GROWTH, HAEMATOLOGY AND SERUM BIOCHEMISTRY OF AFRICAN CATFISH *CLARIAS GARIEPINUS* FED DIETS CONTAINING MIXTURE OF PROCESSED MORINGA (*MORINGA OLEIFERA*) LEAF AND KERNEL MEAL

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ABSTRACT

The potential of blending soaked moringa (Moringa oleifera) leaf and fermented moringa kernel as a substitute for fishmeal in the diet for Clarias gariepinus was investigated. The leaves were soaked overnight while the seeds were defatted and subsequently fermented with Rhizopus stolonifer. Four approximately isonitrogenous (40 % crude protein) and iso-energetic (20 kJ/g) diets were formulated to contain graded levels (0, 20, 40 and 60 %) of mixture (1:1) of soaked leaf and fermented kernel of moringa. The diets were fed to triplicates group of C. gariepinus (average weight, 8.87 ± 0.02 g) for 56 days. Growth performance in terms of weight gain and specific growth rate in the group that received control diet was significantly higher (p<0.05) than those that were reared on diets containing 40 and 60 % mixture of soaked moringa leaf and fermented kernel but statistically similar (p>0.05) to the group fed with 20% of the mixture. The feed conversion ratio was statistically similar (p>0.05) in the control and dietary treatments. The haematological profile of the control group was similar (p>0.05) to all other treatment groups in all the parameters examined; suggesting that there were no deleterious health effects arising from incorporation of processed moringa leaf and kernel in the diets for C. gariepinus.

Keywords: *Clarias gariepinus*, Moringa leaf and kernel, Growth performance, Nutrient utilization

INTRODUCTION

Fishmeal is the most utilised animal dietary protein ingredient in aquaculture diets because of its high protein content, balanced amino acid profile, high digestibility and palatability (Tacon and Metian, 2008). It is also a good source of essential fatty acids, digestible energy, macro and trace minerals and vitamins, and generally acts as feed attractant for most finfish species (Cho and Kim, 2011). However, constant rising prices of fishmeal which is a direct results of depleted wild stocks, inconsistent supply and increased demand has been driving the search for substitutes for decades. In addition to concerns about sustainability issues, the potential presence of organic and inorganic contaminants in fishmeal has also necessitated enhanced efforts to thoroughly evaluate veritable alternatives (Naylor *et al*., 2000). The challenge facing the aquaculture industry is therefore to identify economically viable alternatives to fishmeal that are safe both for fish consumption and humans consuming farmed fish. Gatlin *et al*. (2007) noted that an alternative ingredient to fishmeal must be widely available, competitively priced, easy to handle and store among other considerations. In spite of their limitations, quite a number of plant products have been investigated for their potential in supplementing or even replacing fishmeal in the diet of fish because they are

abundant and renewable (Aslaksen *et al.,* 2007; Kumar *et al.,* 2011; Olude and Badmus, 2015). Initiatives such as addition of amino acids, flavourings and exogenous enzymes to ensure balance of amino acid profile, improve palatability and digestibility (Mukhopadhyay and Ray, 1999; Kureshy *et al.,* 2000); processing of the plant feed ingredients to improve their protein quality and digestibility and reduce or remove antinutritional factors have been taken by several investigators with varied results (Mukhopadhyay and Ray, 1999; Kureshy *et al.,* 2000; Drew *et al.,* 2007).

