# Partial Replacement of Fish Meal with Some Alternative Protein Sources in the Diet of Tilapia Oreochromis niloticus (Linn)

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### Abstract

15 test diets were formulated, using soybean meal, groundnut cake, wheat bran, and blood meal. Their suitability to replace fish meal as a protein source in *Oreochromis niloticus* diet was assessed. Results showed that a proper combination of these alternative protein sources can provide between 42 and 45% of protein required by *Oreochromis niloticus* (33.32% dietary protein, dry matter). In such combination soybean meal, is capable of replacing up to 25% of fish meal as protein source. Blood meal exceeding 6%, groundnut cake beyond 10%, soybean meal above 20%, and wheat bran beyond 10% dietary inclusion level, retarded fish growth.

Keywords: Tilapia, Fishmeal, Alternative protein sources

#### Introduction

The high protein level required by fish for maximum growth has been established. Growth of fishes and utilisation of feed are reported to be optimal with proteins of animal origin, mainly fish meal characterised by being of high nutritive value (Dabrowska and Wojno, 1977). The high price of fish meal and shortage on the world markets have made it necessary to look for substitutes (Tacon and Jackson 1985; Webster et al. 1992). Unfortunately, attempts to replace the fish meal component of practical fish feeds with alternative protein sources have resulted in only variable success and have generally led to reduced feed efficiency and growth (Tacon and Jackson, 1985). Regardless of the limited success, the formulation of feeds containing high levels of plant proteins has become an important objective in fish nutrition research. Attempts to reach substitution levels of more than 50% of fish meal protein, by mixing two or more alternative protein sources, have been scarce although some of the results look promising (Jackson et al., 1982; Smith et al., 1988). The essential amino acid composition of alternative protein sources for fish are not comparable with that of fish meal. They lack the apparent chemical score on individual basis, to provide possible alternative to fish meal (De Silva and Anderson,1995). However, a combination of different alternative protein sources which may possess different limiting amino acids have been strongly suggested (Jackson et al., 1982; Tacon and Jackson, 1985). The objective of this study is to assess the suitability of soybean meal, groundnut cake, wheat bran, and blood meal to partially or fully replace fish meal in the diet of *Oreochromis niloticus* (Linn).

#### **Materials and Methods**

#### EXPERIMENTAL DIETS

15 diets were formulated to yield a protein content of 33.32% dry matter (dm) by calculation. The proximate composition of these diets are presented in Table 1.

