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RESEARCH PAPER

MIXTURES OF OILSEED MEALS AS DIETARY PROTEIN SOURCES IN DIETS OF JUVENILE NILE TILAPIA (*Oreochromis niloticus L.*)

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ABSTRACT
The suitability of different mixtures of soybean meal (SBM), cottonseed meal (CSM) and groundnut cake (GNC) as ingredients to replace fish meal in the diets of Nile tilapia (Oreochromis niloticus L.), was evaluated over a 56-day growth period. Nine isonitrogenous (320 g.kg^{-1}), isolipidic (100 g.kg^{-1}) and isoenergetic (18 KJ.g^{-1}) test diets were formulated in which different mixture combinations of SBM, CSM and GNC proteins replaced fish meal (FM) protein at levels of 50% and 75%. The control diet had FM as the sole protein source. Fish were fed at 6 - 4% body weight per day. The growth experiment was conducted in plastic tanks in a recirculation system. Each dietary treatment was in triplicate. Growth performance and feed utilization of fish fed with the oilseed meal mixtures indicated that up to 50% replacement could be more effective than a single source for the substitution of fish meal in tilapia diets. This was particularly evident with the diet containing equal proportions of all oilseed meals (EQ50). Combination of oilseed meals in different proportions was more effective than the single individual sources. This could be due to a compensatory effect which led to some reduction of antinutritional factors coupled with improved essential amino acid profile in the diet as a result of mixing.

Keywords: *Oilseed meals, Nile tilapia, protein sources, mixtures, growth performance*

INTRODUCTION

Attempts to partially or completely replace the fish meal component of fish feeds with alternative protein sources have resulted in variable success notably in reduced feed efficiency and fish growth at higher dietary inclusion levels (Tacon and Jackson, 1985; Jackson *et al.*, 1982). In most cases single plant protein sources were evaluated at various inclusion levels to substitute fish meal in the diet (Agbo *et al.*, 2011a; Agbo *et al.*, 2011b; Amisah *et al.*, 2009; Nyina-wamwiza *et al.*, 2007; El-Saidy and Gaber, 2002; Mbahinzireki *et al.*, 2001; El-Sayed, 1999). The majority of plant protein sources researched on by these authors was oilseed meals. Although, oilseed meals have high protein levels and favourable essential amino acid (EAA) profiles they are known to contain a variety of growth inhibiting antinutritional factors (Francis *et al*., 2001; NRC, 1993). When a higher level of plant protein is included, the antinutrients in the diets exceed the tolerance limit of the test animal and this often leads to reduced growth, feed utilization and mortalities in some cases. The use of different plant protein sources in combination could prevent high inclusion levels of any single antinutrient in the diet (Francis *et al*., 2001).

The essential amino acid compositions of alternative protein sources for fish are generally not comparable with that of fish meal. Chemical score data show that there is no single foodstuff that can serve as an alternative to fish meal (De Silva and Anderson, 1995). Therefore, combining different alternative protein sources which possess different limiting amino acids that could complement each other has been strongly recommended (Tacon and Jackson, 1985; Jackson *et al*., 1982).

Several researchers have reported comparatively better growth performance of fish fed diets containing different combinations of plant protein sources. Earlier studies by Olukunle (1982) showed that a mixture of groundnut, sunflower seed and sesame meals resulted in better growth of *O. mossambicus* than single meals. Similarly, Hossain (1988) also conducted similar work with common carp (*Cyprinus carpio*) and concluded that different plant protein sources in various combinations were more effective than protein sources derived from a single plant. Borgeson *et al*. (2006) found improved performance of *O. niloticus* fed with a diet containing mixtures of soybean and maize gluten meals as partial substitutes for fish meal protein. Attempts to reach substitution levels of more than 50% of the fish meal protein, by mixing two or more alternative protein sources is scanty in the literature although some of the results look promising (Soltan *et al*., 2008; Borgeson *et al*., 2006; Fontainhas-Fernandes *et al*., 1999; Jackson *et al*.,1982).

Information on the use of plant protein mixtures in Nile tilapia feeds is generally limited (Agbo *et al*., 2014; Agbo, 2008; El-Saidy and Gaber, 2003). The main objective of this study, therefore was to evaluate the quality and suitability of different mixtures of soybean meal (SBM), cottonseed meal (CSM) and groundnut cake (GNC) as alternative source of protein to replace fish meal in Nile tilapia diets focusing on growth performance, feed utilization, carcass quality and cost effectiveness of the diets.

MATERIALS AND METHODS

Experimental system and animals

This study was conducted at the tropical aquarium of the Institute of Aquaculture, University of Stirling, UK. Fish were reared in plastic tanks (30 L) in a recirculation system which was supplied with aerated water at a flow rate of 1 $L.min^{-1}$ with temperature maintained at 27 ± 1^{o}C. The fish were subjected to a constant photoperiod of 12 hours light/12 hours darkness. Water quality parameters measured weekly during the experiment were as follows (mean ± SD): temperature, 26.10 ± 0.44^{o}C; pH, 7.20 ± 0.16; ammonia, 0.06 ± 0.03 $mg.L^{-1}$; nitrite, 0.25 ± 0.0 $mg.L^{-1}$; nitrate, 20 ± 0.0 $mg.L^{-1}$ and dissolved oxygen, 7.20 ± 0.67 $mg.L^{-1}$. Twenty mixed-sex *O. niloticus* known as the “Red-Stirling strain” of mean weight 2.46 ± 0.12 g were stocked randomly into plastic tanks in triplicates per treatment. Initial and final weights of fish were individually obtained under anaesthesia with Benzocaine (50 $mg.L^{-1}$).

