E-ISSN: 2347-5129
P-ISSN: 2394-0506
(ICV-Poland) Impact Value: 5.62
(GIF) Impact Factor: 0.549
IJFAS 2018; 6(5): 23-30

www.fisheriesjournal.com
Received: 05-07-2018
Accepted: 06-08-2018

Esien David-Oku
Department of Biochemistry, Faculty of Basic Medical Sciences, University of Calabar, Calabar, Nigeria

Esane Ernest Anani
Department of Biochemistry, Faculty of Basic Medical Sciences, University of Calabar, Calabar, Nigeria

Oluwatosin Elizabeth Ntaji
Department of Biochemistry, Faculty of Basic Medical Sciences, University of Calabar, Calabar, Nigeria

Roseline Okokon Edide
Department of Home Economics, Cross River State College of Education, Akamkpa

Juliet Ifeoma Obiajunwa
Department of Biochemistry, Faculty of Basic Medical Sciences, University of Calabar, Calabar, Nigeria

Henrieta Nkechi Ene-Obong
Department of Biochemistry, Faculty of Basic Medical Sciences, University of Calabar, Calabar, Nigeria

Correspondence
Esien David-Oku
Department of Biochemistry, Faculty of Basic Medical Sciences, University of Calabar, Calabar, Nigeria

Growth performance and nutritional impacts of *Moringa oleifera* Leaf and shrimp meals supplemented diets on *Clarias gariepinus* (African Catfish)

Esien David-Oku, Esane Ernest Anani, Oluwatosin Elizabeth Ntaji, Roseline Okokon Edide, Juliet Ifeoma Obiajunwa and Henrieta Nkechi Ene-Obong

Abstract
Objective: This study evaluated the impact of graded dietary inclusion of *Moringa oleifera* leaf meal and shrimp meal on growth performance and nutritional quality of the African catfish.
Method: Fish diets were formulated (M_0, M_{10}, M_{20} and M_{30}) to contain 0, 10, 20, and 30% of *Moringa oleifera* leaf meal (MLM) respectively, with 20% of shrimp meal added to each as animal protein source. Two commercial fish feeds, foreign (Coppens™) and local (Vital™), were taken as controls. *Clarias gariepinus* fingerlings (N= 180) were randomly distributed into 18 plastic tanks (n=10); 3 tanks each for the six respective formulated and commercial diets. Feeding was twice daily for 6 weeks. Feed intake, growth performance, and nutritional quality of the fish were assessed using standard methods.
Results: Food intake, protein efficiency ratio, total weight gain and specific growth rate were in the order of $M_{10} > M_{20} > M_0 > M_{30}$. M_{10} fishes were richer in crude fat and protein (25.00, 48.03% respectively) than M_0 (18.49, 46.18%), M_{20} (20.41, 46.07%) and M_{30} (18.80, 45.38%); but similar to the control groups (26.25 - 26.76; 50.60 - 51.97%). Crude fibre and carbohydrate contents of the MLM fishes were significantly greater ($p<0.05$) than the controls but within the group, there were dose dependent increases. Total ash seemed similar (> 10.00%) in all the groups under study.
Conclusion: The results of this study show that dietary supplementation of 10% *Moringa oleifera* leaf plus 20% shrimp meals is optimal for effective growth performance and nutrient quality in African catfish.

Keywords: *Clarias gariepinus*, African catfish, *Moringa oleifera* leaf meal, shrimp meal, growth performance, nutrient quality

1. Introduction
Fish farming also known as aquaculture plays a major role in economic development and foreign exchange earnings of nations [1]. In Nigeria, artificial fish farming is practiced in earthen, concrete and tarpaulin (collapsible) ponds, thus providing facilities that enhance fish growth and development. Several studies have been done on various fish species with the ultimate aim of improving growth, quantities and qualities as well as preventing diseases [2]. The African catfish has been found to be very suitable for aquaculture in Africa [3] for several reasons: it grows fast and feeds on a large variety of agriculture byproducts; it is hardy and tolerates adverse water quality conditions; can be raised in high densities, resulting in high net yields [4].
One of the major challenges of fish farming in most parts of Nigeria is the high cost of feeds (both local and imported); the imported foreign feeds being the most preferred because of better outcome. This of course is a function of high protein quality and quantity in the foreign feeds.
For this reason, farmers prefer to start with foreign feeds, grow and finish with local feeds/supplemented diets. High cost and scarcity of fish meal for on-farm production of supplementary feeds has opened a great door for the exploitation of non-conventional feedstuffs from animal and plant sources [5]. *Moringa oleifera* leaf meal (MLM) and shrimp meal have become choice candidates in several animal feed studies [6-8].

