

Soybean Flour - Poultry Meat Meal Blend as Dietary Protein Source in Practical Diets of *Oreochromis niloticus* and *Clarias gariepinus*

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Abstract

Two parallel experiments were conducted to evaluate the utilization of defatted and toasted soybean flour (SF) blended with poultry meat meal (PMM) as a dietary protein source in the practical diets of *Oreochromis niloticus* ($M \pm SE = 8.2 \pm 0.5$ g) and *Clarias gariepinus* ($M \pm SE = 19.4 \pm 1.8$ g) fingerlings. They were fed four isonitrogenous and isoenergetic diets containing blends of SF and PMM in ratios of 25:75, 50:50, 75:25 and the reference diet (100:00) for eight weeks, at 4% body weight per day.

A trend of difference in nutrient utilization was observed in both investigations as diet II (50:50) recorded better food conversion ratio, specific growth rate, and protein efficiency ratio than any other diet. These differed significantly with the diets ($P < 0.10$). This was probably as a result of a better and an ideal essential amino acid balance in the diet than any other one. There was increase in the level of sulphur containing amino acids (methionine + cystine) with increasing incorporation of PMM as blend with SF in all the diets except the reference diet (without PMM). This order was reversed for phenylalanine + tyrosine, and leucine that was in excess. Methionine deficiency and excessive leucine were ameliorated while phenylalanine + tyrosine level decreased.

Carcass composition differed significantly with respect to the diets only in *O. niloticus* ($P < 0.05$). Carcass lipid and ash increased with increasing levels of PMM.

Introduction

The quest for a suitable plant protein alternative to fish meal has been the focus of aquaculture for many years (Wee and Wang 1987; Ng and Wee 1989; Lim and Dominy 1990; Shiau et al. 1990). Generally, high dietary levels of plant proteins or complete substitution of animal proteins has resulted in poor growth and feed efficiency in fish (Jackson et al. 1982; Viola et al. 1983; Dabrowski et al. 1989; Lim 1992). This has been attributed to antinutritional factors or toxic substances; improper balance of essential nutrients such as amino acids, energy and minerals; high amounts of fiber and carbohydrate; decrease in palatability of the feed; and reduction of pellet quality especially water stability (Lim and Dominy 1989).

Of the plant proteins, soybean is the most widely used (Shiau et al. 1990) and it constitutes 30-40% of the feed for warmwater fishes (Lim and Dominy 1989). While there is projected 5% dwindling supply of fish meal between 1990 and 2000, there has been an astronomical 176% increase in the production level of soybean in the past 20 years with an additional 40% increase anticipated by the year 2000. In addition to its availability in most world markets, it is of good quality comparable to animal protein (Rumsey 1993). There is widespread use of soybean in aquaculture diets in view of its favorable economic and nutritional qualities. However, there is a need to resolve its inherent drawbacks aforementioned.

It has been observed that its nutrient imbalance (particularly amino acids) could be improved by the incorporation of low-cost animal protein such as poultry meat meal. Past experiments have established the successful utilization of poultry by-product meal in fish diets (Fowler 1981a, 1981b, 1982, 1991; Steffens 1994). An economic improvement in soybean meal-based diets for catfish could be achieved by the use of animal proteins which cost less than fish meal (Mohsen and Lovell 1990). Therefore, this study sought to investigate the nutritional value of poultry meat meal as blend with soybean flour in diets of *Oreochromis niloticus* and *Clarias gariepinus*

Materials and Methods

Fingerling *O. niloticus* ($M \pm SE = 8.2 \pm 0.5$ g) and *C. gariepinus* ($M \pm SE = 19.4 \pm 1.8$ g) were stocked in duplicate (10 fish per tank) in 50-l tanks and fed four diets containing soybean flour (SF) - poultry meat meal (PMM) blends, viz: reference diet (100:00), diet I (75:25), diet II (50:50) and diet III (25:75), at 4% body weight per day, twice daily at 0930 and 1700 hours for 8 weeks. Fishes were bulk weighed fortnightly and feeding rates adjusted accordingly. The system was cleaned regularly. Water flow rate was maintained at 2 l min^{-1} per tank. Water quality parameters were monitored fortnightly and recorded thus: temperature $26 \pm 0.0^\circ\text{C}$; dissolved oxygen (DO) $5.2 \pm 0.3 \text{ mgL}^{-1}$; pH 6.6 ± 0.1 ; $\text{NH}_3\text{-N}$ $0.3 \pm 0.1 \text{ mgL}^{-1}$; $\text{NO}_2\text{-N}$ $0.2 \pm 0.0 \text{ mgL}^{-1}$; $\text{NO}_3\text{-N}$ $12.5 \pm 2.5 \text{ mgL}^{-1}$; Ca-hardness $61.8 \pm 3.3 \text{ mgL}^{-1}$, and total hardness $77.3 \pm 1.5 \text{ mgL}^{-1}$.

