

PROTEIN SOYBEAN AS SUBSTITUTE OF FISHMEAL IN PRACTICAL DIETS FOR CATFISH *CLARIAS BATRACHUS* (INDIAN MAGUR)

Dr. Ravindra Singh

Associate Professor, Department of Zoology, K.B.P.G College Mirzapur, U.P., India

Abstract

Plant protein soybean was evaluated as replacements for fish meal in practical diets prepared with 40% crude protein to assess the performance of *Clarias batrachus* fed different levels. Diets-1-4 contained 65, 130, 195, 260 g kg⁻¹ of raw soybean whereas diets 5-8 contained processed soybean with same level as that of diets- 1-4. Other ingredients like sesame oil cake, rice bran, wheat flour, mineral premix and cod liver oil were also added in diets to fulfill the nutritional requirements. Fish meal (as chief protein source) was used to prepare reference diet (Diet-R). The study was conducted in fresh water in 18 aquariums in duplicates equipped with aeration systems. After 60 days, the weight gain of catfish fed diet-5 (with 65 g kg⁻¹) and diet-6 (with 130 g kg⁻¹) processed soybean was higher than that observed in fish fed on reference as well as diets 1-4 and 8th. Other growth parameters Growth % gain, Specific growth rate (SGR), Viscero somatic index (VSI) and Hepato somatic index (HSI) were also recorded high in diets 5th and 6th. This indicates that soybean meal could be substitute of fishmeal in aquaculture feed.

Keywords: *Clarias batrachus*, Fish meal, Growth parameters, Soybean meal, Crude protein.

INTRODUCTION

Fish culture now-a-days has been growing at a rapidly and is considered as the fastest growing food industry (10% per annum). capture fisheries in past years declined and replaced by farmed fish production. Aquaculture production is directly related to proper management of preparation of balanced nutritious diets. Because of these good-quality diets success in semi-intensive and intensive production systems are achieved (Sookying & Davis, 2011). Further, in any diet protein is considered as most important ingredient that's why its quality is a very important nutritional aspect in aquaculture (Garcia *et al.*, 2008). Currently, fish meal is major sources of protein used in aquaculture diets. In present scenario, fishmeal is found to be most costly ingredient due to its demand which increased more than two fold in recent years (FAO, 2012). Along with this also its consumption in fish food increased from 0.8 million tons to 1.7 million tons in recent years (Tacon & Metian, 2008). Hence as the production of aquaculture increased accordingly the demand of this protein is increased. Studies have verified that there are many other alternative protein sources are available which can replace fishmeal without affecting fish performance (Ogello *et al.*, 2014). Among different protein sources available, plant protein is considered as best source in aquaculture and in between plant source, soybean protein has been most acceptable candidate used for

replacement of fishmeal because of its high protein content (Ao *et al.*, 2010; Min *et al.*, 2009). Also, soybean protein has a low carbohydrate and fiber content, high digestibility, and a good amino acid balance, compared with other protein sources of vegetable origin (Gamboa *et al.*, 2013). From advantages, it is clear that soybean is very important which may replace fishmeal effectively in developing aquaculture. Hence there is need to much more implementation of this protein source in fish diet. The objective of this study was to add soybean as substitute of fishmeal in practical diets for catfish *Clarias batrachus* (Indian magur).

MATERIAL AND METHODS

Eight diets (1-4 contained raw soybeans, 5-8 contained processed full-fat soybean) with 40% crude protein were formulated using locally available materials. All ingredients were mixed well and with the help of mechanical pelletizer pellets of 0.5 mm were produced. These were then dried (60° - 62°C) in oven and stored for later use. To all diets was added a mineral and amino acid (methionine + lysine) premix. 1% chromic oxide (Cr₂O₃) was added as an external indigestible marker for digestibility estimations.

Processing of soybean

Processing of soybean was done eliminate anti-nutritional factors (ANFs). For this soybeans were hydrothermally treated at 15 psi and 121°C for 15 min. Fishmeal-based diet was used as the reference/control diet.