| Ingredients              | Diet  |       |       |       |       |       |       |       |       |       |       |       |       |       |       |
|--------------------------|-------|-------|-------|-------|-------|-------|-------|-------|-------|-------|-------|-------|-------|-------|-------|
| %                        | 1     | 2     | 3     | 4     | 5     | 6     | 7     | 8     | 9     | 10    | 11    | 12    | 13    | 14    | 15    |
| Fish meal                | 43    | 26    | 5     | -     | 5     | -     | -     | 26    | -     | 15    | 8     | 24    | 24    | 15    | 10    |
| Soybean M.               | -     | 5     | 5     | -     | 5     | 25    | 25    | 18    | 60    | 25    | 20    | 20    | 20    | 32    | 40    |
| G.nut Cake               | -     | 10    | 15    | -     | 10    | -     | 10    | -     | 10    | -     | 10    | 10    | 10    | 10    | 10    |
| Blood meal               | -     | 5     | 10    | 33    | 20    | 20    | 15    | 5     | -     | 10    | 10    | 6     | 6     | 8     | 8     |
| Wheat bran               | 10    | 10    | 15    | 14    | 18    | 20    | 15    | 10    | 10    | 10    | 20    | -     | -     | -     | -     |
| Sunflower oil            |       | 7     | 6     | 10    | 7     | 6.4   | -     | 6     | -     | 5.2   | -     | 3.1   | 3.1   | 2.0   | 0.3   |
|                          | 10    |       |       |       |       |       |       |       |       |       |       |       |       |       |       |
| Fish oil                 | -     | -     | -     | -     | -     | -     | 4.5   | -     | -     | -     | -     | -     | -     | -     | -     |
| Vitamin Mix <sup>1</sup> | 4.7   | 4.7   | 4.7   | 4.7   | 4.7   | 4.7   | 4.7   | 4.7   | 4.7   | 4.7   | 4.7   | 4.7   | 4.7   | 4.7   | 4.7   |
| Potato Meal              | 32.3  | 32.3  | 29.3  | 38.3  | 30.3  | 23.9  | 20.8  | 30.3  | 15.3  | 30.1  | 27.3  | 32.2  | 32.2  | 28.3  | 27.0  |
| % Proximate              |       |       |       |       |       |       |       |       |       |       |       |       |       |       |       |
| composition              |       |       |       |       |       |       |       |       |       |       |       |       |       |       |       |
| Moisture                 | 16.58 | 17.92 | 19.91 | 19.96 | 8.14  | 15.29 | 13.88 | 13.36 | 12.55 | 16.04 | 19.77 | 5.91  | 5.91  | 3.54  | 4.82  |
| Protein                  | 35.94 | 30.38 | 28.98 | 37.94 | 30.99 | 34.22 | 29.76 | 33.90 | 29.52 | 34.28 | 30.24 | 32.96 | 32.96 | 34.99 | 35.66 |
| Fat                      | 16.04 | 14.39 | 13.40 | 11.44 | 12.42 | 13.84 | 14.50 | 14.34 | 16.91 | 12.81 | 7.41  | 8.67  | 8.67  | 6.48  | 4.68  |
| Energy                   |       |       |       |       |       |       |       |       |       |       |       |       |       |       |       |
| Content KJ/g             | 21.68 | 21.40 | 21.33 | 22.28 | 21.61 | 22.17 | 22.04 | 21.48 | 22.01 | 21.64 | 20.76 | -     | -     | -     | -     |

| Table 1: Formulation an | d proximate compositio | n (% drv matter) | of experimental diets |
|-------------------------|------------------------|------------------|-----------------------|
|                         |                        |                  |                       |

<sup>1</sup>Vitamin and Mineral mix (Spezialfutter Neuruppin - VM BM 55/13 Nr. 7310) supplied per 100g of dry feed : Vitamin A 15000 IU; Vitamin D3 2500 IU; Vitamin E 500mg; Vitamin K3 23mg; Vitamin B1 42mg; Vitamin B2 18mg; Vitamin B6 21mg Vitamin B12 59μg; Nicotinic acid 100mg; Biotin 544.65μg; Folic acid 13mg; Pantothenic acid 123mg, Inositol 1230mg; Vitamin C 66.7mg; Antioxidants (BHT) 121.87mg; Calcium 20.2%

33.32% dietary protein dm, is estimated as optimal for the requirement of *O. niloticus* (Linn) (Ogunji and Wirth in press). Fish meal, soybean meal, blood meal, groundnut cake and wheat bran at varied proportions were mixed in each diet. Cracked soybean seeds (Soja Schrot; obtained from Institute of Animal Nutrition, Leipzig, Germany) were used in diet 13, 14 and 15. The soybean was heated for 60 minutes at 105°C (Viola et al., 1983) in a heating oven to deactivate any trypsin inhibitor which is capable of interfering with protein digestion. This was intended to verifying the method and to compare its effectiveness with the industrially deactivated supplies used in most of the test diets. The heated soybean seed was then homogenised. All dry diet components including vitamin and mineral preparation were thoroughly mixed with sunflower oil. Water was added and the feed pressed into pellets of 1mm diameter. 15 fingerlings (initial weight  $\pm$  3g) were stocked in each of the 9 experimental tanks. Detailed experimental conditions and the analyses of fish and feed samples for protein, crude fat, ash and energy have been previously described (Ogunji and Wirth in press). High Performance Liquid Chromatography (HPLC) (Merck Hitachi) was used for amino acid analysis of feed and fish samples.

