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This is of particular interest to policy makers, scholars, international agencies and private organisations involved in the tisheries industry.

A COMPARATIVE ASSESSMENT OF GROWTH PERFORMANCE, HAEMATOLOGY AND SERUM BIOCHEMISTRY OF HYBRID CATFISH FED NON-CONVENTIONAL PLANT PROTEINS IN HOMESTEAD CONCRETE TANKS

BY

Oyin Olukunle¹, V.O. Taiwo² and J.O. Adejinmi²

ABSTRACT

An 84-day experiment was carried out to evaluate the potentials of three non-conventional plant proteins – groundnut cake (GNC), soybean cake (SBC) and sesame seed cake (SSC) singly and combined in the diets of hybrid catfish grown in homestead concrete tanks.

Triplicate groups of 30 juveniles average weight of $26.9 \pm 0.18g$ were fed four diets which were formulated to contain 30% crude protein with 25% of the GNC (diet 1) component replaced by SBC (diet 2), SSC (diet 3) and a combination of the three in diet 4. All the diets contained an equal amount of animal protein (25.43g). At the end of 84 days of feeding, growth parameters, feed and carcass proximate analyses, mortality, haematology and serum biochemical parameters were measured.

The results showed that the diets affected the growth of the juveniles significantly (P<0.05). The percentage weight gain of the fish fed diet 1 was 204% of the initial weight, while those fed diets 2, 3 and 4 were 405%, 465% and 445% respectively. Protein efficiency ratio (PER) was significantly higher ((P<0.05) in diets 2, 3 and 4 than in diet 1, with diet 4 having the highest PER. The percent specific growth rate (SGR) recorded for each diet was not significantly different (P>0.05) from each other. Percentage mortality were 22.2%, 13.3%, 5.6% and 3.3% in fish fed diets 3, 4, 2 and 1, respectively.

There were no significant variations in haematological parameters of fish fed all the various diets, except for those on diet 3 which exhibited lymphopenia and heterophilia (P<0.05). Similarly, there were no significant variations in serum biochemical parameters, except for hyperfibrinogenaemia, hypoglycaemia and significantly increased AST activities (P<0.05) in fish fed diets 3 and 4. There were significant increases in the serum levels of cholesterol and triglyceride in fish fed diets 2 (P<0.05), 3 and 4 (P<0.01).

The findings in this study showed that even though SBC and SSC inclusions in fish diet resulted in better growth performance than GNC inclusion, the relatively high levels of crude fat in these plant proteins, especially SSC, may in fact be injurious to fish by causing fat deposition in tissues, fatty hepatic and renal damage, artherosclerosis and coronary heart disease both of which will predispose the fish to stress, innocuous diseases and mortality.

Key words: Growth performance, Haematology, Serum biochemistry, Hybrid catfish, Non-conventional plant protein

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INTRODUCTION

Catfish, especially *Clarias* species is acceptable to the generality of Nigerians except for a small fraction of the populace who will not eat the fish for religious reasons. However, catfish is very expensive for most Nigerians, as it costs approximately ¥200 (\$1.90 or £1.43) per kilogram (Olukunle, 1996). The average cost of production could be brought lower if cost-effective non-conventional plant protein supplementation in diet is achieved and between 15-20% of the population such as retirees, housewives and the unemployed high school graduates could be involved in production of catfish in homestead ponds or any available space where water is not a constraint.

By the time the average family protein needs is satisfied, and the ease of handling catfish production is mastered, the incentive to produce for commercial purpose will be aroused. The problem of availability of tablesize catfish all year round will be reduced and prices will be lowered.

Clarias gariepinus and *Heterobranchus longifilis*, the parent stock, and their hybrid have carnivorous tendencies, hence they require high proteinaceous feeds (40 – 45% crude protein) for faster growth. Hybrid catfish on the other hand adapts better to artificial feeding than the parent stock, hence it grows faster and uniformly on waste products. The observed growth patterns in *Clarias* and *Heterobranchus* are unequal mixtures of shooters and smaller sizes in harvested catfish ponds. This observation is a result of the cannibalistic tendencies reported by catfish growers especially where feeding is sporadic. Most fish farmers cannot afford expensive artificial feeds based on fishmeal, hence the need to use cheap local alternatives that can be found in non-conventional plant protein sources.