Experimental design

Fry of catfish were obtained from fish farm, Ramgarh, Gorakhpur and put in glass aquaria (60 × 30 × 30 cm). Temperature was maintained at 25 ± 1°C and light cycle regulated to 12:12 light/dark. Aquarium water was renewed daily with water adjusted to laboratory temperature (25°C). During a 10-day acclimation period, fish were fed the fishmeal reference diet (Diet-R). Fish (mean body weight of 0.55 g) were randomly distributed in aquaria (at 15 fish per aquarium) with two replicates. All fish were fed twice daily at 08:00 and 16:00 hours at a rate of 5% BWd⁻¹ over a period of 60 days.

Parameters

At the end of experiment, the fish from all the treatments were individually weighed in gram and measured in millimetre. The fishes were then processed for subsequent analyses. VSI, HSI, Growth parameters like Live weight gain (g), Growth % gain, SGR, Protein efficiency ratio (PER), feed conversion ratio (FCR), gross protein retention (GPR), and gross energy retention (GER) were calculated using standard method (Steffens, 1989). Digestive enzyme activity were calculated following different procedures, protease (Kumar, 1984), amylase (Kruger & Osborne, 1986), lipase (Worthington, 1991; Zamani *et al.*, 2009) and cellulose (Sadasivam, 1996). Estimation of glycogen from Liver and Muscle was done (Dubois *et al.*, 1956).

Proximate analysis was done following (Horwitz *et al.*, 1970). Statistical analysis: One way ANOVAs was applied using OPSTAT of CCSHAU, Hisar.

RESULTS AND DISCUSSION

Growth performance in terms of live weight gain, growth percent gain in body weight, and specific growth rate increased with an increase in the inclusion levels of processed full-fat soybean in the diets however maximum increased was recorded in diet-5 and 6 where 65 and 130 g kg⁻¹ of soybean incorporated in cat fish diet and decreased in growth performance was significantly (P < 0.05) observed with increased amount of raw soybean in the diets. FCR was greatly improved and PER was enhanced in groups fed on

diets containing processed full-fat soybean in comparison to fishes fed on raw soybean based diets and reference diet (Table 1). (Yu *et al.*, 2013), study indicated that up to 40% fish meal (FM) protein, based on the broken-line analysis of SGR, can be replaced with soybean meal (SBM) in diet for obscure puffer juveniles with supplemental lysine, methionine and taurine. Results of present study are in agreement with observations of (Antolovic *et al.*, 2012; Kikuchi & Furuta, 2009; Lim *et al.*, 2011; Yaghoubi *et al.*, 2016; Yigit *et al.*, 2010; Yu *et al.*, 2013) which claiming that approximately 16-40% FM protein can be replaced with SBM in diets for carnivorous fish species. Muscle glycogen was recorded lower in fish fed on processed soybean diets while values of this parameter was remained high in group fed on raw soybean as well as reference diets. Liver glycogen was observed high in group fed on processed soybean as compared to others groups. VSI, HSI (Table 1) and digestive enzyme activity (specific protease, amylase, and cellulase) (Table 2) were recorded higher in fish fed diets containing processed full-fat soybean in comparison to the fish fed the raw soybean diets (Table 2).

Further, the digestive enzyme activity appeared to depend on the inclusion levels of soybean in the diets. High levels of raw beans in diets appeared to depress digestive enzyme activity compared to diets containing the processed full-fat bean, also diet-5 and diet- 6 was found to be good diet for catfish where inclusions of processed soybean were 65 and 130 g kg⁻¹. Increase of fat and fibre was significantly ($P < 0.05$) higher in fish fed diets containing full-fat soybean compared to fish fed raw soybean-based diets (Table 3).

