EVALUATION OF DIFFERENTLY HEATED SOYBEAN MEALS ON THE GROWTH AND NUTRIENT UTILIZATION OF THE AFRICAN CATFISH *CLARIAS GARIEPINUS* (BURCHELL)

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Abstract

This study was conducted to assess the growth and nutrient utilization of the African catfish, *Clarias gariepinus*, fed differently heated soybean meals. Four hundred and eighty male and female catfish (approximately 182 ± 10 g for females and 208 ± 5 g for males) were distributed into groups of 30 fish (sex ratio 1:1). The fish were stocked, in duplicates for each dietary treatment into 16 labelled hapas. Seven approximately iso-nitrogenous (31% crude protein) and iso-caloric diets (3.5 kcal g^{-1}) diets prepared from raw soybean and soybean autoclaved at 116 °C and 1.2 kg/cm^2 pressure) for 5, 10, 15, 20, 25 and 30 minutes marked D0, D5, D10 D15, D20, D25 and D30 were fed to the fish for 84 days. A fishmeal-based diet (DFM) served as the control diet. The male and female broodfish fed DFM diet had the highest mean weight gain (MWG) (198 ± 23 g; 224 ± 5 g) and percentage weight gain (PWG) (95%; 102%) respectively. The MWG, PWG, feed conversion ratio (FCR), specific growth rate (SGR), apparent net protein utilization (ANPU), hepato-somatic index (HSI) and condition factor (K_f) of the fish fed diets DFM and D25 were significantly different ($p<0.05$) from those fed the other diets. The female broodfish had higher weight increases and were in healthier condition than the male broodfish irrespective of the dietary treatment. The study showed that soybean autoclaved at 116 °C (1.2 kg/cm^2 pressure) for 25 minutes (D25) was found to be the best substitute for fishmeal in diets for the culture of *Clarias gariepinus* brooders.

Keywords: *Clarias gariepinus*, soybean, weight gain, growth, conversion, condition factor.

1. Introduction

Fishmeal constitutes 40-60% of the total cost of aquaculture feed production. It is scarce and expensive particularly in tropical Africa (Keembiyehetty and Gatlin III, 1997). Thus, reduction in fishmeal component of fish diets ultimately reduces feed costs and minimize competition with other users (FAO, 2000). In recent times fish feed researchers and manufacturers have been trying to reduce the cost of fish feed production by reducing or replacing fishmeal component of feeds with locally available oilseed plant proteins and their by-products (Carter and Hauler, 2000).

Soybean has received considerable attention worldwide as one of the most promising sources of plant proteins for use as a substitute for fishmeal because it is highly nutritious and readily available (Lim and Akiyama, 1992). The suitability of soybean in feed manufacture was attributed to a number of factors. Among vegetable protein sources, it has one of the best amino acid profile (rich in essential amino acids except methionine, cysteines and tryptophan). It is highly digestible and has low cholesterol (Ranhotra and Anderson, 1989; Solomon et al., 1996). Despite these advantages however, the presence of anti-nutritional factors and the low level of the sulphur-amino acid in soybean meal (compared with animal protein sources) have been identified as limitations to its use as a substitute for fishmeal in fish diet formulation (Lim and Akiyama, 1992). Feeding raw or improperly heated soybean to animals has been shown to cause growth depression and other associated disorders (Spinelli, 1980). When adequately heated however, the nutritive value of soybean improves (Lovell, 1979).

The present study investigated the growth and nutrient utilization of the African catfish, Clarias gariepinus (Burchell) broodstocks fed diets prepared with differently heated soybean meals as the primary protein source.

2. Materials and methods

Portions of full-fatted, dried soybean seeds (cultiver samsoy 2, TGX 636-02D) were broken into small granules by the hammer mill, winnowed and then autoclaved at 116°C (pressure 1.2 kg/cm^2) for 5, 10, 15, 20, 25 or 30 minutes respectively. One portion of the soybean seeds was left un-autoclaved. These differently autoclaved soybean portions were sun-dried and milled separately.