The formulation and proximate composition of the experimental diets are shown in Table 1. The SF used was of commercial grade with a 73.63% protein solubility index as analyzed according to Araba and Dale (1990). High quality PMM was supplied by Chettles UK Ltd, Nottingham. A California Pellet Mill (model CL2) equipped with a steam conditioner was used to prepare 3 mm pellets which were subsequently dried overnight by convection at 60°C . After cooling, diets were packed in sealed black polythene bags and stored at -30°C until needed.

Chemical analyses of feedstuffs, diets and carcass (initial and final) were performed according to AOAC (1990). Eight specimens were used for the initial carcass analysis, while four specimens per tank were processed for the final carcass analysis per species. Moisture was by oven-drying at 105°C , protein by micro-Kjeldahl technique using Kjeltex Auto 1030 Analyser, lipid by

solvent extraction using Soxtec System 1043 Extractor, crude fiber with Fibertec System 1020 Hot Extractor, and ash by incinerating in a muffle furnace. An Alpha-Plus Amino Acid Analyser (LKB Bichrom Ltd, Cambridge) was used for amino acid analysis of the diets. Biological parameters measured included specific growth rate (SGR), food conversion ratio (FCR), protein efficiency ratio (PER) and apparent net protein utilization (ANPU) (Steffens 1989). Gross energy was calculated by using the following multiplier factors: carbohydrate, 4.1 kcalg⁻¹; protein, 5.4 kcalg⁻¹; and lipid, 9.5 kcalg⁻¹ (Jobling 1983).

Data were subjected to one-way Analysis of Variance (ANOVA) and multiple means comparison using the Turkey Test conventionally at 5% probability level and 10% when necessary to define a trend (Steele and Torrie 1960). Percentage data were transformed by arc-sine transformation (Zar 1984).

Table 1. Inclusion levels of ingredients and proximate composition of diets fed to *O. niloticus* and *C. gariepinus* for 56 days. Figures in parentheses are the blending ratios of soybean flour and poultry meat meal.

Ingredients	Reference diet (100:00)	Diet I (75:25)	Diet II (50:50)	Diet III (25:75)
Soybean flour	80.5	54.0	32.5	14.8
Poultry meat meal	-	18.0	32.5	44.5
Wheat flour	2.8	12.8	20.8	27.4
Soy oil	8.7	7.3	6.2	5.2
Vitamin premix ¹	2.0	2.0	2.0	2.0
Mineral premix ²	4.0	4.0	4.0	4.0
Binder (CMC) ³	2.0	2.0	2.0	2.0
Proximate analysis (% as fed)				
Moisture	7.6	6.3	5.7	5.3
Crude protein	38.9	38.1	38.8	38.3
Lipid	10.2	10.3	10.8	11.1
Crude fiber	4.7	3.8	2.8	2.6
Ash	8.4	9.5	10.8	11.1
Gross energy (kcalg ⁻¹)	4.5	4.5	4.5	4.5

¹ Vitamin premix providing the following vitamins (mgKg⁻¹ premix): vitamin A, 1,000; vitamin D, 4.0; vitamin E, 7,000; vitamin K, 1,500; vitamin C, 37,500; thiamine, 4,250; riboflavine, 3,000; pyridoxine, 1,250; pantothenic acid, 5,250; niacin, 12,500; biotin, 90; folic acid, 1,000; vitamin B₁₂, 1.25; choline, 74,050; inositol, 25,000.