Moringa oleifera is one of the most useful tropical trees belonging to the Moringaceae family; it is fast growing, drought-resistant and high-yielding (Teixeira *et al*., 2014). Makkar and Becker (1996) reported that *M. oleifera* leaves, kernel and the fat free kernel meals contain 26.4, 36.7 and 61.4 % crude protein, respectively. The leaves and pods are also known to be a rich source of vitamins and minerals such as calcium, phosphorus, magnesium, ascorbic acid and tocopherol. Although the kernel meal is a good source of protein, it is deficient in lysine, leucine, phenylalanine + tyrosine and threonine when compared to the standard FAO (2011) protein. Interestingly, Egwui *et al.* (2013) pointed out that the high content of these deficient amino acids in the leaf meal could adequately compensate for them in the kernel meal. They further postulated that combination of kernel and leaf meals in desired proportions might result in obtaining a plant-based protein that would favourably replace fishmeal in fish feeds. Torstensen *et al.* (2008) had earlier suggested that by sensibly blending plant protein ingredients, the qualities that are lacking in one would be compensated by the other. However, Olude and Badmus (2015) in a previous experiment blended moringa leaf and kernel observed poor growth and nutrient utilization in *C. gariepinus* fed beyond 20 % fishmeal replacement. They suggested that the inferior performance at higher levels of supplementation might be due to nutritional-stress factors associated with leaf and kernel of moringa. Makkar and Becker (1996) reported that moringa leaves contain a negligible amount of tannins, but a high level of crude saponins, while glucosinolates, lectins and alkaloids are the major anti-nutrient substances in moringa kernel meal. Fermentation, by either bacterial or fungal organisms, has been used in many studies to reduce the negative effects of some antinutrients on digestibility and growth performance of fish (Alegbeleye *et al.,* 2012; Olude *et al.,* 2016). Soaking in water (Olude *et al.,* 2008; Alegbeleye and Olude, 2009) or alkali (Vadivel and Pugalenthi, 2009) was also affirmed to reduce antinutrient from plant proteins either singly or in combination with other processing methods. The present investigation was therefore undertaken to determine whether soaking of moringa leaf and microbial fermentation of its defatted kernel will improve their utilization by *C. gariepinus* fingerlings.

MATERIALS AND METHODS

Processing of Leaf and Kernel: Fresh leaves and seeds of *Moringa oleifera* were obtained from a residential location in Ipaja, Lagos, Nigeria. The leaves with the stalks were soaked overnight in still tap water (w: v, 1:1) in a bowl after which they were placed on a wire mesh in order to drain excess water as described by Madalla *et al.* (2013). Subsequently, the leaves were removed from the stalk and air-dried at room temperature until a constant weight was obtained. The dried leaves were milled into powder using electrical blender (Kenwood FP190). The resultant meal was sieved using a 595 µm sieve to remove the chaff and to ensure a uniform size profile as previously described (Alegbeleye *et al*., 2012). Similarly, moringa seed kernels (5 kg) were ground and defatted using n-hexane at the Oilseed Laboratory of Federal Institute of Industrial Research (FIIRO), Oshodi, Nigeria according to the method described by Honig *et al*. (1969). The defatted kernel was fermented at the Applied Microbiology Laboratory, University of Lagos, Nigeria using *Rhizopus stolonifer* (1.20×10^3 cfu/g) for 48 hours at 36^{o} C. The fermented kernel was oven dried at 70° C for 48 hour, milled using Kenwood blender and packaged in labelled polythene bags.

Diet Preparations: All other feed ingredients (fishmeal, soybean meal, maize, vitamin-mineral premix, methionine and lysine) were purchased from Sabina Pad Enterprises Limited, a commercial Feed Mill at Oko-Oba, Agege, Lagos State. All dietary ingredients were analyzed for their proximate composition. Pearson square method was used to formulate four iso-nitrogenous (40 %) and iso-energetic diets (20 kJ/g); incorporating graded levels (0, 20, 40 and 60 %) of mixture (1:1) of soaked moringa leaf meal and fermented kernel cake (Table 1). The ingredients were properly mixed and pelleted through a 2 mm die using kitchen hand crank pelletizer. The pellets were dried at room temperature for 2 days. They were packed in properly labelled cellophane bags, and stored in a refrigerator at 4^{o}C to preserve their quality before the commencement of the feeding trial.