The raw and the variously autoclaved soybean portions were used separately along with other feedstuff (Table 1) to compound seven approximately iso-nitrogenous (31%) and iso-caloric (3.5 kcal g^{-1}) diets marked D0, D5, D10, D15, D20, D25, D30 (Table 2). Fishmeal was used as the sole protein source for the control diet (DFM). These

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Table 1: Mean proximate composition (%) (± S.E.)* and the gross energy content (kcal g^{-1}) of the ingredient providers used in formulating the experimental diets.

	Fishmeal	Yellow maize	Brewery dry grain	D0''	D5"	D10"	D15"	D20"	D25"	D30"
Crude Protein	51.0 ± 1.1	7.9 ± 1.9	24.4 ± 2.3	40.2 ± 3.2	40.0 ± 4.1	40.3 ± 4.5	40.5 ± 6.2	41.3 ± 5.0	42.9 ± 4.2	40.1 ± 5.0
Moisture	5.5 ± 1.3	8.6 ± 2.0	7.8 ± 1.2	8.2 ± 3.0	5.9 ± 2.1	5.8 ± 1.2	2.3 ± 1.9	2.6 ± 1.1	4.2 ± 2.1	5.2 ± 1.5
Lipid	16.6 ± 2.0	2.0 ± 0.2	3.4 ± 0.3	13.7 ± 0.5	14.2 ± 1.1	14.2 ± 1.2	15.7 ± 1.4	15.9 ± 2.1	16.2 ± 2.0	14.8 ± 1.9
Ash	25.5 ± 1.2	1.4 ± 0.1	7.5 ± 1.4	6.6 ± 1.1	6.8 ± 1.1	7.7 ± 2.1	7.5 ± 1.5	7.0 ± 2.4	7.8 ± 7.8	8.0 ± 2.4
Crude fibre	0.0	8.0 ± 1.1	10.3 ± 1.3	9.8 ± 3.2	8.2 ± 1.6	5.8 ± 1.1	5.7 ± 0.5	5.1 ± 0.5	6.7 ± 1.7	5.0 ± 1.6
**NFE	1.4 ± 0.8	72.1 ± 2.4	46.5 ± 3.8	21.5 ± 1.4	24.9 ± 3.1	26.2 ± 2.1	28.3 ± 5.2	28.1 ± 4.8	22.2 ± 6.5	26.9 ± 1.8
Gross energy	4.6 ± 0.4	4.4 ± 0.5	3.1 ± 0.8	3.3 ± 0.2	3.4 ± 0.5	3.2 ± 0.8	3.4 ± 0.5	3.5 ± 0.8	3.6 ± 0.5	3.3 ± 0.4

*Mean and Standard error $(\overline{X}(SE))^*$, D0"; D5"; D10"; D15"; D20"; D25"; D30" represents the duration of heating of soybean

** NFE = Nitrogen-free Extract = 100 – (Crude protein + Crude fibre + Lipid content + Moisture content + Ash)

❖Values with the same superscript in each column are not significantly different from each other ($p>0.05$)

ingredients were properly mixed together and pelleted to a particulate size of 8mm. The diets were kept in labeled polythene bags and stored in a freezer at -10°C until feeding trial commenced.

A year old, matured male and female C. gariepinus fish (approximately 182 ± 10 g for females and 208 ± 5 g for males) were obtained from a commercial hatchery in Osun State of Nigeria. They were reared in rectangular, cot-like hapas (4mx3mx1m) made from fine mesh nylon mosquito nets placed in concrete tanks (6mx5mx1.5m) already filled with raw water from the Opa Reservoir, Obafemi Awolowo University, Ile Ife, for two weeks acclimatization.

Thirty healthy specimens of the fish (sex ratio 1:1) all of fairly equal sizes were distributed into each of sixteen labelled hapas at the stocking density of three fish per m^2. The fish were stocked in duplicates for each dietary treatment. They were fed at 3% of their body weight twice daily within 8.00-9.00 a.m and 6.00-7.00 p.m for a period of 84 days. Renewal of water, cleaning of hapas, ration adjustment and length-weight data collection were done every two weeks.

The proximate composition of the feedstuff (Table 1) and diets (Table 2) was determined at the Food and Meat Laboratory of Animal Science Department of the Obafemi Awolowo University, Ile-Ife, using standard analytical methods (AOAC, 1990). The gross energy content was determined by oxygen bomb calorimetry using a Gallenkamp ballistic bomb calorimeter essentially as described by Ackermann et al. (1969).