² Mineral premix providing the following minerals (gKg⁻¹ premix): calcium orthophosphate, 727.8; magnesium sulphate, 127.5; sodium chloride, 60; potassium chloride, 50; iron sulphate, 25; zinc sulphate, 5.5; manganese sulphate, 2.5; copper sulphate, 0.8; cobalt sulphate, 0.5; calcium iodate, 0.3; chromic chloride, 0.1.

³ CMC=carboxymethyl cellulose

Results

Experiment 1 - *O. niloticus*

No significant differences were observed in the mean initial weights (8.2±0.5 g) and the mean final weights (23.1±1.5 g) of *O. niloticus* fed the four diets (P>0.05), but a trend of variation was observed in their SGR, FCR,

PER and ANPU significant only at $P=0.10$. Diet II was best utilized with the following values: SGR, $2.0\% \text{ day}^{-1}$; FCR, 1.5; and PER, 1.9. Conversely, the reference diet was poorest in nutrient utilization with the following values: FCR, 1.9; SGR, $1.6\% \text{ day}^{-1}$; PER, 1.5; and ANPU, 23.5%. Mortality was only recorded for the reference diet and this was insignificant ($P>0.05$) (Table 2). Diets I and III produced growth responses between those of diet II and the reference as depicted in Fig. 1.

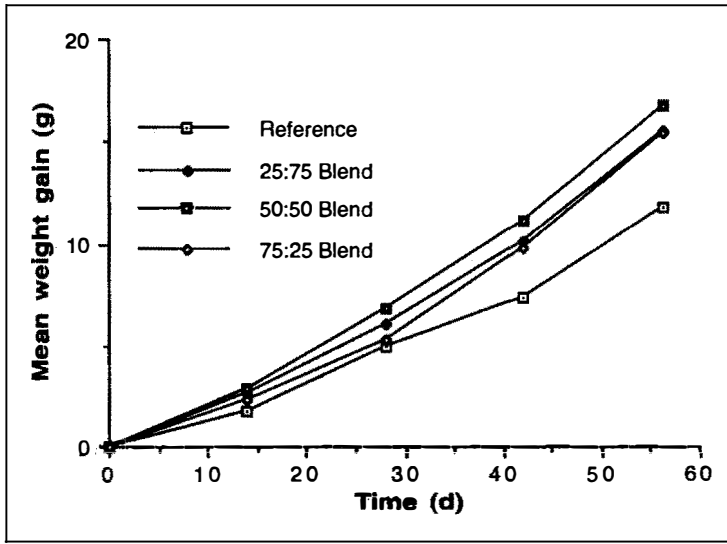


Fig. 1. Growth response of *O. niloticus* fed soybean flour-poultry meat meal blend-based diets for 56 days.

Table 2. Evaluation of soybean flour-poultry meat meal blend utilization in practical diets of *O. niloticus* fed for 56 days. Figures in parentheses are the blending ratios of soybean flour and poultry meat meal.

Parameter	Reference diet (100:00)	Diet I (75:25)	Diet II (50:50)	Diet III (25:75)	\pm S.E.M.
Mean initial weight (g)	8.3 ± 0.6^a	8.2 ± 0.6^a	8.2 ± 0.2^a	8.1 ± 0.5^a	± 0.5
Mean final weight (g)	20.0 ± 1.8^a	23.7 ± 0.1^a	25.0 ± 2.0^a	23.6 ± 1.1^a	± 1.5
SGR ($\% \text{ day}^{-1}$)	1.6^a	1.9^{ab}	2.00^b	1.9^{ab}	± 0.0
FCR	1.9 ± 0.1^b	1.6 ± 0.1^a	1.5 ± 0.1^a	1.6 ± 0.0^a	± 0.1
PER	1.5 ± 0.1^a	1.8 ± 0.2^b	1.9 ± 0.1^b	1.8 ± 0.0^b	± 0.1
ANPU (%)	23.5 ± 0.0^a	27.5 ± 0.1^{ab}	29.5 ± 0.0^b	30.0 ± 0.0^b	± 0.0
Mortality (%)	2.6 ± 2.6^a	0.0^a	0.0^a	0.0^a	± 0.7

Data in the same row carrying different superscripts show a trend of variation that is significant ($P<0.10$).

