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Growth performance and haematological responses of African Mud Catfish (*Clarias gariepinus*) fed dietary levels of *Mucuna pruriens* meal

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ABSTRACT

Feeding trial was conducted in 15 aquaria tanks each of which had a size of (1.8 x 1.3 x 0.36m^3) to assess the performance of *C. gariepinus* fingerlings fed diets containing toasted *Mucuna pruriens* seed meal (MBM) as an alternative protein source to soybean meal. Five isonitrogenous (crude protein – 40%) diets were formulated containing toasted *M. pruriens* seed meal (MBM) at D1 (0% MBM), D2 (25% MBM), D3 (50% MBM), D4 (75% MBM) and D5 (100% MBM) and were fed at 5% body weight to triplicate groups of 15 fingerlings (Av. initial weight of 3.70±0.01g) of *C. gariepinus* for twelve (12) weeks. Growth performance and feed utilization parameters indicated that D2 (25% MBM) diet gave the highest weight gain. Similarly, the highest specific growth rate (1.41±0.01) was recorded in D2 (25% MBM), followed by D1 (0% MBM) (1.38+0.01) and the least value (1.25±0.01) was obtained in D4 (75% MBM). The feed conversion ratio, haematological parameters (PVC, HB, RBC and WBC) values were significantly different (P<0.05) across the treatments. The results from this study indicate that toasted *M. pruriens* seed meal could be used to replace soybean meal up to 25% in the diet of *C. gariepinus* fingerlings without compromising growth and feed utilization.

Keywords: Mucuna bean, plant protein, *C. gariepinus*, haematology

INTRODUCTION

The consumption and demand for fish as a cheap source of protein is increasing by the day in Africa, due to the level of hunger and poverty. Fish and fishery products play a vital role in food security and meeting the nutritional needs of the human population in developing and developed countries (FAO, 2014). The fast growth of the aquaculture industry experienced in the last two decades is as a result of the progressive intensification of production systems and use of quality fish feeds, which meet the nutritional requirement of cultured fish (FAO, 2013). However, the greatest challenge facing the industry today is the high cost of feed ingredients which has made feed industries and farmers to compromise quality for affordability (FAO, 2008). The rise in price of fish feed ingredients, especially fish and soybean meal has greatly inhibited the growth of Aquaculture due to high demand and limited supply.

Soybean meal is one of the most nutritious of all plant protein sources (Batal, 2000). Due to its high protein content, high digestibility and amino acid profile, it is widely used as feed ingredients for many aquaculture species (Storebakken *et al*., 2000). However, wider utilization and availability of soybean is limited by increasing demand for human consumption and for animal feed industries (Siddhuraju and Becker, 2001) hence the need to focus on using less expensive and readily available plant protein sources to replace soybean without reducing the nutritional quality for fish feed production. Such a protein source being sought for must have low or no direct demand for human consumption so that they can contribute maximally towards satisfying the demand for aquaculture feed industries (Francis *et al*., 2001).

Mucuna pruriens commonly known as velvet beans contain a high amount of carbohydrate and macro- and microelements. The plant is used as food and herbal medicine (Siddhuraju *et al*., 1996). *M. pruriens* seeds are an underutilized legume grown predominantly in Africa, Asia and in some part of America (Vadivel and Janardhanan, 2000). The seeds are rich in minerals, carbohydrates, fibre and has a high protein concentration of 23-35%. The nutritive value of *M. pruriens* seeds is similar to the highly-priced soybean. It compares well with other legume crops such as jack bean (*Canavalia ensiformis*), yam bean (*Sphenostylis stenocapa*), kidney bean (*Phaseolus lunatus*), pigeon pea (*Cajanus cajan*), and bambara nut (*Voandzeia subterranean*), (Ologhobo, 1992; Ukachukwu and Obioha, 1997). Haematological components of blood are also valuable in monitoring toxicity especially with feed constituents that affect the formation of blood (Dienye and Olumuji, 2014). The objective of this study was to determine the rate of growth, haematological indices and the economic viability of feeding *C. gariepinus* with various inclusion levels of *M. pruriens* seed meal (MBM).