Turkey-HSD one-way ANOVA coupled with Duncan's multiple range test were performed on the data generated to facilitate between treatment comparisons at 5% level of significance (Duncan, 1955). Analyses were performed using the statistical software package SPSS Version 10 for Windows.

3. Results

The proximate composition of the feedstuff and diets as well as the mineral content of the experimental diets fed to C. gariepinus are shown in Tables 1, 2 and 3.

Soybean meal used in this study, whether raw or heated, contained crude protein that was in the range of 40.1 % to 42.9 % (Table 1).

Table 2: Experimental diet formulations (% dry matter basis). computations. the gross energy content (kcal g^{-1}) and the proximate composition (%) of the experimental diets.

	DIETS							
Formulation	**DFM**	**D0**	**D5**	**D10**	**D15**	**D20**	**D25**	**D30**
Soybean	0.00	33.35	34.43	34.43	34.43	34.43	34.2	34.58
Fishmeal	37.77	4.42	3.34	3.34	3.34	3.34	3.57	3.19
Brewery wastes	37.78	37.78	37.78	37.78	37.78	37.78	37.78	37.78
Yellow maize	22.45	22.45	22.45	22.45	22.45	22.45	22.45	22.45
Mineral/Vitamins[1]	1.00	1.00	1.00	1.00	1.00	1.00	1.00	1.00
Vegetable oil	1.00	1.00	1.00	1.00	1.00	1.00	1.00	1.00
Total	100.00	100.00	100.00	100.00	100.00	100.00	100.00	100.00
Computations								
Crude Protein	31.70	30.52	29.99	30.96	31.25	30.89	30.83	31.01
Gross Energy	4.28	3.38	3.43	3.46	3.37	3.86	3.93	3.06
Proximate composition $(\overline{X}(SE))^*$								
Crude protein	31.2 (1.1)[a]	30.5 (0.6)[a]	30.9 (2.3)[a]	30.5(3.1)[a]	29.5 (0.9)[a]	30.6 (1.4)[a]	31.2 (2.2)[a]	31.0 (2.5)[a]
Moisture content	3.7 (0.8)[a]	7.0 (1.2)[a]	7.0 (2)[a]	6.6 (0.1)[a]	5.5 (0.1)[a]	5.1 (1)[a]	4.9 (1.9)[a]	4.5 (0.6)[a]
Lipid Content	5.8 (0.1)[a]	3.4 (1.4)[a]	3.5 (0.1)[a]	4.0 (1.1)[a]	4.7[a]	4.7 (1.2)[a]	4.8 (0.9)[a]	3.8 (0.1)[a]
Ash	11.7 (1.2)[a]	8.1 (0.4)[b]	7.7 (1.0)[c]	6.8 (2.0)[c]	7.2 (1.41)[c]	7.4 (0.8)[c]	7.9 (0.3)[b]	9.3 (0.4)[b]
Crude fibre	1.5 (1.1)[a]	6.1 (0.1)[b]	6.9 (0.1)[b]	7.9(0.1)[c]	7.8 (1)[b]	5.7 (0.1)[c]	5.9 (0.3)[c]	6.2 (0.6)[b]
NFE[2]	46.1 (1.2)[a]	44.9(1.6)[b]	44.0 (1.4)[b]	44.2 (1.4)[b]	45.3 (1.5)[b]	45.9 (0.1)[b]	46.3 (1.2)[a]	45.2 (1.4)[b]

*Mean and Standard error $(\overline{X}(SE))^*$, Values with the same superscript in each column are not significantly different from each other ($p>0.05$)

[1]Vitamin premix - A Pfizer livestock product containing the following per kg of feed:
Vit. A. 4 000 000i.u; Vit. D3, 800 000i.u; Vit.E, 10 000mg; Vit. K3; 1 200mg;
Vit.B1, 1 000mg; Vit. B2. 1 500mg; Vit. B6, 1 500mg; Niacin, 10 000mg; Panthothenic acd. 3 500; Biotin 15mg; Vit. B12, 10mg; Folic acid. 200mg; Chlorine chloride, 120 000mg; Manganese. 60 000mg; Iron. 15 000mg; Zinc. 15 000; Copper. 800mg; Iodine. 400mg; cobalt, 80mg; Selenium 40mg; anti-oxidant. 40 000mg.
[2]NFE = Nitrogen-free Extract = 100 – (Crude protein + Crude fibre + Lipid content + Moisture content + Ash)

