



GROUNDNUT CAKE AS ALTERNATIVE PROTEIN SOURCE IN THE DIET OF *CLARIAS GARIEPINUS* FRY

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ABSTRACT

In Aquaculture, fish feed is conventionally formulated to contain high amount of fishmeal (32-40%) and this is very expensive thus increasing the cost of fish production. Also, it competes with man (focus consumer) for fish (food product of Aquaculture). Groundnut cake is alternative protein source but deficient in some vital amino acids (such as lysine and methionine) that are present in fishmeal. A 30-day feeding trial was conducted to study the effects of lysine and methionine supplemented groundnut cake diets on the growth, feed utilization and survival of *C. gariepinus* fry. Five treatment diets at 42% crude protein were prepared to contain fortified groundnut cake (GNC) with lysine (L) and methionine (M) each per 100kg of feed. Diet 0 (only GNC), Diet 1 (GNC + 0.25kg L&M), Diet 2 (GNC + 0.35kg L&M), Diet 3 (GNC + 0.45kg L&M) and Diet 4 (GNC + 0.55kg L&M). Water quality parameters were measured according to standard methods. Three-day old fry of similar sizes (0.005 ± 0.002 g to 0.008 ± 0.002 g) were fed treatment diets in triplicate groups of 50 fry per replicate. After the feeding trial, fry fed Diet 0 showed lowest growth and feed utilization parameters except feed conversion ratio (FCR) (0.10 ± 0.04) compared with other diets. Fry fed Diet 3 and or Diet 4 showed better growth performance (growth and feed utilization parameters). Lower feed conversion ratios (FCR) ranged between 0.04 ± 0.02 (Diet 3) and 0.08 ± 0.02 (Diet 1) were recorded. GNC diets did not affect water quality parameters. Temperature, pH and dissolved oxygen except ammonia were within the acceptable ranges for fish culture. Fortified GNC with lysine and methionine totally replaced fishmeal in the feed of *C. gariepinus* fry without affecting its growth and survival. GNC supplemented with at least 0.45kg each of L & M per 100kg of feed is suggested for fish feed.

KEYWORDS: Plant protein source, growth performance, essential amino acids, *Clarias gariepinus* fry, feed efficiency

INTRODUCTION

In Aquaculture, fish feed is conventionally formulated to contain high amount of fishmeal (32-40%) (Richard and Chapman, 2007). Fish meal is very expensive thus increasing the cost of fish production. Also, it competes with man (focus consumer) for fish (food product of Aquaculture). Groundnut cake is alternative protein source. It is deficient in some vital amino acids (such as lysine and methionine) that are present in fishmeal (Eyo and Olatunde, 1998). Its amino acid quality improves in artificial diets when reinforced with lysine and methionine.

Aquaculture in Nigeria is in the development stage because it has not been able to meet the demand and supply of the ever-increasing populations. It is acknowledged as the efficient means of providing food which is rich in protein (Milla, 2003). Ojotiku (2008) noted that interest in fish culture is growing very rapidly in Nigeria but the scarcity of fingerlings of culturable fish such as *Clarias gariepinus* tends to constitute a major constraint to the rapid development of fish farming in Nigeria. This is supported by Akinsanya and Otubayo (2006) who confirmed that *C. gariepinus* from the family Clariidae is generally considered as one of the most important tropical catfish species for aquaculture in West Africa. Early catfish producers depend primarily on natural pond organisms to provide nutrients essential for fish growth. Fish production was often enhanced by the

addition of fertilizers to pond water to stimulate the growth of natural food organism (Robinson and Li, 1999). Prepared feeds, mixtures of feed processed into various forms were used to supplement natural productivity. Supplemented feeds were largely steam-pelleted feeds that provide protein, energy, but were generally deficient in micronutrients such as vitamins, minerals and essential amino acids.

There has been a competition to the use of animal protein both by the feed producers and the populace due to growing interest in fish farming. Richard and Chapman (2007) stated that aquaculture feeds characteristically contain a higher percentage of protein than feeds used for poultry, swines and cattle. A typical commercial production diet formulated for tilapia or catfish contain approximately 32%-40% proteins, while on a commercial grower diet formulated for most terrestrial animals rarely exceeds 20% in total protein. Protein in the majority of formulated fish diets (world wide) depends greatly on fishmeal which is more costly than high quality plant-based protein sources, such as soybeans (Richard and Chapman, 2007).