Security on fish farms has been a continuous problem in the last three decades in Nigeria. Itinerary poachers have reduced many enthusiastic fish farmers to penury. Hence, many fish ponds have been abandoned, mostly with huge capital losses. The idea of growing fish in homestead ponds where a reasonable level of safety is assured will be a welcomed solution. Therefore, the aim of the present study was to assess comparatively three different plant protein sources – groundnut, soybean and sesame seed cakes – individually and in a combination of the three in mass production of hybrid catfish cheaply and economically with the hope of alleviating poverty and protein malnutrition in the ambit of the home.

MATERIALS AND METHODS

Four concrete homestead ponds of $2m \times 3m \times 1.5m$ dimension (capable of holding $9m^3$ of water) used are located at the University of Ibadan. Three hundred and sixty hybrid catfish juveniles of an average weight of $26.9 \pm 0.18g$ were purchased from a reputable private commercial fish farm situated in Ibadan, Oyo State, Nigeria. Thirty fingerlings were randomly stocked in each of three hapa nets of $1m^3$ dimensions and suspended into each of the four ponds $\frac{3}{4}$ -filled with fresh "rested" tap water. The fish were conditioned for 24 hours during which no food was offered. Fresh tap water was added to the holding tanks every other day through inlets. The water quality parameters were maintained at acceptable optimal levels for tropical fish (FAO, 1987).

Four isonitrogenous (30% crude protein) and isocaloric (29.81 \pm 0.19kcal/g) experimental diets were compounded as recommended by Omar (1996) for *C. gariepinus* grown in concrete tanks. All the diets contained 25.43g/100g fish meal as the animal protein source and 37.08g/100g plant proteins as either groundnut cake (GNC) (diet 1), soybean cake (diet 2), sesame seed cake (diet 3) and an equal mixture of SBC and SSC (diet 4). The composition of the diets is shown in Table 1. The ingredients were purchased locally, milled in a fabricated hand-milling machine and sieved to a particle size of 0.05mm. The sesame seed was dehulled, debittered, pre-pressed and oil-extracted before milling as described by Olukunle (1996). Diet components were thoroughly mixed and pelleted using a Moulineux kitchen mincer, and oven dried at 50°C for 6 hours. Samples of the four diets were analyzed for proximate composition for moisture, crude protein, lipid, ash and crude fibre contents as described by A.O.A.C. (1990). The fish were fed twice daily at 5% body weight (Khwuanjai et al., 1996) for 84 days. Batch weighing of fish in each hapa net was done every fortnight and weight of feeds adjusted accordingly. Proximate careass analysis was carried out on five randomly selected fish per hapa net before the commencement (initial) and on the 84th day of the feeding trial as previously described for the diets. Mortality in each hapa net was monitored and recorded every other day.

At the end of the 84-day feeding trial, pooled blood was obtained from the caudal peduncles of five randomly selected fish per hapa net (that is 15 fish per tank), half into clean plain vacutainer tubes and half into tubes containing lithium heparing anticoagulant. Haematological studies were carried out immediately using the anticoagulated blood, while the clotted blood samples were centrifuged at 1,200 G for 5 minutes at 29.5°C, and the serum stored at -25°C until used for biochemical analysis. Haematology and serum biochemical studies were carried out as described by Taiwo and Anosa (1995), Fadina et al. (1999) and Oyelese et al. (1999). The

parameters determined include packed cell volume (PCV), haemoglobin (Hb) concentration, red blood cell (RBC) counts, total and differential white blood cell (WBC) counts, total protein (TP), albumin (ALB), globulin (GLOB), fibrinogen, serum concentrations of Na⁺, K⁺, Cl⁻, HCO₃⁻, urea, creatinine, cholesterol, triglyceride, alanine aminotransferase (ALT), asparigine aminotransferase (AST) and glucose. Mean corpuscular volume (MCV), mean corpuscular haemoglobin concentration (MCHC) and albumin:globulin ratio (AGR) were calculated (Jain, 1986).