As the duration of heating of the soybean increased, the lipid content increases while the moisture content decreases. The crude fibre content of the soybean-based diets (6.1-7.9%) was significantly higher ($p<0.05$) than the value (1.5%) recorded for fishmeal-based diet. The control diet had gross energy (4.28 kcal g^{-1}) and ash content (11.7%) that were significantly higher ($p<0.05$) than those of the soybean-based diets (3.06-3.93 kcal g^{-1} and 6.8-9.3%) respectively. The fishmeal-based diet was significantly richer in the assayed minerals when compared to the other diets, except for phosphorus content whose values ranged within a narrow amplitude irrespective of the diet (Table 3).

The nutrient utilization and growth performance of the male and female C. gariepinus fed the different diets are presented in Tables 4 and 5 respectively. Relatively high FCR was recorded for the soybean-based diets and most especially from those prepared from the raw and the inadequately - heated soybean meals (D5, D10). The protein intake, PER and ANPU of the fish fed DFM and D25 diets were significantly higher ($p<0.05$) than those fish fed the other diets. Irrespective of the sex of the fish, there was no reduction in weight of fish. However, the male and female broodfish fed the fishmeal-based diet had significantly higher ($p<0.05$) mean weight gain, percentage weight gain, specific growth rate and protein efficiency ratio than those fish fed the other diets. The females were found to have higher weight increases than the males and were in healthier condition than their male counterparts fed the same diets. In both male and female fish, the condition factor of the broodfish fed experimental diets D0, D5, D10, D15 and D30 was less than 1:00, an indication of poor growth quality or poor health.

4. Discussion

The crude protein contents recorded for the differently heated soybean meals used in this study are close to what a number of authors have reported for soybean (Koch *et al.* 1978; Keembiyehetty and

Table 3: The mineral content[1] of the experimental diets ($\mu g\ g^{-1}$)

Element	DFM	D0	D5	D 10	D 15	D20	D25	D30
Fe	16.10	5.29	6.59	10.00	6.02	11.19	11.30	4.64
Cu	0.15	0.07	0.03	0.07	0.06	0.19	0.20	0.03
Zn	1.77	0.01	0.01	0.00	0.00	0.01	0.01	0.00
Mg	39.79	0.36	0.43	0.39	0.36	3.17	3.60	0.36
Ca	418.45	8.47	3.20	2.98	2.96	66.36	76.36	3.02
P	7.56	6.80	5.70	5.90	6.28	6.86	7.40	5.38

[1]Mean of three determinations

Table 4: Nutrient utilization of the male and female broodstock fish fed the differently treated diets.

	DFM	D0	D 5	D10	D15	D20	D25	D30
Male								
T.F.I (g)	548.40^{a}	479.90^{b}	480.20^{b}	500.20^{b}	519.40^{b}	529.30^{a}	539.00^{a}	469.90
D.F.I (g)	6.53^{ab}	5.71^{b}	5.72^{b}	5.95^{b}	6.18^{ab}	6.30^{b}	6.41^{a}	5.58^{b}
P.I (g)	2.02^{a}	1.80^{ab}	1.77^{ab}	1.80^{ab}	1.92^{a}	1.95^{a}	1.99^{a}	1.73^{ab}
FCR	2.77^{a}	16.00^{b}	6.61^{c}	6.00^{d}	6.20^{d}	3.80^{ae}	3.27^{ae}	5.70^{c}
PER	1.16^{a}	0.20^{d}	0.49^{cd}	0.55^{c}	0.52^{c}	0.85^{c}	0.98^{a}	0.57^{c}
ANPU	20.21^{a}	5.28^{b}	9.88^{c}	9.95^{c}	10.26^{c}	11.52^{c}	14.11^{d}	10.22^{c}
Female								
T.F.I (g)	548.40^{a}	479.90^{b}	480.20^{b}	500.2^{b}	519.4^{ab}	529.3^{a}	539.00^{a}	469.90
D.F.I (g)	6.53^{a}	5.70^{b}	5.72^{b}	5.95^{ab}	6.18^{ab}	6.30^{a}	6.41^{a}	5.58^{b}
P.I (g)	2.04^{a}	1.77^{a}	1.77^{a}	1.85^{a}	1.92^{a}	1.95^{a}	1.99^{a}	1.73^{a}
PER	1.32^{a}	0.33^{b}	0.50^{bc}	0.70^{c}	1.00^{c}	1.02^{a}	1.09^{a}	0.89^{c}
FCR	2.45^{a}	9.70^{b}	6.50^{bc}	4.61^{c}	3.23^{a}	3.17^{a}	2.97^{a}	3.6^{ac}
ANPU	22.11^{a}	5.62^{b}	10.48^{d}	10.94^{d}	11.22^{d}	11.82^{d}	14.65^{c}	9.032^{d}