Diet formulation and preparation

The soybean meal (crude protein, 500.3 $g.kg^{-1}$), cottonseed meal (crude protein, 441.4 $g.kg^{-1}$), and groundnut cake (crude protein, 430.5 $g.kg^{-1}$) used in this study were obtained from commercial sources in Ghana while, fish meal (crude protein, 716.3 $g.kg^{-1}$) and wheat grain (crude protein, 95.2 $g.kg^{-1}$) were supplied by Ewos Ltd (Bathgate, UK).

Nine isonitrogenous (320 $g.kg^{-1}$ protein), isol-

Table 1: Specification of dietary protein levels (%) in experimental diets used in this study

Diet No.	Designation	Percentage of total protein contributed by various sources in the diets			
		Fish meal	Soybean meal	Cottonseed meal	Groundnut cake
1	Control	100.00	-	-	-
2	EQ50	50.00	16.67	16.67	16.67
3	SBM50	50.00	25.00	12.50	12.50
4	CSM50	50.00	12.50	25.00	12.50
5	GNC50	50.00	12.50	12.50	25.00
6	EQ75	25.00	25.00	25.00	25.00
7	SBM75	25.00	37.50	18.75	18.75
8	CSM75	25.00	18.75	37.50	18.75
9	GNC75	25.00	18.75	18.75	37.50

EQ50 = 50% equal contribution of protein from test ingredients to mixture; SBM50 = half of 50% contribution from soybean meal to mixture; CSM50 = half of 50% contribution from cottonseed meal to mixture; GNC50 = half of 50% contribution from groundnut cake to mixture; EQ75 = 75% equal contribution of protein from test ingredients to mixture; SBM75 = half of 75% contribution from soybean meal to mixture; CSM75 = half of 75% contribution from cottonseed meal to mixture; GNC75 = half of 75% contribution from groundnut cake to mixture

ipidic (100 $g.kg^{-1}$ lipid) and isoenergetic (18 $KJ.g^{-1}$) diets were formulated using mixtures of SBM, CSM, and GNC as protein sources for this experiment to satisfy the nutrient requirements of Nile tilapia (NRC, 1993). Composition of the different oilseed meal mixtures used for diet formulation is presented in Table 1 and designation of diets shown correspondingly. Fish meal was substituted with different mixtures and combinations of SBM, CSM and GNC at 50% and 75% of total protein. Diet formulation is presented in Table 2. Methods of preparation of experimental diets were the same as reported previously by Agbo *et al*. (2014). The diets were stored at -20^{o}C until fed. Fish were hand-fed three times a day (09:30 h, 13:00 h and 16:00 h) to apparent satiation and the experiment lasted eight weeks (56 days). At the end of the growth trial, faeces were collected for digestibility study using a modified settling column system similar to the Guelph system (Cho *et al.,* 1985) as described by Agbo *et al.* (2009).

Biochemical composition of diets and fish

Ingredients, diets, fish carcass and faeces were analysed in triplicates for proximate composition according to standard methods (AOAC, 1990) and chromic oxide of diets and faeces analysed using the method by Furukawa and Tsukahara (1966). Energy was determined using an Adiabatic Autobomb Calorimeter (Parr 6100, USA) with benzoic acid as a standard. Amino acid content was analysed using LKB 4151 Alpha-Plus Amino Acid Analyser (LKB Biochrom Ltd, UK). Phosphorus was measured following the method of Stirling (1985) using a spectrophotometer (Cecil Elegant Technology–Aquarius-P). Some antinutritional factors such as phytic acid, trypsin inhibitors and saponin were analysed following earlier methods used by Agbo *et al*. (2009).

Analysis of growth performance and feed utilization

Performance in growth and feed utilization were determined in terms of weight gain (WG), specific growth rate (SGR), feed intake (FI),

Table 2: Composition of diets ($g.kg^{-1}$ as-fed) fed to juvenile *Oreochromis niloticus* with varying inclusion levels of oilseed meal mixtures and relative prices