Fish provides the cheapest source of animal protein in the world and a good source of essential amino acids (Tobor, 1990). With the ever increasing cost of fish meal in the tropics, (Halver, 2008) noted that several researchers have attempted to incorporate plant protein into formulated fish diet in order to reduce the quantity and cost of fish meal

without reducing the quality of the feed. According to Floyd (2002) fishmeal contains some amino acids and fatty acids that are not present in tissues from terrestrial plants. The most limiting amino acids in plant protein are lysine and methionine (Wikipedia, 2001; Eyo, 2003).

Groundnut cake with crude protein content of 40-45% is a good supplement. It promotes growth and is palatable to fish. Groundnut cake protein is known to be deficient in lysine and methionine and also has a limited amount of tryptophan and threonine but amino acid quality improves in artificial diets when reinforced with lysine, methionine and tryptophan (Eyo and Olatunde, 1998). Groundnut is a valuable source of vitamins E, K and B. It is the richest plant source of thiamine (B₁) and also rich in niacin, which is low in cereal FAO (2000). This study was designed to determine growth performance, feed utilization and survival of *Clarias gariepinus* fry fed different lysine and methionine supplemented groundnut cake diets.

MATERIALS AND METHODS

Study Area

The project was carried out in the finfish hatchery unit of African Regional Aquaculture Center (ARAC) Aluu, Port Harcourt. The hatchery unit consists of both the outdoor and the indoor hatchery. The indoor hatchery has two rectangular concrete tanks of 7m x 7m x 1m each, six rectangular concrete tanks of 7m x 3.5m x 1m each, six concrete tanks of 6m x 4m x 1m each and also two circular tanks of 7m in diameter. The outdoor hatchery has 45 experimental concrete tanks of 1m x 0.5m x 0.5m each.

Source of fry

Three-day old fry of similar sizes (0.005 ± 0.002 g to 0.008 ± 0.002 g) from the ARAC hatchery unit were used.

Formulation and preparation of experimental diet

The treatment diets at 40% crude protein were prepared in the feed mill unit of ARAC as Diet 0, Diet 1, Diet 2, Diet 3 and Diet 4. Diet 0 contained only groundnut cake (GNC) (control). Diet 1, Diet 2, Diet 3 and Diet 4 contained lysine and methionine supplements of 0.25kg, 0.35kg, 0.45kg and 0.55kg each for every 100kg feed (Table 1). The feed was ground into powder and sieved to suit the fry gapes. It was air-tight packaged in small plastic container and kept in the fridge (Sahoo *et al.*, 2004). Table 2 shows the proximate analysis of the experimental diets.

Table 1. Percentage composition of experimental diets.

Ingredients	D ₀ %	D ₁ %	D ₂ %	D ₃ %	D ₄ %
Wheat bran	3.52	3.52	3.52	3.52	3.52
Groundnut cake	87.63	87.63	87.63	87.63	87.63
Garri	5.00	5.00	5.00	5.00	5.00
Palm oil	2.5	2.5	2.5	2.5	2.5
Premix	0.25	0.25	0.25	0.25	0.25
Bonemeal	1.00	1.00	1.00	1.00	1.00
Vitamin C	0.10	0.10	0.10	0.10	0.10
Lysine(kg)	-	0.25	0.35	0.45	0.55
Methionine (kg)	-	0.25	0.35	0.45	0.55

Table 2. Proximate analysis of treatment diets

Moisture content (%)	Ash (%)	Ether extract (%)	Crude protein (%)	Fiber (%)
10.39	8.7	2.4	42.0	0.25

Source of water

Conditioned borehole water was used for this experiment.

Experimental design and feeding trials

Fry of similar sizes were reared in fifteen transparent aquaria (30cm diameter x 25cm depth) at 50 fry per replicate (aquarium). Initial individual total lengths and weights of randomly selected 10 fry were recorded for each replicate. Fry were fed three times daily (07.00hrs, 13.00hrs and 18.00hrs) at 10% body weight for 30 days. Water renewal was done regularly before each feeding to remove uneaten feeds and to prevent fouling and about 80-90% of the culture water was always replaced every morning. The survival of fry was determined everyday by counting and recording the mortalities.