All data obtained per fish tank were pooled and subjected to analysis of variance (SAS, 1987) and the means separated for significant variations from the control feed (diet 1) using Duncan's multiple range test (Duncan, 1959) at 95% and 99% confidence intervals (P<0.05; P<0.01).

RESULTS

Tables 1 and 2 show the composition and proximate analysis of the four diets used in this experiment. From the proximate analysis, it was apparent that crude protein levels were slightly but insignificantly higher (P>0.05) in diets 1 and 2 than those of diets 3 and 4 (Table 2). However, there was a significantly higher (P<0.05; P<0.01) level of ether extract (crude fat) in diets 2,3 and 4, and especially highest in diets 3 which contains 12.5% plant protein supplementation with SSC.

			Diets	
Ingredients	1	2	3	4
Fishmeal	25.43	25.43	25.43	25.43
Corn meal	22.25	22.25	22.25	22.25
Groundnut cake	37.08	27.81	27.81	27.80
Soybean cake		9.27	2	4.64
Sesame seed cake		- -	9.27	4.64
Animal oil	2.50	2.50	2.50	2.50
Plant oil	2.50	2.50	2.50	2.50
Brewer's waste	5.00	5.00	5.00	5.00
Bone meal	2.00	2.00	2.00	2.00
Vitamin mix	2.00	2.00	2.00	2.00
Salt	0.25	0.25	0.25	0.25
Binder	1.00	1.00	1.00	1.00

Table 1: Composition of test diets fed hybrid Clarias (g/100g diet)

Table 2: Proximate composition of test diets fed hybrid Clarias (% dry matter)

		Dicts	
1	2	3	4
10.80	9.44	8.93	9.36
34.44	34.88	31.16	32.27
1.93	6.64	13.33	10.20
12.15	12.52	12.60	11.00
6.45	7.08	7.56	8.03
34.23	29.44	26.42	29.14
	1 10.80 34.44 1.93 12.15 6.45 34.23	1 2 10.80 9.44 34.44 34.88 1.93 6.64 12.15 12.52 6.45 7.08 34.23 29.44	Diets 1 2 3 10.80 9.44 8.93 34.44 34.88 31.16 1.93 6.64 13.33 12.15 12.52 12.60 6.45 7.08 7.56 34.23 29.44 26.42

On growth performance characteristics, Table 3 shows that fish feed with diets 2, 3 and 4 grew much faster (P<0.05; P<0.01) than those fed with diet 1 by a factor of at least 100%. Diet 3 recorded the highest daily and total weight gains of 1.48+/-0.02g and 465.0g or 124.2% and 126.9%, respectively of those fish fed diet 1 (Table 3). However, the fish fed diets 1 and 4 had the highest and lowest FCR (1:3.2 and 1:1.71), respectively. PER values were significantly higher (P<0.05) in diets 4, 2 and 3 (in that order) than in diet 1. Proximate carcass compositions of fish fed test diets for 84 days (Table 4) reveal that while moisture contents did not differ significantly (P>0.05) within the experimental fish, those fed diets 2 and 4 had the highest (P<0.05) levels of erude protein, there was no significant variation (P>0.05) in ash contents, except those on diet 3 which had a

significantly higher (P<0.05) ash content than all the others. Also, significantly higher (P<0.01) levels of crude fat were detected in fish fed diets 2, 3 and 4 compared to those fed diet 1. Crude fibre levels were also very high (P<0.05) in fish fed diets 2, 3 and 4 (Table 4). Mortality records (Table 5) show that 22.2% and 13.3% of fish fed diets 3 and 4, respectively died between the 6th and 12th weeks of the feeding trial. These are significantly higher (P<0.05) than 5.6% and 3.3% mortality rates recorded for fish fed diets 2 and 1, respectively.