El-Sayed A-FM (2004) also evaluated many alternative plant protein sources in fish diet as partial or complete dietary replacement for fish meal. In present investigation mortality throughout the experiment was relatively nil which is differ from De Graaf and Janssen (1996) who observed only 41.5% survival rate for *Clarias batrachus*. The feed conversion ratio was significantly lower observations of present study are different from that by Olukunle *et al.* (2002) who reported the best FCR and SGR in fish fed the control diet. Present finding results showed that fish meal can be replaced partially with soybean meal at 25% and 50% inclusion levels which effectively increased growth parameters. These results are different from findings of Agbebi *et al.*, (2009) where fish meal can be replaced completely with blood meal at 100% with no adverse effects on the growth, survival and feed conversion ratio of *Clarias batrachus* juveniles. This difference may be due to origin of alternative protein source. Digestive enzyme activity in catfish are different those of cyprinids where long gut is present which is adapted to digest and absorb nutrients from plant feedstuffs (Bairagi *et al.*, 2002; Garg *et al.*, 2002). As catfish is omnivorous in nature hence length of gut is comparatively smaller then herbivorous fish that is the reason that plant protein could not be 100% replaced here.

Table 1. Effect of soybean meal as substitute of fish meal on growth Parameters, nutrient retention, VSI and HSI of cat fish under laboratory conditions

Parameter	Diet-R	Diet-1	Diet-2	Diet-3	Diet-4	Diet-5	Diet-6	Diet-7	Diet-8
Initial (wt.)	0.55±10	0.55 ± 10	0.55 ±10	0.55 ±10	0.55 ±10	0.55 ±10	0.55 ±10	0.55 ±10	0.55 ±10
Final (wt.)	1.19 ±11	1.09 ± 09	0.98 ±12	0.96 ±14	0.92 ±16	1.13 ± 09	1.11 ± 05	1.01± 05	1.09 ±09
Live wt. gain	0.64 ±13	0.56 ±11	0.43 ±11	0.41 ±14	0.37 ±14	0.58 ± 04	0.54 ± 06	0.46 ± 06	0.54 ±08
Growth %	116.36 ±09	101.81± 07	78.18 ±07	74.54 ±08	67.27 ±09	105.45±06	98.18 ±07	83.63±08	98.18 ±11
SGR	1.28 ±11	1.17 ±10	0.96 ±12	0.92 ±13	0.85 ±13	1.20 ± 07	1.14 ± 06	1.01± 06	1.14 ±08
FCR	20.16 ±10	20.72 ±10	14.54 ±10	14.21 ±15	13.04 ±15	11.50 ± 04	11.30 ±03	12.82 ±05	12.76±06
PER	0.07 ± 07	0.06 ± 07	0.07 ± 08	0.07 ± 08	0.07 ±11	0.11 ± 02	0.10 ± 02	0.06 ± 04	0.09 ±03
L. glycogen	1.14 ± 09	1.16 ± 06	1.14 ± 08	1.14 ± 07	1.15 ± 09	1.90 ± 04	1.86 ± 02	1.64 ±04	1.66 ±04
M.glycogen	0.71 ± 05	0.36 ± 05	0.65 ± 05	0.71 ± 09	0.63 ± 08	0.36 ± 01	0.38 ± 01	0.27 ± 03	0.34 ± 02
VSI	0.24 ± 02	0.12 ± 03	0.44 ± 03	0.38 ± 04	0.42 ± 05	0.22 ± 02	0.30 ± 02	0.45 ± 04	0.28 ± 04
(HSI)	0.02 ± 01	0.01 ± 01	0.10 ± 02	0.08 ± 03	0.09 ± 03	0.10 ± 02	0.05 ± 01	0.03 ± 01	0.07 ± 01

All the values are mean ± S.E. of mean.

Table 2. Effect of soybean meal as substitute of fish

Enzyme activity	meal on digestive enzymes activity of cat fish.								
	Diet-R	Diet-1	Diet-2	Diet-3	Diet-4	Diet-5	Diet-6	Diet-7	Diet-8
Protease	1.60 ± 04	0.72 ± 04	0.65 ± 05	0.60 ± 05	0.58 ± 04	1.50 ± 01	2.85 ± 02	1.70 ± 03	1.60 ± 04
Amylase	0.10 ± 02	0.50 ± 03	0.30 ± 04	0.30 ± 07	0.30 ± 03	0.50 ± 01	0.56 ± 02	0.52 ± 01	0.40 ± 05
Cellulase	0.23 ± 04	0.37 ± 04	0.35 ± 06	0.42 ± 09	0.49 ± 01	0.50 ± 00	0.55 ± 03	0.54 ± 00	0.57 ± 06
lipase	0.25 ± 05	0.32 ± 05	0.50 ± 07	0.23 ± 09	0.25 ± 05	0.52 ± 01	1.07 ± 04	0.80 ± 01	0.80 ± 02

All the values are mean ± S.E. of mean.