All statistical analyses were carried out by the Duncan multiple range method using SPSS for Windows (version 9). From the experimental data obtained, weight gain, specific growth rate (SGR) and food conversion ratio (FCR) were calculated.

Food conversion rate (FCR) = Food fed (g) × Live weight gain (g) Specific growth rate (SGR % d ) = 100 (ln  $W_2$  - ln  $W_1$ )/ $T_2$ - $T_1$  × 100  $W_2$  = Final weight of fish and  $W_1$  = Initial weight of fish.

 $T_1$  and  $T_2$  = time (day)

Protein to energy ratio (P/E ratio) was calculated as mg protein / kJ energy.

### Results

### CHEMICAL COMPOSITIONS OF THE FEED MIXTURE

The nutrient contents and amino acid composition of the diet ingredients are presented in Table 2. The amino acid contents of cracked soybean seed decreased after heat treatment. Protein content of the test diets calculated to be around 33.32% before formulation, ranged from 28.98 to 37.97% dry matter at proximate analysis. However, conclusions in this work were made based on data from dietary protein contents between 30.28% and 34.28%. The amino acid composition of test diets are presented in Table 3. Energy content of the test diets ranged from 20.78 and 22.68 kJ/g while the protein to energy ratios were between 14.57 and 17.03 mg/ kJ (Table 4).

## FEED PERFORMANCE

Growth data, food conversion ratio, protein to energy ratio and percentage mortality of fish groups fed different test diets are presented in Table 4. Fish group 1 (containing 43% fish meal) performed better than other fish groups. They gained an average weight of 19.51 g per fish in 8 weeks. The SGR and FCR were 3.46%/d and 1.11 respectively. Only 2.2% mortality was observed. Among the other diets formulated by partially or completely substituting fish meal, diet 2, 8 and 13 containing 26 and 24% fish meal performed better. The food conversion ratio recorded in fish group 2, 8 and 13 were not significantly different from fish group 1. The SGR of fish group 1 was not significantly different with fish group 8, but it differs with groups 2 and 13 (Table 4). The amino acid composition of diet 2, 8 and 13 are relatively similar but slightly lower than that of diet 1 (Table 3). Diet 1 with a higher proportion of fish meal was readily accepted and consumed by the fish. Diet 6 containing 20% blood meal and 25% soybean meal was least consumed by the fish (11.64 g per fish, Table 4). Diet 4 had the best amino acid composition when compared with other diets (Table 3), yet did not enhance a better fish performance. The SGR and FCR of this group were 1.11%/d and 5.78, respectively. The highest mortality of 66.7% was observed in the fish group fed diet 7.

### Discussion

After heat treatment amino acids in the cracked soybean seed (Soja schrot) decreased in concentration. Lysine, leucine and arginine among other amino acids were lost in the range of 11.45%, 12.98%, and 9.96% (dry matter) respectively (Table 2). This agrees with the result of Voila et

al. (1983) who observed that soybean meals heated at 105°C and 17% moisture for 60 and 120 minutes respectively lost about 10% lysine. In diet 9, where soybean meal provided the entire dietary protein, fish food intake was low, fish growth was reduced and food conversion became poor (Table 4). It has been observed that growth tends to reduce in fish fed diets with soybean meal replacing all the fish meal (Jackson et al., 1982; Webster et al., 1992). The amino acid composition (Table 3) reveals no great difference between diet 9 and 1 (which was formulated with 43% fish meal and resulted to the best performance of the fish). However, there may be a sub-optimal amino acid balance. The biological value of amino acids from soybean meal may be lower than indicated (Murray, 1982). Dabrowski et al. (1989) stated that amino acid availability, especially methionine, was reduced if soybean-meal protein was used in excess of 50% of the diet. It may be possible that diet 9 was deficient in phosphorous. Substitution of animal meals, which contain bones, by seed proteins creates phosphorous deficiency, which turns out to be the only really critical nutrient when fish meal is replaced by soybean meal (Viola et al., 1988). Watanabe et al., (1997) suggest that adding high percentages of soy products in fish diets can cause unpalatability and unacceptability leading to diminished growth. The texture and taste of test diets are bound to differ with increasing levels of incorporation of plant material which also affects the acceptability of the diets by fish (De Silva et al., 1989). In this study it was observed that when soybean meal constitutes more than 20% of the diet, food intake, weight gain and food conversion ratio were negatively affected (diet 6,7,9,10,11,14,15).