			Diets		
Parameters		1	2	3	4
Initial weight (g)	_	27.3±0.5	26.3±0.6	26.7±0.3	26.6±0.4
Final weight (g)		83.3±1.1	130.0±2.3*	150.6±2.1**	145.3±2.9**
Total weight gain (g)		56.0±0.5	103.5±1.1*	124.3±1.7**	118.7±2.9**
Daily weight gain (g)		0.66±0.02	1.24±0.03*	1.48±0.02**	1.41±0.03**
% weight gain		204.90	405.13*	465.00**	445.00**
Total feed intake (g)		534.20	589.60*	738.10**	492.96
S.G.R. (%)		0.021	0.024	0.025	0.025
F.C.R.		1:3.2	1:1.84*	1:1.99*	1:1.71*
P.E.R.		0.10	0.18*	0.17*	0.24**

Table 3: Growth performance of hybrid Clarias fed test diets

S.G.R. = Specific growth rate; F.C.R. = Feed conversion ratio;

P.E.R. = Protein efficiency ratio

*,** Data significantly different from Diet 1 at P<0.05 and P<0.01, respectively.

Table 4: Proximate carcass composition (% dry matter) of hybrid Clarias before and after 84 days of feeding with test diets

		_			
Parameter	Initial	1	2	3	4
Moisture content	9.88	8.83	9.86	11.60	8.99
Crude protein	59.32	69.58*	65.58*	57.71	69.69*
Ether extract	6.12	5.12	12.34**	11.63**	10.20**
Ash	13.40	15.48	10.89	18.10*	10.24
Crude fibre	0.3400	0.9885*	0.9963*	0.9643*	0.8774*

*.** Data significantly different from initial values at P<0.05 and P<0.01, respectively.

Table 5: Proximate careass composition (% dry matter) of hybrid *Clarias* before and after 84 days of feeding with test diets

Diets					
Week	T	2	3	4	
4		-	-	-	
6		1	2	1	
8		1	3	1	
10	2	1	8	6	
12	1	2	7	4	
		•			
Total	3(3.3%)	5(5.6%)	20(22.2%)	12(13.3%)	

Hacmatology and serum biochemistry of the test fish are shown in Tables 6 and 7, respectively. There were no significant changes (P>0.05) in red blood cell and white blood cell parameters of the fish fed all the four experimental diets, except for relative lymphopenia and heterophilia (P<0.05; P<0.01) exhibited by fish fed diet 3 (Table 6). Similar results were obtained for most serum biochemical parameters. However, fish fed diets 3 and 4 showed hyperfibrinogenaemia, hypoglycaemia (P<0.01) and increased levels of serum AST activity (P<0.05) than those fed diets 1 and 2 (Table 7). Similarly, hypercholesterolaemia and hypertriglyceridaemia (P<0.05; P<0.01) were exhibited by hybrid Clarias fingerlings fed diets 2, 3 and 4.