Table 3 shows the amino acid composition of the diets vis-a-vis their requirements in *O. niloticus* as established by Santiago and Lovell (1988). Methionine + cystine was observed to be the first limiting amino acid followed by phenylalanine + tyrosine, threonine and lysine, in that order. There was an excess of leucine in all the diets. Increasing PMM incorporation increased methionine and reduced leucine in the diets. Carcass composition of

fingerlings fed the four diets is shown in Table 4. Protein, lipid and ash varied significantly with the diets ($P < 0.05$). Protein content was highest (16.2%) in fingerlings fed diet III and lowest (15.4%) in fingerlings fed diet I. Fingerlings fed diet III had the highest lipid content of 8.2% and the highest ash content of 4.6%, while the lowest lipid and ash values of 5.6% and 3.6%, respectively, were recorded in fishes fed the reference diet. There was a general increase in carcass lipid and ash with increasing PMM in the diets. No morphological changes were observed in the fishes.

Table 3. Amino acid composition of soybean flour-poultry meat meal blend-based diets fed to fingerlings of *O. niloticus* for 56 days presented against its requirements. Figures in parentheses represent amino acid content in the diets expressed as percentages of their requirements.

Amino acid (% protein)	Reference diet	Diet I	Diet II	Diet III	Amino acid requirement*
Arginine	5.6(133.3)	5.3(126.2)	5.2(123.8)	5.0(119.0)	4.2
Histidine	2.1(123.5)	2.0(117.6)	1.8(105.9)	1.8(105.8)	1.7
Isoleucine	3.3(106.5)	3.1(100.0)	2.9(93.5)	2.8(90.3)	3.1
Leucine	5.9(174.6)	5.6(164.7)	5.6(164.7)	5.5(161.8)	3.4
Lysine	4.6(90.2)	5.0(98.0)	4.7(92.2)	5.1(100.00)	5.1
Methionine + cystine	1.1(34.4)	1.3(40.6)	1.4(43.8)	1.5(46.9)	3.2
Phenylalanine + tyrosine	5.2(94.5)	4.8(87.3)	4.5(81.8)	4.2(76.4)	5.5
Threonine	3.1(81.6)	3.2(84.2)	3.3(84.2)	3.4(89.5)	3.8
Valine	3.4(121.4)	3.4(121.4)	3.5(125.0)	3.7(132.1)	2.8

* Source: Santiago and Lovell (1988).

Table 4. Carcass composition of *O. niloticus* fed soybean flour-poultry meat meal blend-based practical diets for 56 days. Figures in parentheses are the blending ratios of soybean flour and poultry meat meal.

Diet	Moisture	Protein	Lipid	Ash
Reference(100:00)	74.1 ^a	15.9 ^{ab}	5.6 ^a	3.6 ^a
Diet I (75:25)	73.4 ^a	15.4 ^a	6.5 ^{ab}	4.2 ^{ab}
Diet II (50:50)	70.6 ^a	16.0 ^b	7.3 ^{ab}	4.5 ^b
Diet III (75:25)	71.7 ^a	16.2 ^b	8.2 ^b	4.6 ^b
Initial sample	70.5	15.3	8.7	3.4

Column figures with the same superscript are insignificantly different from each other ($P > 0.05$).

Experiment 2 - *C. gariepinus*

Table 5 depicts no significant differences observed in the mean initial weight (19.4 ± 1.8 g) and the mean final weight (69.2 ± 7.7 g) of *C. gariepinus* fed the four diets ($P > 0.05$), though at $P = 0.10$, a trend of growth and nutrient utilization was recorded. Best performance was observed in fishes fed diet II with values of SGR, $2.7\% \text{ day}^{-1}$; PER, 2.8; ANPU, 47.5%; and FCR, 1.0. The reverse was the case for fishes fed the reference diet as they performed poorly

with values of FCR, 1.6; SGR, 1.7% day⁻¹; PER, 1.8; and ANPU, 31.5%. Diet I had better FCR and PER than diet III except for ANPU. Insignificant mortality was recorded only in diets I and III ($P>0.05$). Diets I and III showed a performance intermediate of the reference diet and diet II as in Fig. 2.