Ingredient	Diets Control 1	EQ50 2	SBM50 3	CSM50 4	GNC50 5	EQ75 6	SBM75 7	CSM75 8	GNC75 9	*Price of ingredients ($GH¢.kg^{-1}$)
FM	420.0	210.0	210.0	210.0	210.0	105.0	105.0	105.0	105.0	1.17
SBM	-	100.2	150.4	75.2	75.2	150.4	225.4	112.7	112.7	0.48
CSM	-	113.5	85.2	170.4	85.2	170.4	127.7	255.5	127.7	0.18
GNC	-	116.4	87.4	87.4	175.0	174.6	131.0	131.1	262.0	0.40
WG	203.0	206.0	205.5	206.0	205.2	209.0	209.4	208.0	208.8	0.10
SF oil	57.0	47.0	53.8	51.7	35.4	42.0	52.1	49.1	24.6	0.50
α- Cel.	30.0	15.9	17.5	12.0	19.5	10.0	10.9	02.6	14.0	0.05
Corn St.	205.0	106.0	105.2	102.3	109.5	53.6	53.5	51.0	60.2	0.10
CMC	20.0	20.0	20.0	20.0	20.0	20.0	20.0	20.0	20.0	0.25
Mpremix[1]	40.0	40.0	40.0	40.0	40.0	40.0	40.0	40.0	40.0	1.50
Vpremix[2]	20.0	20.0	20.0	20.0	20.0	20.0	20.0	20.0	20.0	1.45
Cr_2O_3	5.0	5.0	5.0	5.0	5.0	5.0	5.0	5.0	5.0	
Diet cost ($Gh¢.kg^{-1}$)	0.56	0.42	0.43	0.40	0.42	0.34	0.36	0.33	0.34	

FM = Fish meal, SBM = Soybean meal, CSM = Cottonseed meal, GNC = Groundnut cake, WG = Wheat grain, SF oil = Sunflower oil, α- Cel. = α- Cellulose, Corn St. = Corn starch, Mpremix = Mineral premix, Vpremix = Vitamin premix, Cr_2O_3 = Chromic oxide, CMC= Carboxymethyl cellulose (Sigma, C5013).

[1]*Contained (as g kg^{-1} of diet): $MgSO_4,7H_2O$, 20.40; NaCl, 8.00; KCl, 6.04; Fe $SO_4,7H_2O$, 4.00; $ZnSO_4,4H_2O$, 0.88; $MnSO_4,4H_2O$, 0.41; $CuSO_4,5H_2O$, 0.13; $CoSO_4,7H_2O$, 0.08; $CaIo_3,6H_2O$, 0.05; $CrCl_3,6H_2O$, 0.02 (according to Jauncey and Ross 1982).*

[2]*Contained (as mg kg^{-1} of diet): Thiamine (B_1), 85.00; Riboflavin (B_2), 60.00; Pyridoxine (B_6), 25.00; Pantothenic acid, 105.00; Inositol, 500.00; Biotin, 1.80; Folic acid, 20.00; Ethoxyquin, 4.00; Choline, 1481.00; Nicotinic acid (Niacin), 250.00; Cyanocobalamin (B_{12}), 0.03; Retinol palmitate (A), 20.00; Tocopherol acetate (E), 140.00; Ascorbic acid (C), 750.00; Menadione (K), 30.00; Cholecalciferol (D_3), 0.08 (according to Jauncey and Ross 1982).*

**Prices of ingredients in Ghana cedis (USD 1.00 = GH¢ 0.90, 2007 exchange rate).*

feed conversion ratio (FCR), protein efficiency ratio (PER), apparent net protein utilization (ANPU) and energy retention (ER) as follows:

WG (%) = (final body weight – initial body weight)/ initial body weight x 100

SGR (% day^{-1}) = 100 × [ln(final body weight) - ln(initial body weight)]/no. of days

FI (g) = Total feed intake per fish/no. of days,

FCR = feed intake/live weight gain

PER = live weight gain/crude protein intake

ANPU (%) = 100 × (final fish body protein (g) - initial fish body protein (g))/crude protein intake (g)

ER (%) = 100 × (final fish body energy - initial fish body energy)/gross energy intake

The apparent digestibility coefficients (ADC) for the nutrients of the diets were calculated as follows (Bureau *et al*., 1999):

ADC (%)=100×[1-(%nutrient in faeces/%

nutrient in feed)×(%marker in feed/marker in faeces)]

Digestible protein and energy were calculated as follows:

Digestible protein (DP, g.kg^{-1}) = dietary crude protein (g.kg^{-1}, dwb) x $ADC_{protein}$

Digestible energy (DE, kJ.g^{-1}) = gross energy

(kJ.g^{-1}, dwb) x ADC_{energy}

Where *dwb = dry body weight (g)*

Whole body composition was determined where whole fish body samples were analyzed for moisture, crude protein, crude lipid and ash and the results expressed as percentages of live weight before and after the experiment. Hepatosomatic index (HSI) was determined at the end of the experiment by randomly selecting 20 fish from each treatment and euthanized by overdose of benzocaine. The fish were then weighed, dissected and their livers removed and HSI determined as; *HSI = liver weight/ body weight x 100.*

Analysis of cost effectiveness of diets

Cost effectiveness of diets was assessed using Profit Index (PI) according to El-Sayed (1990) as follows; *PI = value of fish/cost of feeding.* The value of fish and cost of diets were calculated using market prices in Ghana cedis (GH¢) per kilogram.

Statistical analysis

Each experimental diet was fed to three groups of fish in a completely randomized design. Statistical analyses in this study were conducted using SPSS Statistical Package (Version 15.0, SPSS Inc., Chicago, IL). Data were subjected to one-way ANOVA and the Tukey's Multiple Comparison Test applied to evaluate differences between means at $P < 0.05$. All percentages were arcsine transformed before analysis (Zar, 1984).