M. oleifera, a member of the Moringaceae family, is one of the most useful trees in the world with all the parts having nutritional and pharmacological properties [9]. The leaves are rich in protein, vitamins and mineral nutrients and thus widely eaten by humans and animals. Most studies have shown decreased performance of broilers fed *Moringa* leaf meal (MLM) with inclusion levels as low as 5-7.5% [6-8]. Similar findings have been reported with higher inclusion level of MLM in formulated diets for fish. [6, 7, 10, 11] Hence, only limited amounts of *Moringa* leaf meal can be safely used in fish diets. Previous studies [8, 12, 13] showed that the inclusion of *M. oleifera* leaf meal above 10% resulted in lower specific growth rate, feed conversion ratio, protein efficiency and also poorer carcass composition of Nile tilapia. Another study [14] reported that up to 20% inclusion did not affect feed composition ratio and protein efficiency in the diet of African catfish. However, higher inclusion levels caused increase in serum enzymes which suggests some cellular damage [15].

Shrimp meal has been reported to be valuable feedstuffs in dairy cow, [16] lambs, [17] pigs, [18] and also in replacing fish meal or soy bean meal in broilers diet. [18, 19] It has been reported to be a very good growth stimulant in *Clarias batrachus* and *Macrobrachium rosenbergii* because of its high crude protein content [20]. According to Nwanna [21], chemically preserved shrimp meal can profitably replace fishmeal at about 20% in African catfish (*Clarias gariepinus*) diet. It has been reported [22] that shrimp meal can be included in the diet of blue tilapia (*Oreochromis aureus*) up to 6% with no negative impact on growth performance. More so, higher inclusion levels of shrimp meal in the diet of fish have been implicated in compromised growth rate, feed conversion ratio and protein efficiency ratio [23]. There is a lack of information on the nutritional response of African catfish to a combination of MLM and shrimp meal, considered as cheap sources of protein in fish feed formulations. Recently, we formulated fish diets using a combination of graded doses of *Moringa oleifera* leaf meal (MLM) and a fixed proportion (20%) of shrimp meal as protein sources [24]. In this study we evaluated the effects of these MLM and shrimp meal supplemented diets on feed intake, growth performance and nutritional quality of African catfish, *Clarias gariepinus*.

2. Materials and Methods

2.1 Collection of materials

Moringa leaves (*Moringa oleifera* Lam.) were collected from a moringa farm located at Obudu Local Government Area of Cross River State; identified and deposited in the herbarium of the Department of Plant and Ecological Studies in the University of Calabar, Calabar with voucher number P.E.S/Herb/UC/395. Shrimp waste/dust was collected from traders (women) at Nsidung Beach market, Calabar South Local Government Area of Cross River State, Nigeria. Foreign (Coppens^{TM}) and local (Vital^{TM}) commercial feeds used as controls (C_1 and C_2, respectively) in this study, were purchased from a local vendor in Calabar South Local Government Area of Cross River State, Nigeria.

2.2 Processing of *Moringa oleifera* leaf and shrimp meals

Five hundred (500) grams of freshly harvested leaves were washed and air-dried under shade until they were crispy to touch while still retaining their greenish coloration. The leaves were then threshed to strip off dry leaves from stalks to reduce the crude fiber content in the meal. The dried leaves were ground into a fine powder using a hammer mill. The resulting Moringa leaf meal (MLM) was sieved and stored in plastic bags at room temperature until when needed. The shrimp waste/dust was oven dried at 45^{o}C for 4 hours and thereafter, ground into fine powder and stored in tight plastic container for further use.