Table 3. Effect of soybean meal on proximate composition of cat fish

Parameters	Diet-R	Diet-1	Diet-2	Diet-3	Diet-4	Diet-5	Diet-6	Diet-7	Diet-8
	96.4 ± 04	97.6 ± 05	95.8 ± 05		87.8 ± 09	91.4 ± 03	92.8 ± 03	98.2 ± 05	91.8±06

CONCLUSION

The catfish shows high growth upto 25 to 50% replacement of fish meal by soybean protein. No further improvement in growth was recorded as level of soybean increased. However, growth performance was reduced at 100% of replacement of fish meal by soybean protein. The results of this study showed that in diet of catfish only 50% soybean protein can be effectively replaced by plant protein.

REFERENCES

1. Agbebi, O., Otubusin, S., & Ogunleye, F. (2009). Effect of different levels of substitution of fishmeal with blood meal in pelleted feeds on catfish *Clarias gariepinus* (Burchell, 1822) culture in net cages. *European Journal of Scientific Research*, 31(1), 6-10.
2. Antolović, N., Kožul, V., Antolović, M., & Bolotin, J. (2012). Effects of partial replacement of fish meal by soybean meal on growth of juvenile saddled bream (Sparidae). *Turkish Journal of Fisheries and Aquatic Sciences*, 12(2), 247-252.

2. Ao, X., Kim, H., Meng, Q., Yan, L., Cho, J., & Kim, I. (2010). Effects of diet complexity and fermented soy protein on growth performance and apparent ileal amino acid digestibility in weanling pigs. *AsianAustralasian Journal of Animal Sciences*, 23(11), 1496-1502.
3. Bairagi, A., Ghosh, K.S., Sen, S. K., & Ray, A. K. (2002). Enzyme producing bacterial flora isolated from fish digestive tracts. *Aquaculture International*, 10(2), 109121.
4. De Graaf, G., & Janssen, H. (1996). Artificial reproduction and pond rearing of the african catfish *Clarias gariepinus* in Sub-Saharan Africa. A hanbook: FAO, Roma (Italia).1-73.
5. Dubois, M., Gilles, K.A., Hamilton, J.K., Rebers, P.T., & Smith, F. (1956). Colorimetric method for determination of sugars and related substances. *Analytical Chemistry*, 28(3), 350-356.
6. El-Sayed, A.F.M. (2004). Protein nutrition of farmed tilapia: searching for unconventional sources. Paper presented at the New dimensions in farmed tilapia: Proceedings of the Sixth International Symposium on Tilapia, *Aquaculture*, 392-402.
7. FAO. (2012). The state of World Fisheries and Aquaculture *FAO Roma*, 24-26.
8. Gamboa Delgado, J., Rojas-Casas, M.G., Nieto-López, M. G., & Cruz-Suárez, L.E. (2013). Simultaneous estimation of the nutritional contribution of fish meal, soy protein isolate and corn gluten to the growth of Pacific white shrimp (*Litopenaeus vannamei*) using dual stable isotope analysis. *Aquaculture*, 380, 33-40.
9. García Ulloa Gomez, M., López-Aceves, L.A., PoncePalafox, J.T., Rodríguez-González, H., & Arredondo Figueroa, J.L. (2008). Growth of fresh-water prawn *Macrobrachium tenellum* (Smith, 1871) juveniles fed isoproteic diets substituting fish meal by soya bean meal. *Brazilian Archives of Biology and Technology*, 51(1), 57-65.
10. Garg, S., Kalla, A., & Bhatnagar, A. (2002). Evaluation of raw and hydrothermically processed leguminous seeds as supplementary feed for the growth of two Indian major carp species. *Aquaculture Research*, 33(3), 151163.
11. Horwitz, W., Chichilo, P., & Reynolds, H. (1970). Official methods of analysis of the Association of Official Analytical Chemists. *Official methods of analysis of the Association of Official Analytical Chemists*.1-1015.
12. Kikuchi, K., & Furuta, T. (2009). Inclusion of blue mussel extract in diets based on fish and soybean meals for tiger puffer *Takifugu rubripes*. *Fisheries Science*, 75(1), 183-189.
13. Kruger, J.M., & Osborne, C.A. (1986). Etiopathogenesis of uric acid and ammonium urate uroliths in nonDalmatian dogs. *Veterinary Clinics of North America: Small Animal Practice*, 16(1), 87-126.
14. Kumar, A. (1984). T. Friedmann: Gene therapy - Fact and fiction. In *Biology's New Approaches to Disease Based on the Gene*, 29, 355-358.
15. Lim, S.J., Kim, S.S., Ko, G.Y., Song, J.W., Oh, D.H., Kim, J.D., Lee, K.J. (2011). Fish meal replacement by soybean meal in diets for Tiger puffer, *Takifugu rubripes*. *Aquaculture*, 313(1-4), 165-170.
16. Min, B., Cho, J., Chen, Y., Kim, H., Yoo, J., Lee, C., Kim,
17. (2009). Effects of fermented soy protein on growth performance and blood protein contents in nursery pigs. *Asian-Australasian Journal of Animal Sciences*, 22(7), 1038-1042.
18. Ogello, E.O., Munguti, J.M., Sakakura, Y., & Hagiwara, A. (2014). Complete replacement of fish meal in the diet of Nile tilapia (*Oreochromis niloticus* L.) grow-out with alternative protein sources. A review. *International Journal of Advanced Research*, 2(8), 962-978.