	Ι	Diets	
1	2	3	4
31.0±0.7	30.5±0.4	30.0±0.7	31.5±0.5
2.9±0.2	2.7±0.3	2.6±0.3	2.9±0.3
8.3±0.1	8.1±0.1	7.9±0.2	8.3±0.2
108.7±7.5	115.2±5.7	117.6±8.1	108.7±1.6
26.8±0.4	26.4±0.2	26.2±0.7	26.2±0.2
15.2±2.5	13.7±1.2	13.0±1.3	15.7±2.2
12.4±2.3	10.8±2.1	9.5±0.3	12.5±0.9
1.9±0.2	2.2±0.1	2.8±0.2	2.3±0.3
0.3±0.04	0.3±0.02	0.3±0.00	0.4±0.03
0.6±0.01	0.4±0.03	0.4±0.02	0.5±0.02
	1 31.0±0.7 2.9±0.2 8.3±0.1 108.7±7.5 26.8±0.4 15.2±2.5 12.4±2.3 1.9±0.2 0.3±0.04 0.6±0.01	I 2 31.0±0.7 30.5±0.4 2.9±0.2 2.7±0.3 8.3±0.1 8.1±0.1 108.7±7.5 115.2±5.7 26.8±0.4 26.4±0.2 15.2±2.5 13.7±1.2 12.4±2.3 10.8±2.1 1.9±0.2 2.2±0.1 0.3±0.04 0.3±0.02 0.6±0.01 0.4±0.03	$\begin{array}{c cccccc} & \text{Diets} \\ 1 & 2 & 3 \\ \hline 31.0\pm0.7 & 30.5\pm0.4 & 30.0\pm0.7 \\ 2.9\pm0.2 & 2.7\pm0.3 & 2.6\pm0.3 \\ 8.3\pm0.1 & 8.1\pm0.1 & 7.9\pm0.2 \\ 108.7\pm7.5 & 115.2\pm5.7 & 117.6\pm8.1 \\ 26.8\pm0.4 & 26.4\pm0.2 & 26.2\pm0.7 \\ 15.2\pm2.5 & 13.7\pm1.2 & 13.0\pm1.3 \\ 12.4\pm2.3 & 10.8\pm2.1 & 9.5\pm0.3 \\ 1.9\pm0.2 & 2.2\pm0.1 & 2.8\pm0.2 \\ 0.3\pm0.04 & 0.3\pm0.02 & 0.3\pm0.00 \\ 0.6\pm0.01 & 0.4\pm0.03 & 0.4\pm0.02 \\ \hline \end{array}$

Table 6: Haematology (mean ± S.E.M.) of hybrid Clarias fed test diets

* ** Data significantly different from Dict 1 at P<0.05 and P<0.01, respectively.

Table 7: Serum biochemistry (mean ± S.E.M.) of hybrid Clarias fed test diets

			Diets	
Parameters	1	2	3	4
Total protein (g/dl)	8.5±0.1	8.3±0.1	8.4±0.2	8.4±0.1
Albumin (g/dl)	4.3±0.2	4.3±0.1	4.2±0.2	4.3±0.2
Globulin (g/dl)	4.2±0.2	4.0±0.2	4.2±0.1	4.1±0.2
A:G ratio	1.02±0.01	1.05±0.03	1.00±0.01	1.04±0.02
Fibrinogen (g/dl)	0.37±0.03	0.39±0.05	0.55±0.02*	0.50±0.03*
Glucose (mg/dl)	99.5±0,4	98.9±0.6	78.5±3.3*	82.3±2.6*
Na+ (mMol/l)	141.0±0.7	141.0±1.4	141.2±0.0	141.5±0.4
Cl- (mMol/l)	104.0±0.7	102.5±1.3	104.0±0.7	105.0±2.2
K+ (mMol/l)	4.5±0.7	4.2±0.0	4.4±0.7	4.5±0.5
HCO3- (mMol/l)	20.5±0.4	21.5±0.3	20.5±0.4	21.5±0.3
ALT (IU/I)	4.4±0.1	4.1±0.2	4.5±0.2	4.2±0.2
AST (IU/I)	8.3±0.2	8.3±0.1	12.1±0.4*	10.6±1.1*
Urca (mg/dl)	9.5±0.4	10.5±0.4	9.0±0.7	9.5±0.4
Creatinine (mg/dl)	1.0±0.2	1.2±0.0	1.1±0.0	1.2±0.0
Cholesterol (mg/dl)	109.5±0.4	143.5±3.2*	172.6±8.6**	159.3±7.4**
Triglyceride (mg/dl)	75.5±0.7	91.5±0.3*	102.5±1.8**	102.9±0.4**

*** Data significantly different from Dict 1 at P<0.05 and P<0.01, respectively.

DISCUSSION

Plant proteins are currently being used to substitute and augment animal proteins to reduce the cost of fish feed. One veritable source of plant proteins in livestock diets, groundnut cake (GNC), has been in use for over 40 years. However, in Nigeria, the production of GNC is on the decline, human rate of its consumption has risen astronomically because of population explosion; hence the cost of GNC has increased to the extent that its main use in formulating livestock diets has become uneconomical. Soybean cake (SBC) is gradually replacing GNC

in livestock diets because of its availability and lower cost. Utilization of sesame seed cake (SSC) in fish feed is more recent. SSC contains a higher level of methionine than GNC and SBC (Altschul, 1958). Research has shown that the inclusion of SSC in fish diets should not be higher than 25% of the total protein requirement (Olukunle, 1996). However, the level of SSC to be included in a fish diet to give optimum growth performance and good health is yet to be determined.