2.3 Fish diet formulation and processing

Four different diets were prepared using Pearson's method of fish feed formulation to contain 40% crude protein; 20% shrimp meal across board, then 0, 10, 20, and 30% MLM to give M0, M10, M20, and M30 diets, as earlier reported [24].

2.4 Catfish feeding protocol

The research was conducted at the fish farm site of the Institute of Oceanography, University of Calabar, Nigeria. One hundred and eighty (180) African catfish fingerlings (*C. gariepinus*) of average weight 2.60 g were purchased from University of Calabar Fish Farm, Calabar, Nigeria. They were sorted into eighteen (18) rectangular plastic tanks (n=10). The fish were allowed to acclimatize for 7 days during which they were fed on commercial diet. Prior to the commencement of the experiment, all fish were starved for 24 hrs to eliminate food residue in the gut and to increase their appetite. The initial and weekly weight and length of the fingerlings were measured with Mettler electronic weighing balance and a graduated rule respectively. The MLM supplemented diets (M0, M10, M20 and M30), and commercial feeds ($C_1$and C_2) groups were fed to the respectively designated experimental groups. The fish were fed by hand (5% of body weight) daily between 8:00hrs-9:00hrs and 16:00hrs-17:00hrs. The quantities of feed fed were adjusted every two weeks according to the body weight of the fish. They were fed to satiation and the remaining uneaten feed were collected and weighed to estimate actual feed intake.

2.5 Determination of feed intake and growth performance

The growth and nutrient utilization were determined in terms of feed intake (FI), protein intake (PI), percentage relative weight gain (%RWG), percentage relative length gain (% RLG), specific growth rate (SGR), feed conversion ratio (FCR), protein efficiency ratio (PER) and survival rate (SR) [25] as follows:

$$\text{FI } (gfish^{-1}day^{-1}) = \frac{\text{Total feed intake per fish}}{\text{Number of days}}$$

$$\text{PI } (gfish^{-1}\,day^{-1}) = \text{Feed intake x \% protein in diet}$$

$$\%RWG = \frac{\text{Final weight-initial weight}}{\text{Initial weight}} \times 100$$

$$\%RLG = \frac{\text{Final length-initial length}}{\text{Initial length}} \times 100$$

$$FCR = \frac{\text{Feed intake (g)}}{\text{Total weight gain (g)}}$$

$$SGR = \frac{\text{Log}_n\,(\text{Final Body Weight}) - \text{Log}_n\,(\text{initial bodyweight})}{\text{Number of days}} \times 100$$

$$PER = \frac{\text{Net weight gain}}{\text{Protein Intake}}$$

$$SR = \frac{\text{Initial number of fish started-mortality x 10}}{\text{Initial number of fish}}$$

2.6 Water quality parameters
Dissolved oxygen, pH, temperature and CO_2 of the water were monitored weekly using water checker.

2.7 Harvesting/ sacrifice of fish
At the end of the experiment, the fish tanks were completely drained. The fish were collected with a net with handle, and sacrificed. Afterwards, they were dried in an oven.

2.8 Proximate analysis on fish flesh
Proximate analysis on the fish flesh (for moisture, crude protein, crude lipid, ash and essential amino acid composition) was done using standard methods. [26, 27] and results expressed as percentage of live weight.

2.9 Amino acid analysis
Amino acid analysis was done accordingly [28]. Samples were hydrolysed with 6N hydrochloric acid in vacuum at 1100C for 24hrs. Hydrolysed amino acids were quantified using LKB Biochrom Ltd, UK. The amino acid analyser was calibrated using a standard solution (AA-5-18, sigma). The prepared sample was loaded into ion exchange column. The amino acids were sequentially eluted by buffers of varying pH and ionic strength. The acidic amino acids were first removed, followed by neutral amino acids. Once separated, individual amino acids were quantified by reaction with nihydrin and resultant colour intensity was measured by spectrophotometer at 570nm. The colour intensity was directly proportional to the quantity of an amino acid present in the sample. Amino acids were identified by comparing peak retention times to the standard and quantification was done by comparing area under peak to the area under the standard.

2.10 Statistical analysis
All data collected were analyzed using statistical analysis system software (SPSS, 1995 Version 9 for WINDOWS). One way analysis of variance and Duncan's multiple range tests was used to compare between parameters of the different feeding groups.