Measurement of weights and lengths

At five days interval, 10 fry were randomly sampled and weighted in each tank. The total length was measured individually using a meter ruler. Daily food ration requirement was adjusted according to weight. The fry was put on a filter paper and the length of the head to the end of the tail was marked on the paper and this was measured with a ruler calibrated to 0.1cm. For weighing, each fry was carefully siphoned out from the plastic containers. They were put on a filter paper to absorb much of the water on the body before weighing in aluminum foil whose weight has been determined earlier. The weights were recorded using electronic digital balance model LE180 provided with ± 0.0001 g accuracy.

Water quality parameters measurement

The water quality parameters recorded were the temperature, pH, oxygen and ammonia. The water temperature and pH were measured 3 times daily (07.00hrs, 13.00hrs and 18.00hrs) using portable pH meter (pH-009(111) ATC). The dissolved oxygen and ammonia nitrogen levels were measured once daily (07.00hrs) using Freshwater Aquaculture Testing kit Model AQ-2 (Code 3633-03).

Data Analyses

Data were subjected to analysis of variance (ANOVA), Duncan Multiple Range and descriptive statistics using SAS (2003) statistical package.

RESULTS

Growth, feed utilization parameters and survival rate

Growth and feed utilization parameters, and survival rate are shown in Table 3. Fry fed Diet 1 showed significantly lower length, weight, relative growth rate, relative weight gain and specific growth rate. Fry fed Diet 3 and Diet 4 indicated insignificant growth and feed utilization while those fed Diet 0 had the least values of growth and feed utilization parameters. All fry showed a uniform increase in weight in all the diets within the first 10 days of feeding trials. The weight gain difference becomes significant after the 10th days. Survival rate of 100% was recorded for fry fed Diet 2, Diet 3 and Diet 4 while Diet 0 and Diet 1 were $93.04 \pm 1.4\%$ and $87.57 \pm 1.4\%$ respectively.

Water quality parameters

The results of the physico-chemical parameters are shown in Table 4. Dissolved oxygen was similar in all culture

tanks except in Diet 0 tank. Temperature was significantly higher in Diet 4 tank. pH and ammonia were similar in all culture tanks.

Table 3. Growth parameters, feed utilization parameters and survival rate of *C. gariepinus* fry fed different treatment diets

PARAMETER	Diet 0	Diet 1	Diet 2	Diet 3	Diet 4
INITIAL LENGTH (cm)	0.59±0.03 ^{ab}	0.56±0.02 ^b	0.61±0.02 ^{ab}	0.022±0.03 ^b	09.66±0.02 ^a
INITIAL WEIGHT (g)	0.005±0.002 ^b	0.006±0.002 ^b	0.006±0.003 ^a	0.007±0.002 ^a	0.008±0.002 ^a
INITIAL K	4.39±0.85 ^{ab}	4.09±0.64 ^b	3.39±0.4 ^b	6.27±1.1 ^a	2.62±0.79 ^b
FINAL LENGTH (cm)	1.9±0.04 ^d	2.01±0.04 ^c	2.0±0.05 ^c	2.3±0.05 ^b	2.43±0.04 ^a
FINAL WEIGHT (g)	0.06±0.005 ^c	0.07±0.001 ^{b c}	0.08±0.001 ^{ab}	0.09±0.001 ^a	0.09±0.001 ^a
RWG (%)	0.06±0.005 ^c	0.07±0.001 ^{b c}	0.08±0.001 ^{ab}	0.09±0.001 ^a	0.09±0.001 ^a
SGR (%BW/day)	0.64±0.16 ^b	1.06±0.16 ^{ab}	0.88±0.17 ^{ab}	1.12±0.16 ^a	1.26±0.14 ^a
FCR	0.1±0.04 ^a	0.05±0.04 ^a	0.08±0.02 ^a	0.14±0.04 ^a	0.05±0.03 ^a
PER	0.0001±0.00001 ^a	0.0002±0.0001 ^a	0.0002±0.0001 ^a	0.0001±0.0001 ^a	0.0001±0.0001 ^a
Final K	0.66 ±0.06 ^b	0.89±0.11 ^{ab}	1.09 ±0.09 ^a	0.85±0.08 ^{ab}	1.04±0.12 ^a
FE	3.7 ±0.6 ^b	8.46±1.87 ^{ab}	6.15±0.94 ^{ab}	8.98±1.37 ^{ab}	10.33±0.43 ^a
SR	93.04±1.4 ^b	87.57±1.4 ^b	100 ±0.0 ^a	100±0.0 ^a	100±0.0 ^a
RGR	0.03±0.005 ^b	0.048±0.01 ^{ab}	0.39 ±0.02 ^{ab}	0.052±0.01 ^a	0.057±0.01 ^a
PI	0.05±0.02 ^b	0.21±0.001 ^a	0.06±0.07 ^b	0.12±0.1 ^{ab}	0.21±0.3 ^a