❖ Values with the same superscript in each row are not significantly different from each other ($p>0.05$)

TFI – Total feed intake, D.F.I – Daily feed intake, P.I – Protein intake, PER – Protein efficiency ratio, FCR – Food conversion ratio, ANPU—Apparent net protein utilization

Table 5: Weight gains (g), specific growth rate and condition factor (k) $(\overline{X}(SE))^*$ of the male and female broodstock fish fed the experimental **diets.**

	Diets							
	DFM	**D0**	**D5**	**D 10**	**D 15**	**D20**	**D25**	**D30**
Male								
ıt. Wt. (g)	208.0 (13)^a	204.0 (7.8)^a	215.5 (10.7)^b	191.5 (23.5)^a	214.8 (21)^b	214.3 (15)^b	222.2 (12.8)^b	206.9 (14.8)^a
in. Wt.(g)	406.2 (15.7)^a	286.0 (23)^c	288.3 (15.8)^b	271.3 (17.5)^c	298.8 (16)^b	340.0 (23)^a	386.8 (28)^a	291.0 (16.8)^b
I.W.G(g)	198.2 (23)^a	82.0 (7.5)^d	72.0 (4.8)^c	83.7 (6.9)^c	84.0 (10)^c	125.7 (6.6)^b	164.6 (7.9)^b	82.1 (5.8)^c
.W.G	95.3^a	40.2^c	33.8^c	43.7^c	39.0^c	58.7^b	74.1^b	39. 6^c
.W.G g)	2.4 (0.8)^a	0.5 (0.2)^d	0.9^c	1.0 (0.2)^c	1.0 (0.3)^b	1.5 (0.1)^b	2.0 (0.4)^a	1.0 (0.3)^c
.G.R.	0.4 (0.2)^a	0.1^c	0.2^c	0.2^b	0.2^{ab}	0.3^{ab}	0.3^{ab}	0.2 (0.1)^b
	1.1 (0.2)^a	0.6 (0.1)^b	0.8^b	0.8 (0.1)^b	0.8 (0.1)^b	1.1 (0.3)^{ab}	1.1 (0.1)^a	0.8 (0.1)^b
emale								
ıit. Wt.(g)	221.0 (10)^a	227.2 (3)^a	183.3 (24)^b	207.0 (11)^{ab}	182.4 (19)^b	192.0 (58)^{ab}	198.4 (9)^a	206.5 (24)^{ab}
in. Wt.(g)	425.3 (23)^a	285.0 (6)^c	257.4 (97)^b	277.5 (54)^b	272.9 (65)^{ab}	359.0 (73)^a	379.7 (63)^a	304.0 (82)^b
I.W.G(g)	204.0 (58)^a	57.8 (23)^e	74.1 (2.9)^d	70.5 (23)^c	90.4 (24)^b	167.0 (8)^a	181.3 (33)^{ab}	97.5 (6)^c
.W.G	92.3^a	25.4^d	40.4^c	34.1^c	49.6^b	86.9^c	91.4^a	47.2^{bc}
.W.G (g)	2.4 (1)^a	0.7^d	0.9 (1)^c	0.8^b	1.1 (1)^{ab}	2.0 (1)^{ab}	2.2 (1)^a	1.2 (1)^b
.G.R.	0.4^a	0.1^d	0.2^{bc}	0.2 (0.1)^{bc}	0.3 (0.1)^{ab}	0.3 (0.1)^{ab}	0.34^{ab}	0.3 (0.1)^b
.f	1.28 (0.3)^a	0.7 (0.2)^c	0.8 (0.4)^b	0.8 (0.3)^c	1.0 (0.3)^b	1.0 (0.3)^b	1.1 (0.3)^{ab}	0.9 (0.2)^b