19. Olukunle, O., Ogunsanmi, A., Taiwo, V., & Samuel, A. (2002). The nutritional value of cow blood meal and its effects on growth performance, haematology and plasma enzymes of hybrid catfish. *African Journal of Online*, 5(1), 22-29.
20. Sadasivam, S. (1996). *Biochemical Methods: New age international*. 2(2), 124-126.
21. Sookying, D., & Davis, D.A. (2011). Pond production of Pacific white shrimp (*Litopenaeus vannamei*) fed high levels of soybean meal in various combinations. *Aquaculture*, 319(1-2), 141-149.
22. Steffens, W. (1989). *Principles of Fish Nutrition*. Ellis Horwood Limited. 1-384.
23. Tacon, A.G., & Metian, M. (2008). Global overview on the use of fish meal and fish oil in industrially compounded aquafeeds: Trends and future prospects. *Aquaculture*, 285(1-4), 146-158.
24. Worthington, C. (1991). *Worthington enzyme manual related Biochemical*. Freehold, New Jersey, USA. 1730.
25. Yaghoubi, M., Mozanzadeh, M.T., Marammazi, J.G., Safari, O., & Gisbert, E. (2016). Dietary replacement of fish meal by soy products (soybean meal and isolated soy protein) in silvery-black porgy juveniles (*Sparidentex hasta*). *Aquaculture*, 464, 50-59.
26. Yigit, M., Ergün, S., Türker, A., Harmantepe, B., & Erteken, A. (2010). Evaluation of soybean meal as a protein source and its effect on growth and nitrogen utilization of black sea turbot (*Psetta maeotica*) juveniles. *Journal of Marine Science and Technology*, 18(5), 682-688.
27. Yu, D., Gong, S., Yuan, Y., & Lin, Y. (2013). Effects of replacing fish meal with soybean meal on growth, body composition and digestive enzyme activities of juvenile Chinese sucker, *Myxocyprinus asiaticus*. *Aquaculture Nutrition*, 19(1), 84-90.
28. Zamani, A., Hajimoradloo, A., Madani, R., & Farhangi, M. (2009). Assessment of digestive enzymes activity during the fry development of the endangered Caspian brown trout *Salmo caspius*. *Journal of Fish Biology*, 75(4), 932-937.