MATERIALS AND METHODS

Seed collection and processing: *Mucuna pruriens* seeds were obtained from Zaria in Kaduna State, Nigeria. The seeds were soaked in tap water at room temperature (27 °C) for 24 hrs. The seed coat was then removed and the decorticated seeds were boiled for one hour, sun-dried for a while and then oven-dried at 70 °C for 48 h to 10% moisture content. The processed seeds were sent for proximate analysis using the method of AOAC (2005). Fish meal, soybean meal and other feedstuffs were obtained from a commercial market in Port Harcourt, Rivers State, Nigeria. All the feed ingredients were separately milled and screened to fin particle size. Energy and crude protein content of feeds were calculated from the nutrient composition of each ingredient used in the formulation of the experimental diet.

Diet formulation: Feed ingredients were weighed and mixed in appropriate proportions to give the desired nutrient required by the fish. The ingredients were mixed until uniformly blended. Water was added slowly to the mixture with continuous stirring to form a dough. The dough was pelleted with a hand pelletizer. Ingredients and composition of fish diets are shown in Table 1. The experimental diet was analyzed for proximate composition following the methods of the Association of Official Analytical Chemical (AOAC, 2005). Five dry diets containing 39% CP were formulated based on the varying inclusion levels (0, 25, 50, 75, 100%) of *M. pruriens* seeds meal (MBM) were represented as follows; D1, D2, D3. D4 and D5 respectively. The control diet D1 contained 0% of MBM.

Table 1: Different feed ingredients and their percentages in the experimental diets

INGREDIENT	D1 0% MBM	D2 25%MBM	D3 50%MBM	D4 75%MBM	D5 100%MBM
MBM	-	7.73	18.45	34.34	60.29
SBM	26.61	23.19	18.45	11.45	-
Fishmeal	26.61	26.61	26.61	26.61	26.61
Wheat bran	20.66	18.51	15.52	11.08	3.83
Yellow maize	20.66	18.51	15.52	11.08	3.83
Bone meal	1	1	1	1	1
Vitamin premix	0.5	0.5	0.5	0.5	0.5
Lysine	1	1	1	1	1
Methionine	1	1	1	1	1
Vitamin C	0.2	0.2	0.2	0.2	0.2
Salt	0.25	0.25	0.25	0.25	0.25
Starch (binder)	0.5	0.5	0.5	0.5	0.5
Palm oil	1	1	1	1	1
Total	100	100	100	100	100

Experimental area and fish: The experiment was carried out at the aquaculture unit of the University of Port Harcourt demonstration farm, Port Harcourt, Rivers State, Nigeria. A total of 225 fingerlings of *C. gariepinus* with an average weight of 3.71g were acclimatized to experimental conditions for seven days. After acclimatisation, fifteen randomly selected fish of uniform size were stocked into each of 15 aquaria tanks (1.8 x 1.3 x 0.36m^3).

Water quality monitoring: Temperature, pH, dissolved oxygen and ammonia were measured twice a week at 7:00–8:00 before feeding. The temperature was recorded with a digital thermometer, dissolved oxygen was determined by Winkler's method and pH was determined with Hannah E251 pH meter and ionized ammonia; was measured using an ammonia assay kit.

Proximate analysis: Proximate analysis of experimental diets was carried out according to AOAC (2005) procedures. Moisture was determined by drying weighed samples at 105°C to a constant weight and calculating the difference in weight. Nitrogen contents were analyzed by the Kjeltec system (2200 Kjeltec Auto Distillation, Foss Tecator, Sweden) and crude protein was calculated by multiplying the percent nitrogen by 6.25. The ether extract was designated as crude fat and determined using Soxtec System (1045 Soxtec Extraction Unit, Foss Tecator, Hoganas, Sweden). Diethyl ether (boiling point 40-60°C) was used as a solvent and the weight of the extract was expressed as a percent of the tissue weight. Ash content was estimated by incinerating samples in a muffle furnace at 600°C for 6 hours. Total carbohydrate (%) was calculated by subtracting crude protein, fat, moisture, and ash from the total weight. Proximate carcass compositions were analyzed before initiation and after the termination of the experiment.