RESULTS AND DISCUSSION

Biochemical composition of diets

Proximate composition, energy, phosphorous, antinutritional factors and essential amino acid (EAA) composition of experimental diets are presented in Table 3. Crude protein contents were similar and only varied slightly between the diets (323.8 – 334.9 g.kg^{-1}). Energy, crude lipid and NFE levels in all experimental diets were very similar. However, crude fibre content exhibited high variation and ranged between 25.4 g.kg^{-1} and 44.6 g.kg^{-1} where the control diet had the lowest and Diet 6 the highest. Diets 4, 6 and 8, which contained more CSM, had the highest levels of crude fibre. Phosphorous content of the control diet was highest (9.03 g.kg^{-1}), phytic acid ranged from 0.5 g.kg^{-1}– 12.5 g.kg^{-1}, TIs from 0.0 g.kg^{-1} – 3.6 g.kg^{-1}, saponin from 1.1 g.kg^{-1} – 4.7 g.kg^{-1} and gossypol from 0.0 g.kg^{-1} – 1.4 g.kg^{-1}. Generally, methionine + cystine were observed to be the first limiting EAA followed by threonine and lysine, in that order (Table 3). Increasing oilseed meal inclusion reduced the limiting amino acids and increased phenylalanine + tyrosine in the diets.

Growth performance

Growth performance of Nile tilapia fingerlings fed the experimental diets is presented as initial mean weight (IW), final mean weight (FW), weight gain (WG) and specific growth rate (SGR) in Table 4 and shown graphically in Fig. 1. It was observed that growth responses were significantly affected by both mixture composition and the inclusion level of oilseed meal protein right from the end of the second week of the feed trial. In general, growth rate decreased with increase in inclusion of oilseed meal protein mixtures.

Best overall growth response was obtained in tilapia fed the control diet (Figure 1). Percentage WG and SGR were highest for the control diet (704.24% and 3.72 %. day^{-1}, respectively) and lowest for Diet 6 (EQ75) containing 75% plant protein (equally contributed by SBM, CSM and GNC) which had WG of 322.35 % and SGR of 2.56 %.day$^{-1.}$ (Table 4). Weight

Table 3: Biochemical composition of diets fed to Nile tilapia (*Oreochromis niloticus)* in this study

Parameter	Diets Control 1	EQ50 2	SBM50 3	CSM50 4	GNC50 5	EQ75 6	SBM75 7	CSM75 8	GNC75 9
Proximate composition (g.kg^{-1} as-fed)									
DM	941.6	934.7	936.0	947.7	941.0	938.0	937.1	940.8	936.5
CP	323.8	324.0	330.6	328.6	330.4	330.4	331.7	329.0	334.9
CL	108.0	105.6	105.7	105.9	104.4	104.5	108.5	107.3	104.6
CF	25.4	37.1	34.1	40.2	34.6	44.6	35.2	42.1	33.9
Ash	96.9	87.4	85.9	87.8	84.1	81.1	80.3	85.3	81.5
NFE	387.6	380.6	379.7	385.2	387.4	377.4	381.4	377.1	381.7
Cr_2O_3	5.0	5.1	5.1	5.0	5.0	4.9	5.0	4.9	4.9
GE (kJ.g^{-1})	18.60	18.79	18.85	18.78	18.86	18.89	18.92	18.79	19.02
Phosphorous and anti-nutritional factors (g.kg^{-1})									
P	9.03	7.84	7.70	7.86	7.76	7.42	7.18	7.76	7.48
PA	0.5	7.6	7.1	8.5	7.1	11.1	10.5	12.5	10.4
TI	0.0	1.8	2.4	1.5	1.6	2.7	3.6	2.2	2.3
G	0.0	0.6	0.5	1.0	0.5	1.0	0.7	1.4	0.7
S	1.1	3.4	3.3	3.4	3.5	4.5	4.4	4.5	4.7
Estimated essential amino acid composition (% of dietary protein)									
Arginine	5.01	6.11	5.97	6.23	6.14	6.66	6.44	6.83	6.69
Histidine	1.99	2.18	2.20	2.20	2.15	2.28	2.30	2.31	2.23
Isoleucine	3.96	3.71	3.84	3.66	3.63	3.59	3.78	3.51	3.46
Leucine	6.41	6.08	6.24	6.03	5.98	5.92	6.15	5.84	5.77
Lysine	6.23	5.43	5.55	5.41	5.34	5.03	5.20	5.00	4.90
Methionine + Cystine	2.72	2.07	2.06	2.10	2.04	1.74	1.74	1.79	1.70
Phenylalanine + Tyrosine	5.26	5.90	5.96	5.89	5.85	6.21	6.30	6.20	6.14
Threonine	3.23	2.93	2.97	2.97	2.86	2.78	2.84	2.83	2.67
Valine	4.94	4.59	4.69	4.57	4.52	4.42	4.56	4.39	4.30

DM = dry matter, CP = crude protein, CL = crude lipid, CF = crude fibre, NFE = nitrogen free extract, Cr_2O_3 = Chromic oxide, GE = gross energy, P = phosphorous, PA = Phytic acic, TI = Trypsin inhibitor, G = Gossypol, S = Saponin

gains for all oilseed based diets were significantly lower ($P < 0.05$) than the control. SGR followed the same trend with the exception of Diet 2 (EQ50) containing 50% plant protein (equally contributed by SBM, CSM and GNC) which was not significantly different ($P > 0.05$) from the control (Table 4). Results of the present investigation showed that substitution of fish meal by various plant protein sources in different combinations resulted in improved growth performance compared to that of single plant proteins used at the same level in previous studies (Agbo *et al.*, 2011a; Agbo *et al.*, 2011b).