Means with the same letters in the same row are not significantly different (P>0.05)

K-Condition factor, RWG-Relative weight gain, FCR-Food conversion ratio, PER-Protein efficiency ratio, FE-Feed efficiency, SR-Survival rate, RGR-Relative growth rate, PI-Performance Index

Table 4. Water quality parameters of culture tank water during the feeding trials

Water quality parameter	D ₀	D ₁	D ₂	D ₃	D ₄
Temperature (°C)	26.00±0.4 ^b	27.00±0.32 ^{ab}	26.2±0.31 ^b	27.00±0.44 ^{ab}	28.24±0.5 ^a
pH	6.66±0.11 ^a	6.68±0.31 ^a	6.54±0.25 ^a	6.99±0.22 ^a	6.74±0.32 ^a
Dissolvedoxygen (mg/l)	7.40±0.4 ^a	6.67±0.32 ^{ab}	6.57±0.22 ^{ab}	6.63±0.11 ^{ab}	6.41±0.88 ^{ab}
Ammonia (mg/l)	0.41±0.32 ^a	0.32±0.5 ^a	0.37±0.44 ^a	0.34±0.33 ^a	0.32±0.03 ^a

Means with the same letter in the same row are not significantly different (P>0.05)

DISCUSSION

The protein content of GNC (40-45%) was sufficient for the growth and survival of *C. gariepinus* fry. Lack of lysine and methionine in Diet 0 affected the growth performance of the fry. The highest FCR indicated the lowest growth. Lysine and methionine in Diets 1 to 4 enhanced the growth, feed utilization and survival of *C. gariepinus* fry. The recorded lower FCR in these diets showed that they were of higher quality than Diet 0. This further means that the fry converted small amount of these diets to flesh (growth). Immediate removal of left-over feed from the culture tanks might be responsible for the acceptable ranges of the water quality parameters. The higher level of ammonia could be linked to partial removal of the culture water.

The insignificant performance of fry at the first 10 days of feeding trials indicates that larvae cannot make well use of

dry feed until after 10 days. This is in agreement with ACE (1995) that after a period of 10 days the larvae gradually adapt to dry feed. The observed least growth performance and feed utilization in fry fed Diet 0 could be attributed to lack of lysine and methionine in the diet. Floyd (2002) noted that the growth of African catfish *C. gariepinus* fed complete plant protein as main protein source were poorer than those feed complete fish meal because fish meal contains some amino acids not present in tissues from terrestrial plant. But when plant protein supplemented with lysine and methionine, there were improvement in all the diets (D₁, D₂, D₃, and D₄). This remarkable improvement can only be possible when the diets are supplemented with different levels of lysine and methionine. Li and Robinson (Li and Robinson, 1998) and

Chang *et al.* (2003) demonstrated that supplementing plant protein with lysine and methionine in low protein diet is beneficial for catfish growth performance. The present study showed that fry fed Diet 4 and Diet 3 had better growth performance and feed utilization compared to other diets. The lower food conversion ratio for Diets 1-4 was an indication that these diets are more quality feed than Diet 0. Lower food conversion ratio indicates higher food conversion efficiency which results in higher growth (Ugwumba and Ugwumba, 2007). The water quality parameters except ammonia were within the acceptable range for fish culture (Swann, 2006). The higher level of ammonia in culture tanks could be linked to partial removal of culture water. It can then be recommended that groundnut cake can be supplemented with least 0.45kg of lysine and methionine per 100kg feed for better growth performance of *C. gariepinus* fry.

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