Growth and nutrient parameters: The total weight gain (TWG), relative growth rate (RGR) (%), specific growth rate (SGR) (%/day), total feed intake (TFI), feed conversion ratio (FCR), protein efficiency ratio (PER) and survival (%) were determined according to the method of Jamabo *et al*., 2016. All parameters were measured for all the treatments and their replicates. Mortalities were recorded as they occurred.

Haematological parameters: The blood samples were taken from the dorsal fin of the fish following Klontz and Smith (1986) and taken to the laboratory for haematological analysis. The samples were analyzed according to methods adopted in fish haematology (Ivanova, 1983; Haghighi, 2010). The haematological parameters obtained include packed cell volume (PCV), Haemoglobin (Hb) and red blood cell (RBC). The white blood cell (WBC) and differential count (neutrophils and lymphocytes) were determined as described by Dacie and Lewis (2001).

Statistical Analysis: Data was statistically analyzed using SPSS version 16.0. One-way analysis of variance (ANOVA) and Duncan's Multiple Range Test (Duncan, 1955) was applied to compare means between treatments.

RESULTS

Composition of Experimental Diets

The gross composition of the experimental diets (Table 1) indicates that diet (D1) has no MBM inclusion and SBM was completely replaced with MBM, in D5. Diet 2, 3, 4 respectively has 25%, 50%, 75% replacement of soybean by MBM. The proximate composition of the experimental diet is shown in Table 2. The diets were isonitrogenous. Crude protein, dry matter and NFE values were not significantly different ($P>0.05$). Crude fat showed a significant difference ($P<0.05$) with the highest observed in D5 (4.36±0.005) and the least in D1 (4.24±0.005). There was a significant difference ($P<0.05$) in the ash content, moisture and crude fibre across the treatments.

Physico-chemical Parameters of Water

The summary of the Physico-chemical parameters of the experimental waters during the period of the study is shown in Table 3. Water temperature ranges from 26.54 to 26.73, pH values were between 6.54 and 7.34; while, dissolve oxygen values ranged from 4.48 and 6.16. The result reveals no significant difference was observed in temperature ($P>0.05$) among the experimental tanks, while the other parameters had a significant difference (P<0.05).

Growth and Nutrient parameters of *C. gariepinus* fingerlings

The result of the effects of the experimental diets on growth parameters of *C. gariepinus* fingerlings is presented in Table 4. Fingerlings fed D2 diet had the best growth performance and feed utilisation parameters (mean wt. gain, FCR, SGR and PER), while D3 had the poorest value for mean weight gain and PER.

Haematological parameters

The haematological parameters of *C. gariepinus* fed with different levels of Mucuna bean meal are shown in Table 5. The Packed Cell Volume (PCV) was significantly different (P<0.05) across the experimental treatments. The highest PCV was recorded in D3 (15.00±0.57), lowest PCV was recorded in D1 (11.00±0.57). Haemoglobin values were significantly different (P<0.05) across the experimental treatments. The highest HB value was recorded in D3 (5.06±0.15) while the lowest value was recorded in D1 (3.66±0.20). The RBC was significantly different across the treatments. The highest RBC was seen in D3 (3.00±0.12). The lowest RBC was recorded in D1 (2.10±0.06). WBC was significantly different in all the treatments (P<0.05). The highest WBC was recorded in D2 (12.90±0.35). D5 showed the lowest WBC and was not significantly different (P>0.05) from D1and D3.