The weight gain of feed containing protein mixtures at 50% and 75% increased by an average of 184.98% and 164.99% respectively and specific growth rate by 0.70% and 0.89% respectively compared to that of single plant protein source in previous studies (Agbo *et al.*, 2011a; Agbo *et al.*, 2011b; Nyina-wamwiza *et al.*, 2007; El-Saidy and Gaber, 2002; Mbahinzireki *et al.*, 2001). The results obtained here

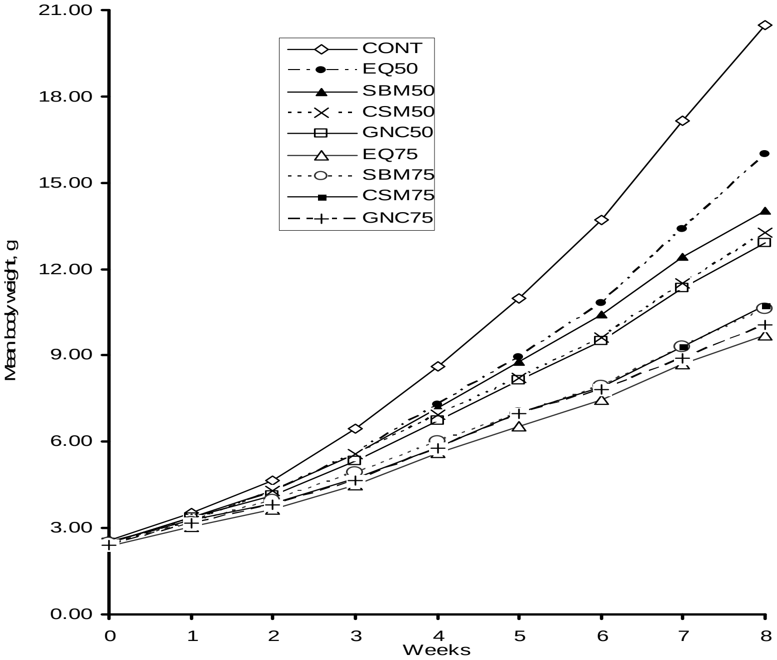

Fig. 1: Growth response of Nile tilapia (*Oreochromis niloticus*) fed diets with oilseed meal mixtures for eight (8) weeks

Table 4: Growth performance and survival of Nile tilapia (*Oreochromis niloticus*) fingerlings fed diets with oilseed meal mixtures for eight (8) weeks

Diets		Growth Performance				
		IW (g)	FW (g)	WG (%)	SGR (%.day^{-1})	S (%)
1	Control	2.55±0.14	20.49±1.91^{a}	704.24±55.69^{a}	3.72±0.12^{a}	100.00±0.00
2	EQ50	2.44±0.13	15.96±1.03^{b}	556.41±57.82^{b}	3.36±0.16^{ab}	93.33±11.54
3	SBM50	2.47±0.07	13.84±0.69^{bcd}	459.57±25.54^{bcd}	3.07±0.09^{bcd}	90.00±10.00
4	CSM50	2.42±0.05	14.30±2.07^{bc}	464.46±48.62^{bc}	3.08±0.16^{bc}	91.67±2.89
5	GNC50	2.50±0.17	12.87±1.02^{bcd}	414.78±23.92^{cde}	2.92±0.08^{bcde}	91.67±5.77
6	EQ75	2.35±0.05	9.95±1.32^{d}	322.35±53.37^{e}	2.56±0.24^{e}	95.00±5.00
7	SBM75	2.47±0.07	10.48±1.16^{cd}	324.19±35.92^{e}	2.58±0.15^{e}	88.33±5.77
8	CSM75	2.54±0.19	11.26±1.64^{cd}	334.51±31.64^{de}	2.62±0.13^{de}	90.00±8.66
9	GNC75	2.39±0.10	11.18±1.89^{cd}	352.37±57.55^{cde}	2.69±0.23^{cde}	90.00±5.00

IW = Initial weight, FW = Final weight, WG = Weight gain, SGR = Specific growth rate, S = Survival rate. Values are means ± SD of three replicates, and values within the same column with different letters are significantly different ($P < 0.05$).

are in agreement with those of Olukunle (1982) who observed better growth performance of *O. mossambicus* fed diets containing combinations of groundnut, sunflower seed and sesame meals compared with diets containing the single ingredients. The effectiveness of using various combinations of ingredients in fish feed has been reported by Tacon *et al.* (1984) who successfully reduced the fish meal level from 50% to 10% by using a mixture of soybean, meat and bone meal, brewers yeast, puffed maize and blood meal in the diet of tilapia without reducing the growth performance. Fontainhas-Fernandes *et al.* (1999) incorporated a mixture of extruded pea and defatted soybean meals, Borgeson *et al.* (2006) soybean and maize gluten meals and Agbo *et al.* (2014) soybean, cottonseed and groundnut meals into Nile tilapia diets and reported improved growth parameters. Hasan (1986) also reported better growth performance of carp fry fed diets containing different mixtures/combinations of linseed, groundnut, mustard and sesame meals.