The amino acid composition of the diets (Table 6) shows that methionine + cystine was the first limiting amino acid, followed by phenylalanine + tyrosine and lysine, in that order. There was excess of leucine in all the diets and increasing the level of PMM in the diets increased methionine + cystine but decreased phenylalanine + tyrosine and leucine. No significant difference was observed in the carcass proximate composition of *C. gariepinus* fed the diets ($P>0.05$). Morphologically, neither toxicity nor deficiency symptoms were observed.

Table 5. Evaluation of soybean flour-poultry meat meal blend utilization in practical diets of *C. gariepinus* fed for 56 days. Figures in parentheses are the blending ratios of soybean flour and poultry meat meal.

Parameter	Reference diet (100:00)	Diet I (75:25)	Diet II (50:50)	Diet III (25:75)	±S.E.M.
Mean initial weight (g)	19.6±2.9 ^a	19.8±0.6 ^a	20.3±1.5 ^a	17.8±1.5 ^a	±1.8
Mean final weight (g)	51.1±1.5 ^a	70.4±7.6 ^a	93.2±5.7 ^a	65.2±12.2 ^a	±7.7
SGR (% day ⁻¹)	1.7 ^a	2.3 ^{ab}	2.7 ^b	2.3 ^{ab}	±0.0
FCR	1.6±0.2 ^a	1.2±0.1 ^a	1.0±0.0 ^a	1.3±0.2 ^a	±0.1
PER	1.8±0.2 ^a	2.4±0.1 ^b	2.8±0.0 ^a	2.2±0.3 ^a	±0.2
ANPU (%)	31.5±0.1 ^a	40.0±0.0 ^a	47.5±0.0 ^a	40.4±0.4 ^a	±0.2
Mortality (%)	0.0 ^a	2.6 ^a	0.0 ^a	2.6 ^a	±0.00

Data in the same row with different superscripts show a trend of variation that is significant ($P<0.10$).

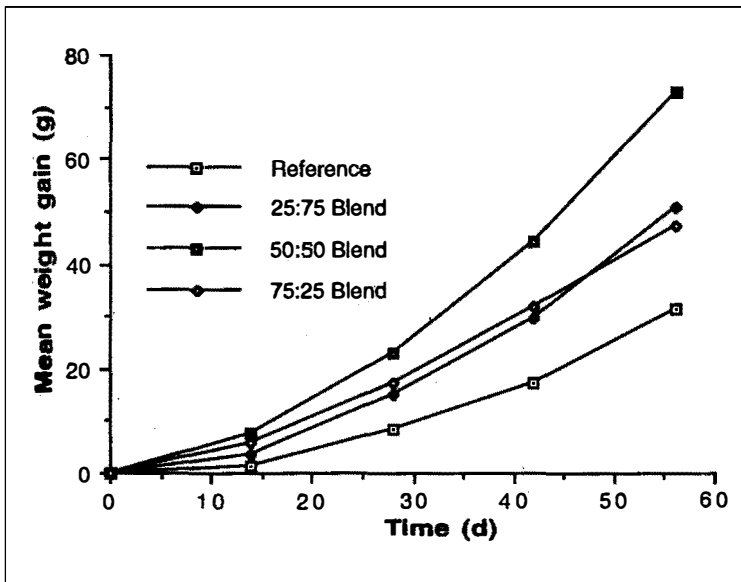


Fig. 2. Growth response of *C. gariepinus* fed soybean flour-poultry meat meal blend-based diets for 56 days.

Table 6. Amino acid composition of soybean flour-poultry meat meal blend-based diets fed to fingerlings of *C. gariepinus* for 56 days presented against its requirements. Figures in parentheses represent amino acid content in the diets expressed as percentages of their requirements.

Amino acid	Reference diet	Diet I	Diet II	Diet III	Requirement*
Arginine	5.6(130.2)	5.3(123.3)	5.2(120.9)	5.0(116.3)	4.3
Histidine	2.1(140.0)	2.0(133.3)	1.80(120.0)	1.80(120.0)	1.5
Isoleucine	3.3(126.9)	3.1(119.2)	2.9(111.2)	2.8(107.7)	2.6
Leucine	5.9(168.6)	5.6(160.0)	5.6(160.0)	5.5(157.1)	3.5
Lysine	4.6(90.2)	5.0(98.0)	4.7(92.2)	5.1(100.00)	5.1
Methionine + cystine	1.1(47.8)	1.3(56.5)	1.4(60.9)	1.5(65.2)	2.3
Phenylalanine + tyrosine	5.2(104.0)	4.8(96.0)	4.5(90.0)	4.2(84.0)	5.0
Threonine	3.1(155.0)	3.2(160.0)	3.2(160.0)	3.4(170.0)	2.0
Valine	3.4(113.3)	3.4(113.3)	3.5(116.7)	3.7(123.3)	3.0

* Requirements of a related species, the channel catfish. Source: National Research Council (1983).