Table 2: Proximate composition of experimental diet

Parameters (%)	D1	D2	D3	D4	D5
Crude protein	39.43±0.003^{a}	39.53±0.003^{a}	39.67±0.003^{a}	39.74±0.002^{a}	39.93±0.002^{a}
Crude fat	4.24±0.005^{a}	4.29±0.008^{b}	4.27±0.008^{a}	4.32±0.005^{a}	4.36±0.005^{b}
Crude fibre	2.27±0.011^{b}	2.23±0.008^{c}	2.35±0.011^{a}	2.19±0.008^{c}	2.33±0.011^{a}
Ash	8.71±0.008^{d}	8.77±0.008^{b}	8.67±0.008^{e}	8.81±0.011^{a}	8.73±0.008^{c}
Moisture	5.66±0.020^{ab}	5.71±0.014^{a}	5.59±0.014^{c}	5.68±0.017^{ab}	5.62±0.014^{bc}
Dry matter	94.34±0.020^{a}	94.29±0.014^{a}	94.40±0.014^{a}	94.32±0.017^{a}	94.37±0.014^{a}
NFE	39.69±0.008^{b}	39.48±0.012^{b}	39.48±0.006^{b}	39.25±0.006^{b}	39.02±0.005^{b}

*Superscripts of the same alphabet in rows are not significantly different (P<0.05); **Superscripts of different alphabets in rows are significantly different (P<0.05)

Table 3: Physio-chemical Parameters of Water

Parameters	D1	D2	D3	D4	D5
Temperature (˚C)	26.54±0.156[a]	26.68±0.129[a]	26.70±0.0136[a]	26.66±0.153[a]	26.73±0.145[a]
pH	7.34±0.074[a]	7.03±0.060[b]	6.93±0.044[b]	6.62±0.120[c]	6.54±0.102[c]
DO (mg/l)	4.74±0.131[b]	6.16±1.022[a]	4.74±0.107[b]	4.50±0.119[b]	4.48±0.145[b]
Ammonia (mg/l)	0.12±0.005[d]	0.20±0.005[a]	0.14±0.004[c]	0.20±0.008[a]	0.17±0.007[b]

*Superscripts of the same alphabet along row are not significantly different (P<0.05)

Table 4: Assessment of feed utilization by the experimental fish

Parameters	D1	D2	D3	D4	D5
Initial weight(g)	3.71+0.02[a]	3.71+0.02[a]	3.70 + 0.01[a]	3.71+0.02[a]	3.70++ 0.02[a]
Weight gain (g)	5.07±0.63[a]	5.24±0.62[a]	4.60±1.29[a]	4.61±1.63[a]	4.71±0.49[a]
SGR %day^{-1}	1.38+ 0.01[c]	1.41 + 0.01[c]	1.26 + 0.01[a]	1.25 + 0.01[a]	1.29 + 0.01[b]
PER	1.13+ 0.01[a]	1.35+ 0.01[a]	1.04+ 0.01[a]	1.15+ 0.01[a]	1.15+ 0.01[a]
FCR	0.69+ 0.01[a]	0.57+ 0.01[a]	0.80+ 0.01[b]	0.73+ 0.03[b]	0.71+ 0.01[a]

*Superscripts of the same alphabet along row are not significantly different (P>0.05); **Superscripts of different alphabets along row are significantly different (P<0.05)

Table 5: Hematological parameters of *C. gariepinus* fingerlings fed with varying inclusion levels of MBM

Parameters	D1	D2	D3	D4	D5
PCV(%)	11.00±0.57[b]	13.00±0.57[ab]	15.00±0.57[a]	14.00±1.15[a]	13.00±0.57[ab]
HB(gdl^{-1})	3.66±0.20[b]	4.36±0.20[ab]	5.06±0.15[a]	4.76±0.43[a]	4.50±0.29[ab]
RBC(x10^{6}l^{-1})	2.10±0.06[c]	2.50±0.12[bc]	3.00±0.12[a]	2.97±0.26[ab]	2.87±0.09[ab]
WBC(x10^{3})	10.47±0.72[b]	12.90±0.35[a]	10.50±0.40[b]	11.67±0.66[ab]	10.27±0.72[b]