Experimental Design: The experiment was carried out at the fish hatchery unit of University of Lagos, Nigeria using 12 plastic bowls (55.2 x 33.5 x 21 cm^3) filled to 2/3 (26 liters) of their volume with fallowed tap water. Triplicates groups of fish (average weight, 8.87 ± 0.02g), previously obtained from a local fish hatchery were randomly allotted to the four experimental diets (Table 1). The fish were fed 3% body weight offered in two equal installments at 0900 and 1600 hours for 6 days every week in a 56-day experiment. On every 7th day, the fish were batch-weighed with a top-loading balance (Camry EK5055, Zhongshan, China) in order to adjust the quantity of feed gift. Water temperature (28 – 30^{o} C), pH (6.5 – 7.5) and dissolved oxygen (3.92 – 4.02 mg/l) were regularly monitored throughout the feeding trial.

Growth Performance Assay: Growth performance and nutrient utilization were computed using the following parameters and relationships: Mean Weight Gain (MWG, g) = final wet weight – initial wet weight; Specific Growth Rate (SGR, %/day) = [(Ln final wet weight – Ln initial wet weight)/days] x 100; Feed Conversion Ratio (FCR) = feed intake / wet weight gain, Protein Efficiency Ratio (PER) = weight gain/dry protein intake.

Biochemical Assay: All analyses for proximate composition were performed according to the method of Association of Official Analytical Chemists (AOAC, 1990). Micro-kjeldahl method was used in determining crude protein following digestion, distillation and titration. Crude fibre was determined as loss on ignition after digesting moisture-free defatted sample with a weak acid (1.25 % H_2S0_4) and a weak base (1.25 % NaOH). Crude fat was estimated by weight difference after extracting oil from sample for two hours using petroleum ether in an auto fat extraction system (HT6, Tecator, Sweden). Similarly, ash content was determined by weight difference after incinerating sample in muffle furnace set at 500^{o} C for six hours. Nitrogen free extract was determined by subtracting the sum of moisture, ash, crude protein, crude lipid and crude fibre from 100. Gross energy content was estimated by calculation, based on physiological fuel values of 23.64 kJ/g for protein, 39.54 kJ/g for lipid and 17.15 kJ/g for carbohydrate according to Henken *et al.* (1986). Tannins was determined by the method of Broadhurst and Jones (1978), while the method of Wheeler and Ferrel (1971) was used to quantify phytic acid.

Two milliliters of blood samples were collected per treatment replicate from the caudal peduncle of the fish at the end of the experiment with the aid of a 2 cm plastic syringe and the blood was dispensed into ethylene diamine tetra-acetic acid (EDTA) anticoagulant bottle for haematological studies. Packed cell volume (PCV), haemoglobin concentration (Hb), red blood cell (RBC) and white blood cell (WBC) counts were determined by the method of Blaxhall and Daisley (1973). Different types of WBC were analysed in percentage into neutrophil, lymphocyte, eosinophil, monocyte and basophil. Mean corpuscular volume (MCV, fl), mean corpuscular haemoglobin (MCH, pg) and mean corpuscular haemoglobin concentration (MCHC, g/dl) were calculated using standard relationships (Blaxhall and Daisley, 1973).

Table 1: Composition (%) of experimental diets in fish meal replacement experiment

Ingredients	Control Diet	20 % MOL + FKC	40 % MOL + FKC	60 % MOL + FKC
Soybean meal	30.00	30.00	30.00	30.00
Fish meal	36.20	32.05	26.88	20.34
Maize	25.80	21.93	17.20	11.12
Moringa	0.00	8.02	17.92	30.54
Vitamin/mineral premix*	2.00	2.00	2.00	2.00
Vegetable oil	3.00	3.00	3.00	3.00
Lysine	1.00	1.00	1.00	1.00
Methionine	1.00	1.00	1.00	1.00
Starch	1.00	1.00	1.00	1.00
Proximate composition (%) of experimental diets				
Moisture	7.01	7.24	7.53	7.88
Crude protein	40.06	39.74	39.3	38.77
Ether extract	9.13	9.94	10.96	12.27
Crude fibre	1.87	2.45	3.15	4.05
Ash	6.35	6.26	6.14	5.99
Nitrogen free extract	35.58	34.37	32.93	31.04
Gross energy kJ/g	19.55	19.69	19.86	20.08