3. Results
3.1 Feed tanks water quality assessment
Dissolved oxygen (DO) and CO_2 were significantly (p<0.05) lower and higher respectively, in the MLM (M_0, M_{10}, M_{20}, M_{30}) tanks than the C_1 and C_2 tanks (Table. 1). The pH of the MLM tanks were also significantly (p<0.05) lower while the temperatures were similar.

Table 1: Fish tank water quality parameters

Sample	Dissolved oxygen (mg/L)	Temperature (ºC)	pH	CO_2 (mg/L)
C_1	6.00	28.33	7.80	0.03
	±0.12	±0.33	±0.12	±0.01
C_2	5.77	28.67	7.57	0.02
	±0.12	±0.33	±0.12*	±0.00*
M_0	4.70	29.00	6.97	0.03
	±0.06*,a	±0.00	±0.12*,a	±0.00
M_{10}	4.40	28.67	6.77	0.05
	±0.00*,a	±0.33	±0.09*,a	±0.01a,b
M_{20}	4.50	29.00	6.60	0.05
	±0.06*,a	±0.00	±0.06*,a	±0.00a,b
M_{30}	4.63	28.67	6.73	0.05
	±0.07*,a	±0.33	±0.12*,a	±0.00a,b

Values are expressed as mean ±SEM of 3 determinants
* = significantly different from C1 at p<0.05
a = significantly different from C2 at p<0.05
b = significantly different from M0 at p<0.05

3.2 Feed Intake of fish placed on different experimental diets.
Except for C_1 with food intake of 14.13 gfish^{-1}day^{-1}, all the other groups had similar food intake (13.60 - 14.07 gfish^{-1}day^{-1}) which was significantly lower (p<0.05) than C_1 (Table. 2). Among the MLM groups, there was a dose dependent reduction in feed intake, with M_{10} having the highest. Protein intake was significantly lower in the MLM supplemented groups (1.63- 2.02 gfish^{-1}day^{-1}) than the commercial feeds groups (7.11-7.43 gfish^{-1}day^{-1}). As with food intake, there was dose dependent reduction in protein intake among the MLM groups with M10 (2.02 gfish^{-1}day^{-1}) being the highest. Surprisingly, M_0 had a superior protein intake (2.12 gfish^{-1}day^{-1}) than the MLM supplemented groups. But in feed conversion ratio (FCR), M_0 and M_{20} were similar; while M_{10} and M_{30} were better and worse, respectively, than M_0. PER of M_{10} (0.90) was significantly greater (p<0.05) than those of M_0, M_{20}, M_{30}, C_1 and C_2 (0.61, 0.72, 0.61, 0.67, and 0.69), respectively. The survival rates of the fish on M_0 and M_{10} were the same (86.67%) and higher than those of M_{20} (83.33%) and M_{30} (76.67%). The commercial feeds C_1 and C_2 recorded 100% survival rate.

3.3 Growth performance of fish placed on different experimental diets
Total weight gain (TWG), relative weight change (RWC), relative length change (RLC), and specific growth rate (SGR) of the fishes fed with commercial feeds were significantly greater (p<0.05) than those on MLM supplemented diets (Table 3). However, there was a dose dependent significant reduction in those parameters from M_{10} to M_{30}, with M_{30} being worse than M_0. Thus M_{10} had the best profile (TWG, 1.87g; RWC, 74.94%; RLC, 65.00%; and SGR, 1.32%/day) among the MLM groups. The average weekly body weight (Figure 1) and length (Figure 2) amply illustrate the disparity in growth performance between the fish on commercial diet and those on MLM supplemented diets.

Table 2: Feed consumption parameters and survival rate of fish placed on different experimental diets.