Mean and Standard error $(\overline{X}(SE))^*$

Values with the same superscript in each row are not significantly different from each other (p>0.05)

Init. Wt. – Initial weight, Fin. Wt.- Final weight, M.W.G – Mean weight gain, P.W.G – Percentage weight gain, D.W.G – Daily weight gain, S.G.R – Specific growth rate, K -- Condition Factor

Gatlin III, 1997). The low mineral content of the soybean-based diets agrees with the report of Weingartner (1987) who concluded that soybeans are not a major source of minerals but, when included in a mixed diet, contribute to overall requirements. The relatively high FCR recorded for the soybean-based diets and most especially those prepared from the raw and the inadequately-heated meals (D5, D10) could probably be due to the high fibre content of the diet and/or low feed intake by the fish. Similar high feed conversion ratios were reported by Olaleye (1991) and Ipinjolu (1997) when various plant-protein feeds were fed to Tilapia and Clarias fish fingerlings respectively. That the protein intake, protein efficiency ratio, apparent net protein utilization, mean weight gain, percentage weight gain and specific growth rate of the fish fed DFM diet were significantly higher ($p<0.05$) than those fish fed the other diets was expected since fishmeal contains all the essential minerals, trace elements and amino acids needed for fish growth in balanced proportions (Lovell, 1981, Luquet and Watanabe 1986). As noted in this study, Viola et al. (1983), Hilton and Slinger (1986) also reported that the energy content of soybean meal is lower than that of fishmeal. This could also contribute to growth depression in the fish fed the soybean-based diets. Such adverse effects on growth have also been reported by some authors (Peres et al. 2003; Fourner *et al.* 2004 and De Francesco et al. 2004). The significant difference in mean weight gain of the broodfish fed diets DFM, D25 and D20 and those fed the inadequately heated soybean-based diets is an indication that there are certain differences in the nutrient composition of the diets conferring better growth factor on the fish. This could be attributed to differences in the heating time of the soybean component of the diet which may correlate with the level of trypsin inhibitor as well as the imbalance in the amino acid composition of soybean. Soybean components of the diets heated for less than 25 minutes could therefore be regarded as being under-heated. Heating the soybean components for more than 25 minutes probably resulted in its overheating. Both over and under - heating of soybean meal ultimately result in a diet of lower nutritional quality. Peres et al. (2003) associated improperly heated soybean fish diet with decrease in apparent protein digestibility index, which tangentially affect fish growth. Similarly, Khalifa et al. (1992) reported that overheating soybean meal might result in denaturation of some essential amino acids, reduction in availability of lysine and arginnine which consequently lower protein utilization efficiency.

Lack of reduction in the initial weight even in treatments where the fish were fed with raw or inadequately heated soybeans is due to the fact that adult animals have a lower amino acid requirement than the young fish (Rackis and Gumbmann, 1981). The low condition factor recorded for the fish fed diets D0, D5, D10, D15 and D30 is an indication of poor growth quality or poor health. The occurrence of significantly higher ($p<0.05$) weight increases and healthier condition of the females above their male counterparts fed the same diets were also reported by Tsadu (1996) and De Silva and Anderson, (1998). This was probably due to the gains in weights of the female gonads full of matured eggs in breeding season.

5. Conclusion

This study showed that soybean autoclaved at 116 °C (1.2kg/cm^2 pressure) for 25 minutes was the best substitute for fishmeal (i.e diet D25) in the diets of the African catfish, C. gariepinus for aquaculture. In culture conditions, the female C. gariepinus grow faster and are in better condition than their male counterparts due to rapid increases in weight being contributed by the female gonads especially in the breeding season.

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