** Hi-nutrient vitamin premix supply/100 g diet : vitamin A palmitate, 1000 IU; cholecalciferol (D), 1000 IU; α-tocopherol acetate (E), 1.1mg; menadione (K), 0.02 mg; thiamine B1, 0.63 mg; riboflavin(B2), 0.5 mg; pantothenic acid, 1.0 mg; pyridoxine (B6), 0.15 mg; cyanocobalamine (B12), 0.001mg; nicotinic acid, 3.0 mg; folic acid, 0.1mg; choline, 31.3 mg; ascorbic acid (C), 0.1mg; ferrous sulphate, 0.05 mg; copper sulphate, 0.25 mg; manganese sulphate, 6.00 mg; cobalt chloride, 0.5 mg; zinc sulphate, 5.0 mg; sodium selenite, 0.02 mg; MOL- soaked moringa leaves; FKC – fermented moringa kernel cake*

Serum glucose was determined colorimetrically, while serum cholesterol was determined using enzymatic endpoint method as described by Roeschlau *et al.* (1974). Total serum protein was determined using Biuret method as described by Tietz (1995). The bromocresol green (BCG) method as described by Doumas *et al.* (1971) was employed in determining serum albumin, while serum globulin was calculated using the relationship: serum globulin (g) = total serum protein (g) – serum albumin (g). Serum aspartate aminotransferase (AST), alanine aminotransferase (ALT), alkaline phosphatase (ALP) and lactate dehydrogenase (LDH) were determined using the methods of Moss and Henderson (1999).

Data Analysis: Experimental data obtained from the feeding trials were analysed using a one-way analysis of variance (ANOVA) as described by Gomez and Gomez (1984), while Duncan's Multiple Range Test (Duncan, 1955) was used to resolve statistical variations in data means using Statistical Package for Social Science (SPSS 15.0, 2006 version, Chicago, USA) and Microsoft Excel for Windows, 2007.

RESULTS

Fermentation reduced the levels of tannin (18.46 g/kg) and phytate (16.60 g/kg) contents in the raw kernels by approximately 78 and 61 % respectively, while soaking on the other hand reduced tannins (75.80 g/kg) in the leaves by 30 %. The crude protein, ether extract and gross energy contents of moringa kernels were also reduced after fermentation (Table 2). There was a general trend of decrease in the growth performance and nutrient utilization (Table 3) of fish reared on diet containing mixture of processed moringa leaf and kernel meal with increased level of inclusion. Growth performance in terms of weight gain and specific growth rate were significantly ($p<0.05$) reduced beyond the 20 % inclusion level relative to the control. However, there was no significant ($p>0.05$) difference in the feed conversion ratio of the control and other dietary treatment.

Table 2: Proximate, gross energy and antinutritional compositions of feed ingredients

Parameters	Raw Moringa Kernels	Defatted Moringa Kernels	Fermented Moringa Kernels	Raw Moringa Leaves	Soaked Moringa Leaves	Fish meal	Soybean	Maize
Moisture %	7.90	9.70	14.30	9.20	10.10	5.25	11.72	6.19
Crude Protein %	40.15	51.83	49.04	30.53	29.85	67.21	44.00	9.80
Ether Extract %	33.14	12.80	7.18	7.29	7.28	14.80	8.60	4.50
Crude Fibre %	9.09	4.06	5.13	6.55	9.36	-	5.00	1.45
Ash %	3.14	4.04	4.49	9.08	9.75	12.66	4.63	1.46
NFE %	6.58	17.57	19.86	37.35	33.66	0.08	26.05	76.60
Gross energy (kJ/g)	24.97	20.93	18.69	17.62	17.30	21.75	19.17	17.59
Tannins (g/kg)	18.46	nd	7.10	75.80	53.25	nd	nd	nd
Phytate (g/kg)	16.60	nd	3.60	13.30	14.80	nd	nd	nd