In this study, growth performance in terms of WG and SGR of fish fed with the control diet were significantly higher than all the test diets. Among the oilseed-based diets tested, WG of Diet 2 was not significantly different from Diets 3 and 4 but significantly higher than diets 5-9 which were mostly 75% plant protein mixtures. SGR followed a similar trend, however, that of Diet 2 was not significantly different from Diets 3, 4 and 5. Indeed, results from this study indicate that poor growth could be attributed to high levels of phytic acid, trypsin inhibitors and gossypol in the diets with 75% plant mixtures (Table 3).

Results from the study also indicated that, Diets 2-5 at 50% substitution of oilseed meal mixtures generally had slightly improved levels of methionine and threonine compared to those in previous studies by Agbo *et al.* (2011a) and Agbo *et al.* (2011b) where similar oilseed meals were used at the same level. Diets at 75% oilseed meal substitution also followed a similar trend in EAA profile. This seems to support the view of Jackson *et al.* (1982) and Tacon and Jackson (1985) who advocated the use of a combination of different plant protein sources as a means of compensating for EAA deficiency in tilapia diets.

Feed utilization

Feed efficiency and utilization data are presented in Table 5. The control diet ranked highest in efficiency of feed utilization expressed in FRC. However, the FCR (2.07) of the control diet was not significantly different ($P < 0.05$) from that of Diets 2-4 (FCR = 2.52 – 2.89) (Table 5). Diet 6 ranked lowest in efficiency of feed utilization in terms of FCR (3.09). However, this FCR was not statistically different from the other diets except Diets 1 and 2. These values agree with those of El-Saidy and Gaber (2003) and Borgeson *et al.* (2006). Feed intake ranged between 26.75g and 36.83g at the end of the experiment. Oilseed meal inclusion at 75% led to a significantly lower feed intake compared to the control diet. Feed intakes were similar to those reported by Agbo *et al.* (2014) and El-Saidy and Gaber (2003).

Protein utilization efficiency decreased as oilseed meal inclusion increased with the control diet having the highest PER (1.51) and ANPU (22.63) and Diet 6 having the lowest PER (0.79) and ANPU (12.08). PER was significantly ($P < 0.05$) lower for Diets 3 - 8 than the control and Diet 2. ANPU and energy utilization followed exactly the same trend as PER with the exception of Diet 4 which was not different from the control (Table 5).

The study revealed that protein utilization indices (PER, ANPU) and ER in fish fed with the oilseed meal mixtures were significantly different (Table 5) from the control with the exception of Diet 2 in case of PER, Diets 2 and 4 in case of ANPU and ER, which were not different from the control. Within the oilseed meal based diets these parameters were not significantly different with the exception of Diet 6 and 7. Generally, Diet 2 had similar ($P < 0.05$) growth performance (SGR) and feed utilization

Table 5: Feed utilization and profit index of Nile tilapia (*Oreochromis niloticus)* fingerlings fed diets with oilseed meal mixtures for eight weeks

Diets		Parameters					
		FCR	FI (g)	PER	ANPU (%)	ER (%)	PI
1	Control	2.07±0.26[a]	36.83±0.90[a]	1.51±0.19[a]	22.63±2.83[a]	17.10±2.14[a]	1.73
2	EQ50	2.52±0.23[ab]	33.98±2.04[ab]	1.23±0.11[ab]	17.62±1.63[ab]	13.37±1.24[abc]	1.91
3	SBM50	2.89±0.27[abc]	32.71±1.20[ab]	1.05±0.10[bc]	16.30±1.46[bc]	12.48±1.12[bc]	1.63
4	CSM50	2.80±0.20[abc]	31.80±2.39[abc]	1.07±0.10[bc]	17.39±1.50[ab]	14.17±1.03[ab]	1.75
5	GNC50	3.17±0.13[bc]	32.83±2.14[ab]	0.95±0.04[bc]	15.03±0.65[bc]	11.62±0.50[bc]	1.51
6	EQ75	3.90±0.55[c]	29.20±2.60[bc]	0.79±0.11[c]	12.08±1.63[c]	6.61±0.97[d]	1.51
7	SBM75	3.64±0.68[c]	28.67±1.41[bc]	0.85±0.16[c]	13.36±2.39[bc]	10.25±1.83[c]	1.56
8	CSM75	3.18±0.47[bc]	26.75±1.80[c]	0.97±0.14[bc]	14.25±2.07[bc]	11.01±1.60[bc]	1.95
9	GNC75	3.55±0.44[bc]	29.45±2.05[bc]	0.85±0.10[c]	13.49±1.52[bc]	10.72±1.20[bc]	1.66

