S. K.S. AND RAY, A. K., 2004. Evaluation of the nutritive value of *Leucaena leucocephala* leaf meal, inoculated with fish intestinal bacteria *Bacillus subtilis* and *Bacillus circulans* in formulated diets for rohu (*Labeo rohita*) (Hamilton) fingerlings. *Aquat. Res.*, **35**: 436-446.

DE SILVA, S. S. AND DAVY, F.B., 1992. Fish nutrition research for semi-intensive culture systems in Asia. *Asian Fish. Sci.*, **5**:129-144.

GHATNEKAR, S. D., AUTI, D. G. AND KAMAT, V.S., 1983. Feeding leucaena to Mossambique tilapia and Indian major carps In: *Leucaena Research in the Asia Pacific Region*. Proceedings of a Workshop held in Singapore, 23-26, November, 1982. IDRC, Ottawa, Canada pp. 61-63.

HASSAN, M. R., ROY, P. K. AND AKAND, A. M., 1994. Evaluation of Leucaena leaf meal as a protein source for Indian major carp, *Labeo rohita*, fingerlings. In: *Fish nutrition research in Asia* (ed. S.S. De Silva), Proceeding of the Fifth Asian Fish Nutrition Workshop. Asian Fish. Soc. Spec. Publ. 9, Asian Fisheries Soc., Manila, Philippines, pp. 69-76.

KHAN, N., QURESHI, N.A., NASIR, M., RASOOL, F. AND IQBAL, K.J., 2011. Effect of artificial diet and culture systems on sensory quality of fried fish flesh of Indian major carps. *Pakistan J. Zool.*, **43**: 1177-1182.

LI, P. AND WANG, X. 2004. Nutritional value of fisheries by-catch and by- product meals in the diet of red drum (*Sciaenops ocellatus*). *Aquaculture*, **236**: 485–496.

PATNAIK, D., SAHU, N. P. AND CHAUDHARI, A., 2005. Effects of feeding raw soybean meal to fry of Indian major carp (*Catla catla*) on growth, survival and protein digestibility. *Israel J. Aquacult.-Bamidgeh*, **57**: 164-174.

RAHMAN, M. A., ZAHER, A. AND AZIMUDDIN, K. M., 2009. Development of fingerling production techniques in nursery ponds for the critically endangered reba carp, *Cirrhinus ariza* (Hamilton, 1807). *Turkish J. Fish. aquat. Sci.*, **9**: 165-172.

RITCHER, N., SIDDHURAJU, A. AND BECKER, K., 2003. Evaluation of nutritional quality of moringa (*Moringa oleifera Lam.*) leaves as alternative protein source for tilapia *(Oreochromis niloticus L.). Aquaculture*, **217**: 599-611.

SHAZADI, T., SALIM, M., UM-E-KALSOOM AND SHAHZAD, K., 2006. Growth performance and feed conversion ratio (FCR) of hybrid fingerlings (*Catla catla* X *Labeo rohita*) fed on cottonseed meal, sunflower meal and bone meal. *Pak. Vet. J.*, **26**: 163-166.

STEEL, R. G. D., TORRIE, G. H. AND DICKEY, D. A., 1997. *Principal and procedures of statistics: A biometrical approach.* (3rd Ed.) McGraw Hill, New York.

UM-E-KALSOOM, SALIM, M., SHAHZADI, T. AND BARLAS, A., 2009. Growth performance and feed

conversion ratio (FCR) in hybrid fish (*Catla catle x Labeo rohita*) fed on feed bran, rice broken and blood meal. *Pak. Vet. J.*, **29**: 55-58.

ZAMAL, H., BARUA, P., BELAL UDDIN AND ISLAM, K.S., 2009. Estimation of growth and financial analysis through the application of ipil-ipil *(Leucaena leucocephala)* leaf meal as supplements to soybean and fish meal in the diet of juvenile monosex tilapia *(Oreochromis niloticus). Int. Aquat. Feed.*, **12**:36-42.

*(Received 20 August 2013, revised 29 September 2013***)**