Nd = not determine

Table 3: Growth performance and nutrient utilization of *Clarias gariepinus* fed diets containing mixture of soaked *Moringa oleifera* leaf (MOL) and fermented kernel cake (FKC)

Growth parameter	Control Diet	20 % MOL + FKC	40 % MOL + FKC	60 % MOL + FKC
Initial weight (g)	8.87 ± 0.03	8.85 ± 0.05	8.90 ± 0.10	8.87 ± 0.03
Final weight (g)	34.40 ± 2.16 ^{a}	26.61 ± 2.72^{ab}	24.00 ± 4.00^{b}	21.15 ± 1.09^{b}
Mean weight gain (g)	25.54 ± 2.18 ^{a}	17.76 ± 2.67^{ab}	15.10 ± 4.10^{b}	12.28 ± 1.08^{b}
Specific growth rate (%/day)	2.41 ± 0.12^{a}	1.96 ± 0.17 ^{ab}	1.75 ± 0.32^{b}	1.55 ± 0.09^{b}
Feed intake (g)	25.43 ± 1.25^{a}	19.13 ± 1.95^{b}	18.60 ± 1.09^{b}	17.31 ± 0.84 ^{b}
Feed conversion ratio	1.00 ± 0.04^{a}	1.09 ± 0.53^{a}	1.31 ± 0.28^{a}	1.42 ± 0.05^{a}
Protein intake (g)	10.14 ± 0.50^{a}	7.65 ± 0.78^{b}	7.44 ± 0.44^{b}	6.92 ± 0.34^{b}
Protein efficiency ratio	2.51 ± 0.10^{a}	2.31 ± 0.11^{ab}	2.00 ± 0.43^{b}	1.77 ± 0.07 ^{ab}
Survival (%)	93.33 ± 3.33	90.00 ± 0.00	85.00 ± 5.00	90.00 ± 0.00

Survival ranged from 80.00 ± 5.77 to 93.33 ± 3.33 and showed no significant ($p>0.05$) difference among dietary groups. Haematological profile of *C. gariepinus* fingerling fed tested diets decreased with increased inclusion levels of mixture of processed moringa leaf and kernel in terms of PCV, Hb, RBC and WBC with value range of 18.33 ± 4.73 – 21.00 ± 1.73 %, 5.90 ± 1.56 – 6.67 ± 0.65 g/dl, 1.67 ± 0.46 – 1.94 ± 0.17 x 10 µl and 9.70 ± 2.25 – 16.00 ± 0.50 x 10^3 µl respectively. These values were however not statistically different ($p>0.05$) in all the dietary groups. Similarly, dietary treatment did not exert any influence on the MCH, MCHC and MCV (Table 4). The serum biochemistry of *C. gariepinus* fed experimental diets showed that there was no significant difference ($p>0.05$) between the control diet and tested diets in terms of total protein, globulin, albumin, cholesterol, ALT, ALP and LDH (Table 5). The AST of the control group was similar ($p>0.05$) to 20 PM group and they are statistically higher ($p<0.05$) than those that received other diets. Serum glucose (0.93 ± 0.23) of the group that received 20 PM was significantly ($p<0.05$) higher than the control group but similar to other dietary groups.