From the specific growth rate (SGR), the addition of a mixture of plant protein sources improved the growth performance of fish fed diets 2, 3 and 4. This is similar to the observation of Jackson et al. (1982). Diet 4 in addition had an improved protein efficiency ratio (PER) of 0.24 over diets 1, 2 and 3. The high PER in diet 4 is an indicator of the efficiency of protein utilization in fish fed this diet. However, the feed conversion ratio (FCR) of diet 4 was the lowest and this suggests that this diet was well accepted by the fish and that the conversion of protein to fish flesh was the best of all the diets.

The highest percentage daily and total weight gains observed in fish fed diet 3 (1.48 and 465%) were not supported by a corresponding high deposition of fish flesh in the carcass. Rather, a high percentage of ash (18.1%) suggesting high deposition of minerals and/or the development of dense bones was observed in fish fed diet 3. Diet 4 supported the highest deposition of flesh in fish with almost 70% crude protein in the fish carcass compared to about 58% in diet 3, a figure lower than the initial crude protein composition of the fish (59.32%). These findings support the view of Jackson et al. (1982) that a mixture of feed ingredients with different limiting amino acids is advisable for use in fish feeds. The findings of Pag (1956) and Olukunle (1996) further corroborate the results of this experiment that a diet with several protein sources gives better protein deposition in fish carcass than a single protein source.

The mortality pattern of fish fed the test diets shows that more deaths occurred in fish groups fed diets containing SSC, that is diet 3 (22.2%) and in diet 4 (13.3%) containing 12.5% and 6.25%, SSC, respectively. Moreover higher death rates were recorded in fish fed diets 3 and 4 between weeks 8 and 12, suggesting a form of stress, which may be attributable to SSC inclusion in the diets. In fact, both haematology and serum biochemistry data of fish fed diets 3 and 4, supported the above assertion that SSC may induce stress which are clinically characterized by lymphopenia, heterophilia and hypoglycaemia in animals (Jain, 1986; Kaneko, 1989). It should be noted that diets containing SSC have very high levels of crude fat, the excess in diet of which is stored in the body as fat in muscle, liver and kidneys leading to easily rupturable fatty liver and kidneys (Robbins et al., 1984) and in serum as triglycerides and cholesterol (Kaneko, 1989). Hyperlipidaemia depicted an increased serum levels of cholesterol and triglycerides in fish fed diets 3 and 4 containing SSC may precipitate artherosclerosis and coronary heart disease (kaneko, 1989) that may in fact be responsible for stress induced mortalities observed in these groups of fish compared to those fed diets without SSC. This clearly shows that SSC, even at a rate of 6.25% of plant protein inclusion, may still be quite far from the level that will ensure good growth performance and health in fish. More importantly, since the ultimate aim of producing fish using these cheap, non-conventional plant protein sources is for human consumption, the risk of very high fat intake from fish by man may be direct transfer of the same stress factors - hyperlipidaemia, artherosclerosis, fatty liver and kidney and coronary heart disease to man and the attendant public health and economic implications.

In conclusion, the findings in this study has corroborated earlier reports that diets with several protein sources give better protein deposition in fish carcass than a single protein source. They also show that GNC, SBC and probably lower levels of SSC inclusion (that is, less than 6.25%) as admixture will give very good growth performance characteristics and ensure good health and productivity in homestead fish farming with lower risks of poaching, economic animal protein food security and overall economic empowerment of housewives, pensioners and low scale fish farmers. It also noted that the use of very high inclusion levels of SSC might be injurious to both fish and man.

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