FRC = Feed conversion ratio, FI = Feed intake, PER = Protein efficiency ratio, ANPU = Apparent net protein utilization, ER = Energy retention and PI = Profit index. Values are means ± SD of three replicates, and values within the same column with different letters are significantly different (P < 0.05)
The prices of ingredients used in this study are presented in Table 1. Price of tilapia is 2.00 GH¢kg^{-1} (USD1.00 = GH¢ 0.90, 2007 exchange rate). Only the costs of feed were considered, other costs were considered to be constant

(PER, ANPU and ER) compared to the control diet which outperformed Diets 3 – 9 in growth and feed utilization. This suggests the superiority of Diet 2 over the other plant derived mixtures for *O. niloticus.* The improved performance of Diet 2 was probably due to improved EAA balance in the study. Mixing the oilseed meals slightly increased the level of methionine + cystine and reduced that of phenylalanine + tyrosine and leucine but they were still above the requirements for tilapia (Table 3). Methionine + cystine were observed to be the first limiting amino acid followed by threonine and lysine, in that order. Increasing the oilseed meal inclusion reduced the limiting amino acids and increased phenylalanine + tyrosine in the diets as was similarly observed by Hossain (1988) and Sadiku and Jauncey (1995). Methionine was identified as the first limiting amino acid in diets with single oilseed meals but its level was slightly improved by the incorporation of SBM which had a higher level of this amino acid than the other oilseed meals (Agbo, 2008).

Although the different oilseed meals complemented each other in terms of amino acid balance in this study, SBM contributed more because of its superior amino acid profile. There was also a reduction in levels of individual antinutritional factors especially TIs and gossypol contents of Diets at 75% inclusion levels (Table 3) particularly for SBM and CSM diets which contained higher levels of TIs and gossypol respectively when they were used individually as protein sources in previous studies (Agbo *et al.*, 2011a; Agbo *et al.*, 2011b). However, PA increased in the mixtures especially at 50% inclusion as compared to previous studies (Agbo *et al.*, 2011a; Agbo *et al.*, 2011b).

Apparent nutrient digestibility

Apparent nutrient digestibility values are presented in Table 6. The control diet had the highest (89.88%) apparent protein digestibility (APD) and Diet 8 the lowest (87.41%). Generally, APD decreased slightly as plant protein inclusion increased. Apparent energy, dry matter and phosphorous digestibilities followed

Table 6: Apparent digestibility coefficients (%) of protein, lipid, dry matter, energy, phosphorus and digestible protein and energy (g.kg^{-1} and kJ.g^{-1} respectively, dry weight basis) in test diets for Nile tilapia (*Oreochromis niloticus*)

Parameters	Diets Control 1	EQ50 2	SBM50 3	CSM50 4	GNC50 5	EQ75 6	SBM75 7	CSM75 8	GNC75 9
DM	81.79	80.60	80.92	78.13	81.17	79.43	78.56	75.31	76.90
CP	89.88	89.08	89.66	87.62	89.75	88.84	88.46	87.41	89.51
CL	96.75	98.85	98.02	97.02	96.91	96.91	96.58	96.07	93.36
GE	83.88	82.74	83.27	80.78	83.34	82.01	81.17	78.10	79.27
P	75.89	73.11	74.48	67.08	72.94	70.53	72.36	64.91	70.53
DP	309.1	308.8	316.7	303.8	315.1	312.9	313.1	305.7	320.1
DE	15.60	15.55	15.70	15.17	15.72	15.49	15.36	14.68	15.08

DM = dry matter, CP = crude protein, CL = crude lipid, CF = crude fibre, GE = gross energy, P = phosphorous, DP = digestible protein, DE = digestible energy

similar trends to APD. Overall, nutrient digestibility for all diets was high and digestible protein and energy varied only slightly among the diets (Table 6).

Apparent protein digestibility (APD) of the diets fed to fish in this experiment was slightly higher than that of single protein source used by Agbo *et al.*, (2011a) and Agbo *et al.*, (2011b) indicating slight APD improvement through mixing of plant protein sources. APD was similar for all diets up to 75% replacement of the FM protein compared with that of the control diet, even though percentage weight gain was significantly lower for the 50% and 75% replacement groups. APD obtained in this study is higher (87.41 – 89.88%) than the values reported by El-Saidy and Gaber (2003) (80.30% - 85.40%) and Hossain *et al.* (1992) (81.44%) for tilapia.

Whole body composition

The chemical composition of whole fish body before and after the study is given in Table 7. All fish displayed a change in whole body composition (compared with the initial whole body composition at the start of the experiment), which consisted mainly of a decrease in percentage moisture and a corresponding increase in total lipid content. The protein content of fish increased in all dietary treatments compared with the initial sample. Lipid content was significantly higher, especially among diets with higher inclusion of oilseed meal mixtures, however, ash was the direct opposite. Hepatosomatic Index (HSI) values did not show any particular trend relating to diet treatment but, the control had the highest value.