	Food intake ($gfish^{-1}day^{-1}$)	Protein intake ($gfish^{-1}day^{-1}$)	Feed conversion ratio	Protein efficiency ratio	Survival rate (%)
C_1	13.92	7.43	2.80	0.67	100.00
	±0.22	±0.12	±0.0	±0.01	±0.00
C_2	14.13	7.11	2.88	0.69	100.00
	±0.10	±0.09*	±0.02*	±0.01	±0.00
M_0	13.64	2.12	10.53	0.61	86.67
	±0.12[a]	±0.03*,[a]	±0.44*,[a]	±0.03*,[a]	±3.33*,[a]
M_{10}	14.07	2.02	7.56	0.90	86.67
	±0.13	±0.01*,[a]	±0.25*,[a,b]	±0.05*,[a,b]	±3.33*,[a]
M_{20}	13.67	1.86	10.30	0.72	83.33
	±0.05[a]	±0.04*,[a,b]	±0.54*,[a]	±0.04[c]	±3.33*,[a]
M_{30}	13.60	1.63	13.69	0.61	76.67
	±0.01[a]	±0.0*,[a,b]	±0.80*,[a,b]	±0.04	±3.33*,[a,b]

Values are expressed as mean ±SEM of 3 determinants
* = significantly different from C1 at $p<0.05$
a = significantly different from C2 at $p<0.05$
b = significantly different from M_0 at $p<0.05$

Table 3: Growth performance parameters of fish placed on the different experimental diets

Sample	Total weight gain (g)	Rel. weight Change (%)	Rel. length Change (%)	Specific growth rate (%/day)
C_1	4.80	196.20	133.20	2.61
	±0.00	±5.91	±7.59	±0.06
C_2	4.90	181.96	125.50	2.46
	±0.06	±6.87	±14.51	±0.06
M_0	1.30	49.44	23.67	0.96
	±0.06*,[a]	±5.30*,[a]	±2.17*,[a]	±0.08*,[a]
M_{10}	1.87	74.97	65.00	1.34
	±0.07*,[a,b]	±0.03*,[a,b]	±8.19*,[a]	±0.01*,[a,b]
M_{20}	1.33	52.40	28.77	1.00
	±0.07*,[a]	±4.82*,[a]	±1.57*,[a]	±0.08*,[a]
M_{30}	1.00	40.72	24.20	0.81
	±0.06*,[a,b]	±3.28*,[a]	±4.91*,[a]	±0.05*,[a]

Values are expressed as mean ±SEM of 3 determinants
* = significantly different from C_1 at $p<0.05$
a = significantly different from C_2 at $p<0.05$
b = significantly different from M_0 at $p<0.05$

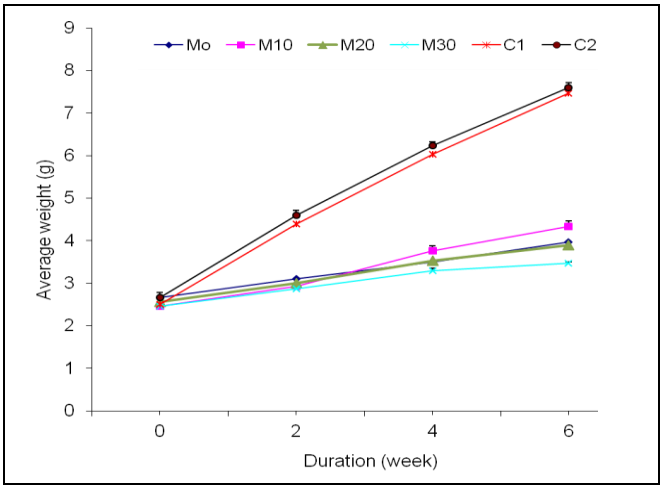

Fig 1: Average Weekly body Weights Of fish Placed On the different Experimental fish Diets

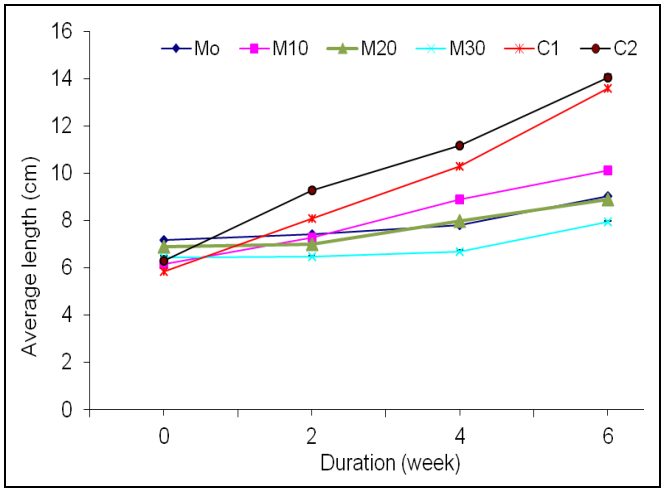

Fig 2: Average Weekly body Length of fish Placed on the different experimental fish diets