Diet 13 formulated with 33% blood meal (Table 1) resulted to reduced fish growth, low feed intake and poor food conversion ratio (Table 4). Blood meal used for diet formulation in this study contained 94.32% protein, dry matter, and had a better amino acid profile than other feed stuffs (Table 2). The amino acid composition of diet 4 seems better when compared with diet 1, but it was deficient in isoleucine and methionine (Table 3). It has been observed that blood meal protein is of low quality, has low digestibility, and the amino acid composition is rather biased being low in isoleucine and methionine (Close and Menke, 1986). Cullison (1979) noted that blood meal is not very palatable to most livestock and hence it is not popular as a protein supplement in livestock feeds. In this study also diet 4 was not well accepted. This may be due to the unpalatable nature of the blood meal component. At a high dietary inclusion of blood meal, poor performance of fish was observed (diet 3,4,5,6,7,11,12,14,15). A dietary blood meal inclusion rate, not exceeding 6% may therefore be recommended for *Oreochromis niloticus* (Linn). This is contrary to the result of Otubusin (1987) who realised a good performance of the same species with a feed containing 10% blood meal.

When 15% groundnut cake was included in diet 3, fish growth was relatively depressed. Inclusion of 5% fish meal in the diet did not improve growth. The reduced fish performance may be a result of inadequate protein content of the diet (28.98%), than the effect of groundnut cake. However, the amino acid profile of the groundnut cake used for diet formulation in this study was extremely poor (Table 2). Wu and Jan (1977) reported a very low specific growth rate of *Tilapia aurea* fed on an all groundnut protein diet. They associated the poor fish performance to the amino acid profile of groundnut. Jackson et al. (1982) reported a rapid growth decrease as the level of groundnut inclusion was increased in the diet of tilapia Oreochromis mossambicus; a problem blamed on the low methionine level in the sample used. Considering the results of this current study a groundnut cake dietary inclusion of about 10% is recommended. The reason for the high mortality recorded among fish group fed diet 7 (66.7%; Table 4) is unclear. However, the spoilage of the diet in the course of the experiment may have been the cause. Aflatoxin (a toxic component of groundnut), has been linked to increase mortality in animal feeding (Lovell, 1989).

From the foregoing, it has become evident that no particular feed stuff under investigation, apart from fish meal may solely provide all the necessary nutrients needed by *Orechromis niloticus* (Linn). This agrees with the observation of De Silva and Anderson (1995). Fish groups fed diet 2, 8 and 13 showed a better performance than other groups when compared with fish group 1. From these observations it appears that a proper combination of soybean meal, blood meal, groundnut cake and wheat bran can provide between 42 and 45% protein need of Orechromis niloticus (Linn). In this combination soybean alone, is capable of replacing up to 25% fishmeal in the diet. Jackson et al. (1982) tried to substitute different plant proteins for fish meal. At low levels of replacement (25%) growth rates were similar, at higher inclusions the performance was considerably reduced. Viola et al. (1986) showed that tilapia responded very well to feeds that contained plant proteins when phosphorus is supplemented in them. When substituting fish meal the amino acid compositions of the diets determine the performance of the fish at any dietary protein level. Attention should therefore be paid to the amino acid profile of alternative protein sources and the resulting test diets. More investigations are needed to further this direction of research. A lot of alternative protein sources are yet to be investigated and the preliminary results need to be harmonised. Results of this study have shown that alternative protein sources combined properly, are capable of substituting fish meal in the diets of Orechromis niloticus (Linn).

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