*Superscripts of the same alphabet along row are not significantly different (P<0.05)

DISCUSSION

Proximate Composition

The range of crude protein (CP) recorded for the experimental diets were close to 32-38% reported by (Craig and Helfrich, 2004) for catfish fingerlings. Ash content of the experimental diets is an indicator of high minerals inherent in the treatments and implies that deficiencies of minerals for the fish will not be observed or recorded. Aderolu and Akpabio (2009) reported that the rate of digestion and nutrient absorption is restricted by high fibres in diets. Keembiychetty and Desilva (1993) stated that it could result in increased weight of excreta and reduced nutrient absorption. The crude fibre in this study was within the acceptable range of fibre requirement of catfish. Processes that reduce fibre content of oilseed meal could improve growth rate and weight gain of fish (Maina *et al.*, 2002).

Physio-chemical Parameters

The water quality parameters obtained in this work were within the range recommended for the culture of *C. gariepinus* (Boyd, 2000; Ajani, 2006). Boyd (2000) reported temperature range of 22 - 27^{O}C, pH range from 6.5 – 9.0 and dissolved oxygen of 6.3 – 9.6mg/l, as optimum and best for high growth performance in cultured tropical fishes. The temperature and pH values for this study falls within the acceptable range for the growth and survival of *C. gariepinus.* However, the dissolved oxygen for this study was quite low due to the feed thus, affected the growth and feed conversion rate. Ammonia has a more toxic form at high pH and less toxic form at low pH. In this study, the ammonia values show that is was toxic at some point and the water was immediately changed whenever it was observed.

Growth and Nutrient Parameters

Growth of fish depends on the nutritive quality of feeds, especially its crude protein (Ibrahim *et al*., 2018). Growth depression which was more pronounced in fish fed diets D3 may be attributed to some anti-nutritional factors in the feed as observed by (Adewumi, 2014). High fibre content in this diet was found to reduce digestibility of this diet. The feed conversion ratio is usually influenced by the quality of feed, pond water quality, and feeding management (Jamabo *et al*., 2016). Feed conversion ratio increases with high inclusion of plant protein, this may be due to the protein sparing effects by the other nutrients in the feed. Dienye and Olumuji (2014) observed that FCR increased with increase inclusion of moringa leaf and that a lower FCR is an indicator of better utilization of feed by fish. The specific growth rates were similar to the value obtained by Fagbenro and Jauncey (1995) in the diet of *C. gariepinus*, while the range of protein efficiency ratio (PER) obtained in this study is similar to result obtained by Nwanna (2003) for *C. gariepinus.*

Haematological Parameters

There was no particular trend in the haematological parameters of the experimental fish. The PCV is an indication that the animal is anaemic. The anaemic condition of the fish in this study might be due to its protein inadequacy to meet the fish nutrient requirements, which might have inhibited erythrocyte production or increased the rate of destruction. Daramola *et al*., (2005) reported that the concentration of haemoglobin (HB) in the cytoplasm of red blood cells indicates of oxygen-carrying capacity of the blood of the individual. The HB values of the fish compared favourably with normal range, indicating that the fish had sufficient blood pigment for proper transportation of oxygen, thus healthy living. Reduction in the concentration of HB and PCV suggested the presence of toxic factors such as haemagglutinin which can have adverse effects on the blood formation (Oyawoye and Ogunkunle, 1998). Decreased RBC in the experimental fish in this study is similar to the observations of Joshi *et al*., (2002) in *Puntius conchonius* and *Clarias batrachus* exposed to different toxicants.

CONCLUSION

Mucuna pruriens meal is an important feed ingredient that can be used to partially replace soybean meal in the diet of *C. gariepinus* up to 25% inclusion without significant reduction in growth performance and nutrient utilization.

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