Whole body composition was little affected by dietary treatments. Total crude protein, moisture content (MC) and gross energy (GE) contents of whole body of Nile tilapia were not influenced by dietary treatments because there was no significant difference among these parameters (Table 7). A similar trend was observed by Agbo *et al.* (2014) and El-Saidy and Gaber (2003) in Nile tilapia; Regost *et al.* (1999) in turbot; Pongmaneerat *et al.* (1993) in carp and Moyano *et al.* (1992) in rainbow trout. Ash content was significantly higher for the control diet and Diet 4 (CSM50) compared to the other diets particularly with higher levels of plant protein (Table 7). However, diets with

Table 7: Whole body proximate composition (% wet weight), energy and hepatosomatic index of Nile tilapia (*Oreochromis niloticus*) fed diets with oilseed meal mixtures

Diets		Parameters					
		MC	**CP**	**CL**	**Ash**	**GE**	**HSI**
1	Control	72.55±0.71	14.87±0.37	7.81±0.22[c]	3.93±0.14[a]	6.37±0.04[a]	3.11±0.40[a]
2	EQ50	73.97±0.92	14.25±0.58	7.62±0.26[c]	3.35±0.08[bc]	6.36±0.25[a]	2.49±0.50[b]
3	SBM50	72.64±0.47	15.19±0.26	7.99±0.13[abc]	3.42±0.04[bc]	6.68±0.14[a]	2.51±0.32[b]
4	CSM50	70.63±1.41	15.75±0.73	9.03±0.42[a]	3.67±0.18[ab]	7.30±0.40[a]	2.74±0.50[ab]
5	GNC50	72.27±1.46	15.37±0.81	8.25±0.41[abc]	3.28±0.20[bc]	6.79±0.50[a]	2.54±0.48[b]
6	EQ75	73.04±0.91	15.02±0.44	7.91±0.25[bc]	3.05±0.11[c]	5.11±0.25[b]	2.65±0.43[ab]
7	SBM75	72.26±1.46	15.32±0.81	8.22±0.40[abc]	3.23±0.17[bc]	6.92±0.17[a]	2.92±0.47[ab]
8	CSM75	73.64±1.61	14.52±0.79	7.81±0.49[c]	3.08±0.20[c]	6.45±0.53[a]	2.02±0.47[c]
9	GNC75	72.03±1.29	15.06±0.62	8.86±0.49[ab]	3.36±0.17[bc]	6.80±0.26[a]	1.89±0.40[c]
	IWBC	74.06	13.88	7.76	3.27	6.16	

MC = moisture content, CP = crude protein, CL = crude lipid, CF = crude fibre, GE = gross energy, HSI = Hepatosomatic index, IWBC = initial whole body composition of fish. Values are means ±SD of three replicates, and values within the same column with different letters are significantly different (P < 0.05).

higher levels of plant proteins produced higher lipid and lower moisture contents as also observed by Nyina-wamwiza *et al*. (2007) for the African catfish (*Clarias gariepinus).*

Cost-effectiveness of diets

The cost of the diets reduced with increase in inclusion levels of oilseed meal mixtures (Table 2). Results of cost analysis expressed as profit index (PI) of diets used in this experiment are presented in Table 5. The PI of Diets EQ50, CSM50 and CSM75 were higher than that of the control diet. The PIs of the remaining diets were lower than that of the control. In this experiment, it was also observed that the culture periods using the oilseed meal based diets would be longer than the control due to their lower SGRs.

The economics of feed production indicated that the costs of the diets were minimised by replacing fish meal with the oilseed meal mixtures (Table 2 and 5). From the results it was observed that the diet with equal contributions of oilseed meal in the mixture (Diet 2) and diets with higher proportions of CSM in the mixture (Diet 4 and 8) were the diets which were more profitable than the control diet. This compares with the results reported by Agbo (2008) where CSM based diets were most profitable possibly because CSM was the cheapest oilseed meal and had a fairly good growth performance. A similar investigation by Olvera-Novoa *et al.* (2002) suggested that it is possible to replace up to 65% of animal protein in *O. mossambicus* fry diets using a mixture of plant proteins (alfalfa leaf protein concentrate, soybean and torula yeast) without adverse effects on fish growth and profitability. However, another study by El-Saidy and Gaber (2003) evaluating a mixture of SBM, CSM and sunflower meal for Nile tilapia showed that plant protein mixtures were more profitable than the fish meal based diet even at 100% replacement. Moreover, Coyle *et al.* (2004) indicated that efficient and economical tilapia growth can be obtained by feeding diets without fish meal using a combination of distillery by-products, meat and bone meal and SBM.

CONCLUSIONS

Results from the study demonstrated that the use of different plant protein sources in various combinations to enhance protein profile of Nile tilapia diets could be more effective than using a single plant protein source for substitution of fish meal in the diet of this commercially important fish species. Careful selection and use of different plant protein sources in various combinations can be a means of compensating for essential amino acid deficiency in any single protein source and also prevent a high inclusion level of any single antinutritional factor in the diet. In the present study, though Diets EQ50, CSM50 and CSM75 had PIs higher than the control, Diet EQ50 has the best prospects for Nile tilapia culture based on observed growth performance, nutrient utilization and economic benefit when compared to the other diets evaluated in this study.

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