DISCUSSION

The observed inferior performance of *C. gariepinus* fed graded levels of mixture of processed moringa leaf and kernel in the present study could be attributed to a number of factors. Firstly, in spite of the fact that processing methods employed under current investigation reduced the antinutritional factors in the leaf meal and kernel cake of moringa considerably, these ingredients still contained substantial quantities of antinutritional factors

Table 4: Haematological profile of *Clarias gariepinus* fingerling fed varying levels of diets containing mixture of soaked *Moringa oleifera* leaf (MOL) and fermented kernel cake (FKC)

Haematological profile	Control Diet	20 % MOL + FKC	40 % MOL + FKC	60 % MOL + FKC
PCV (%)	20.33 ± 2.31	21.00 ± 1.73	19.33 ± 1.16	18.33 ± 4.73
Hb(g/dl)	6.60 ± 0.79	6.67 ± 0.65	6.20 ± 0.79	5.90 ± 1.56
RBC(x 10^6 µl)	1.85 ± 0.21	1.94 ± 0.17	1.67 ± 0.19	1.67 ± 0.46
MCH(g/dl)	35.60 ± 0.87	34.43 ± 1.91	37.09 ± 0.63	35.48 ± 2.53
MCHC(g/dl)	32.45 ± 0.80	31.75 ± 1.75	31.98 ± 2.32	32.24 ± 1.96
MCV(g/dl)	10.97 ± 0.17	10.85 ± 0.40	11.41 ± 0.86	11.00 ± 0.69
WBC(10^3µl)	12.10 ± 6.93	14.97 ± 4.71	16.00 ± 0.50	9.70 ± 2.25
HET(%)	24.00 ± 1.00	21.67 ± 2.89	24.33 ± 3.05	24.00 ± 3.00
LYM(%)	70.67 ± 1.52^b	74.33 ± 3.78^a	70.00 ± 4.36^b	71.33 ± 2.52^b
EOS(%)	3.67 ± 0.58	3.67 ± 0.58	4.33 ± 1.53	3.67 ± 0.58
MON(%)	1.33 ± 0.58^b	0.33 ± 0.58^a	1.33 ± 0.58^b	1.00 ± 0.00^b
BAS(%)	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00

PCV (Pack cell volume), HB (haemoglobin), WBC (White blood cell), RBC (Red blood cell), MCH (Mean corpuscular haemoglobin), MCV (Mean cell volume), HET (Haematocrt), MCHC (Mean corpuscular haemoglobin), LYM (Lymphocytes), EOS (Eosinophils), BAS (Basophils) MON (Monocytes)

Table 5: Serum chemistry parameters of *Clarias gariepinus* fingerling fed varying levels of diets containing mixture of soaked *Moringa oleifera* leaf (MOL) and fermented kernel cake (FKC)

Serum Chemistry	Control Diet	20 % MOL + FKC	40 % MOL + FKC	60 % MOL + FKC
TP (g/dl)	2.93 ± 0.63	3.23 ± 0.63	3.07 ± 0.23	2.77 ± 0.77
GLO (g/dl)	2.30 ± 0.67	2.30 ± 0.50	2.20 ± 0.03	2.06 ± 0.75
ALB(g/dl)	2.93 ± 1.09	3.23 ± 1.09	3.07 ± 0.40	2.77 ± 1.32
CHL(g/dl)	2.30 ± 1.15	2.30 ± 0.87	2.20 ± 0.52	2.07 ± 1.31
GLU(g/dl)	0.63 ± 0.05^a	0.93 ± 0.23^b	0.80 ± 0.00^{ab}	0.70 ± 0.17^{ab}
ALT(IU)	96.93 ± 20.51	98.27 ± 6.49	82.90 ± 2.86	89.53 ± 11.84
AST(IU)	13.43 ± 5.25^b	12.80 ± 1.90^b	7.40 ± 2.52^a	9.07 ± 3.09^{ab}
ALP(IU)	10.40 ± 5.81	16.43 ± 2.76	17.17 ± 2.15	13.57 ± 4.11
LDH(g/dl)	119.00 ± 5.31	116.10 ± 25.91	128.57 ± 42.12	99.33 ± 30.79