Table 7. Carcass composition of *C. gariepinus* fed soybean flour-poultry meat meal blend-based practical diets for 56 days. Figures in parentheses are the blending ratios of soybean flour and poultry meat meal.

Diet	Moisture ^{NS}	Protein ^{NS}	Lipid ^{NS}	Ash ^{NS}
Reference(100:00)	69.9	17.3	8.0	3.5
Diet I (75:25)	71.3	16.9	7.4	3.7
Diet II (50:50)	68.8	17.1	9.2	3.6
Diet III (75:25)	69.6	17.9	9.1	3.9
Initial sample	68.0	16.4	6.2	3.1

NS = Not significant with respect to the diets ($P > 0.05$).

Discussion

The nutrient utilization of the SF:PMM blend-based diets by *O. niloticus* and *C. gariepinus* differed insignificantly at $P=0.05$. However, there was a trend of differences in the mean final weight, SGR, FCR, PER and ANPU of both fishes fed the diets, and these differences were significant ($P < 0.10$). The best mean final weight, SGR, PER and FCR values were recorded for fishes fed diet II in both experiments, suggesting the superiority of the 50:50 SF:PMM blend over the other blends for the culture of *O. niloticus* and *C. gariepinus*. The improved performance of diet II was probably due to improved essential amino acids (EAA) balance. Sadiku and Jauncey (in press) showed that SF is higher in all the EAA than PMM except threonine, valine, methionine + cystine and lysine. The incorporation of PMM obviously increased the level of methionine + cystine and reduced that of phenylalanine + tyrosine and leucine.

Methionine was identified as the first limiting amino acid as shown in Table 4, the risks of its deficiency could be reduced by the incorporation of PMM in the diets. Similarly, it reduced the level of leucine which when present in excess would be toxic to fish (Hughes et al. 1984; Robinson et al. 1984; Tacon 1992) and observed to have caused scoliosis, deformed opercula, scale

deformities, scale loss and spongiosis of epidermal cells in *Onycorhynchus mykiss* (Cho and Cowey 1991). Though there is paucity of information on deficiency and toxicity levels of these amino acids in *O. niloticus* and *C. gariepinus*, absence of structural symptoms of deficiency and toxicity in the two experiments indicated that their levels of deficiency and excessiveness were not critical to both fishes. However, excessive inclusion of PMM would result in phenylalanine + tyrosine deficiency.

Problems of lysine unavailability and sulphur containing amino acids destruction through toasting of SF (Gohl 1987; Snyder and Kwon 1987) could be ameliorated by the incorporation of PMM. A blend of SF and PMM as in diet II gave an improved amino acid balance. Higher levels of SF as in the reference diet and diet I increased the risk of methionine deficiency and leucine toxicity, while that of PMM as in diet III increased the risk of phenylalanine deficiency. This could militate against high inclusion levels of SF and PMM in ideal SF:PMM blend-based diets of *O. niloticus* and *C. gariepinus*.

The increasing order of carcass lipid and ash from the reference diet to diet III in *O. niloticus* with increased inclusion levels of PMM rather than SF was due to higher levels of lipid and ash in diets with high PMM (Sadiku and Jauncey, in press). This was not the trend in *C. gariepinus* which could tolerate higher inclusion levels of this in its diet than *O. niloticus*, as catfishes require more lipid than tilapia (National Research Council 1983). In fact, tilapia has been documented to perform better on a low lipid diet (Jauncey and Ross 1982; Jauncey 1993).

In conclusion, there was nutritional improvement of SF with the incorporation of animal protein. Whether this was equally economical as postulated by Fowler (1991) needs to be further investigated by evaluating the cost effectiveness of this study.

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