TP (Total Protein), GLO (Globulin), ALB (Albumin), CHL (Cholesterol), GLU (Glucose) ALT (Alanine aminotransferase), AST (Aspartate aminotranseferase), ALP (Alkaline phosphatase), LDH (Lactate dehydrogenase)

that were enough to impair growth and nutrient utilization. For instance, tannins and phytates contents increased with inclusion levels of processed moringa leaf and kernel in the tested diets. The antinutritional effects of tannins and phytates are well detailed in various reports incorporating plant-derived ingredients in fish diets. Phytates and tannins are known to complex with proteases, minerals and other enzymes when present in fish diets in certain quantities (Francis *et al.*, 2001; Kumar *et al.*, 2011), thus limiting nutrient availability and digestibility. Becker and Makkar (1999) demonstrated that 2 % hydrolysable tannins elicited reduced feed intake, growth and oxygen consumption in common carp. Sajjadi and Carter (2004) also demonstrated an impaired protein digestibility as a result of dietary phytic acid in Atlantic salmon. They further reported an improved protein digestibility when phytase was added to the diet to establish the fact that the cause of the impaired protein digestibility was phytic acid. The view that utilization of mixture of moringa leaf and kernel cake was limited by its antinutritional composition can be further corroborated by the feed intake of the treatment groups that decreased with increase in inclusion of moringa mixture and significantly

lower than the control group. The observed reduced feed intake was probably the most important factor responsible for the observed result. Although, saponins was not assayed in the current investigation, Richter *et al*. (2003) confirmed its presence in moringa leaf; tannins and saponins are known to impact bitter astringent taste to diets thereby lowering palatability (Francis *et al.,* 2001), which could have been the case in the present study. Afuang *et al.* (2003) and Dongmeza *et al*. (2006) have also reported poor feed intake of diets containing saponin and/or tannins in Nile tilapia when fed diets containing moringa leaf. Another possible reason for the poor utilization of the mixture of processed moringa leaf and kernel in the present study could be their poor biological value. Plant proteins are known to be deficient in key amino acid unlike fishmeal proteins that are known to contain balanced mix of essential amino acid profile (Khattab *et al*., 2009). This observation was also corroborated by Madalla *et al*. (2013) who suggested that aqueous extraction of leaf meal could compound the problem of improperly balanced amino acid profile usually associated with plant proteins through leaching of amino acid. Furthermore, Teixeira *et al*. (2014) noted that major portion of moringa leaf protein is insoluble and has low *in vitro* digestibility, a reason that might also be responsible for the low nutritional value of the mixture in the current study.

Anti-nutrients have been shown by many investigators to negatively alter fish health, usually indicated by suboptimal haematology and serum biochemistry. Ogunji *et al*. (2008) attributed decreasing hematological values in *Clarias gariepinus* to increasing antinutritional factors arising from increasing dietary pigeon pea. This was corroborated by Osuigwe *et al.* (2003) who attributed the low values of RBC, WBC, PCV and Hb concentration in African catfish fed raw jackbean seed meal to the anti-nutritional factors inherent in the plant ingredient. Mbahinzireki (1999) and Garcia-Abiado *et al.* (2005) observed that tilapia fed diets containing cotton seed meal protein showed significant decline in PCV and haemoglobin levels. They attributed the result to the effect of gossypol, an anti-nutritional factor peculiar to cottonseed meal. Although, the processed moringa mixture did not significantly alter most of the haematological and serum biochemistry parameters assayed in the present investigation; there tends to be a general decrease in these parameters, especially beyond the 20% supplementation level, further lending credence to the fact that caution must be applied in the use of this mixture beyond 20% level.

Conclusion: Results from this study have shown that soaked moringa leaf and fermented moringa kernel could replace fishmeal at 20% inclusion level, beyond which growth performance and nutrient utilization was compromised. The observed results can be attributed to high levels of anti-nutrient which limited palatability and consequently feed intake and possibly impaired digestibility. There is need for further investigation on means of improving the utilization of moringa leaf and kernel by catfish.

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