3.4 Proximate composition of experimental fish samples

There was no significant difference in mean contents (%) of crude protein and crude fat between M_{10} (48.02; 25.00, respectively) and the foreign feed (C_1) (51.97; 26.25, respectively) fed fishes (Table 4). M_{20} and M_{30} were similar with M_0 in protein and fat content (45.38 - 46.18; 18.49 - 20.41, respectively). The reverse was the case with crude fibre and carbohydrate where the MLM groups recorded significantly higher contents (0.83 – 2.20; 5.03 – 12.53, respectively) than the C groups (0.20; 1.88 - 2.49, respectively). There was a dose dependent increase in these parameters between M10 – M30. Total ash content of all the groups was < 10%, while moisture content was 9.52 – 10.98%.

Table 4: Proximate (%) composition of the different experimental fish samples

Sample	Moisture	Crude fat	Crude protein	Crude fibre	Total ash	CHO
C_1	9.52	26.25	51.97	0.20	10.18	1.88
	±0.28	±0.80	±0.52	±0.00	±0.05	±0.27
C_2	9.52	26.76	50.60	0.20	10.43	2.49
	±0.33	±0.44	±0.59	±0.00	±0.30	±0.47
M_0	9.76	18.49	46.18	0.21	10.69	17.73
	±0.15	±0.06*,a	±0.09*	±0.01	±0.03*	±2.99*,a
M_{10}	10.21	25.00	48.03	0.83	10.94	5.03
	±0.17	±0.00a,b	±1.93	±0.07*,a,b	±0.55	±1.91b
M_{20}	10.98	20.41	46.07	1.03	10.49	10.98
	±0.05*,a,b	±1.17*,a	±1.10*	±0.01*,a,b	±0.01*	±1.64*,a
M_{30}	9.94	18.80	45.38	2.20	10.52	12.52
	±0.17	±0.24*,a	±0.69*	±0.12*,a,b	±0.07*	±0.38*,a

Values are expressed as mean ±SEM of 3 determinants
* = significantly different from C_1 at $p<0.05$
a = significantly different from C_2 at $p<0.05$
b = significantly different from M_0 at $p<0.05$

3.5 Amino acid content of experimental fish samples.

Among the MLM supplemented groups, M_{10} had the highest content of all the amino acids evaluated (except for Phe and His) which were in most cases, similar to C_1 and C_2 groups (Table 5). The richest content of His was found in M_0 (14.38%) with M_{10} - M_{30} having values < 10% (8.41 - 9.93%).

Table 5: Selected amino acid composition (%) of the different experimental fish samples.

Sample	Methionine	Threonine	Isoleucin	Phenylanine	Alanine	Valine	Leucine	Tryptophan	Lysine	Histidine
C_1	30.10	13.09	14.96	6.59	16.87	11.76	10.93	3.57	17.58	14.02
	±1.58	±0.47	±0.65	±0.16	±1.45	±0.26	±1.12	±0.27	±1.97	±1.18
$=C_2$	28.11	11.90	13.08	6.19	18.06	12.04	11.14	3.62	17.83	14.01
	±1.20	±0.95	±1.24	±0.64	±0.31	±0.01	±0.64	±0.24	±1.55	±1.00
M_0	6.61	8.47	1.27	7.59	13.35	8.98	7.27	2.99	13.79	14.38
	±0.24*,a	±0.57*,a	±1.06*	±0.55	±1.49a	±0.18*,a	±0.90a	±0.42	±1.64	±0.38
M_{10}	18.47	11.95	13.85	7.46	16.43	11.38	9.88	3.13	14.65	9.09
	±0.33*,a,b	±0.94b	±1.33	±0.56	±1.41	±0.23a,b	±0.91	±0.64	±2.23	±1.94
M_{20}	5.91	9.06	10.62	5.10	12.38	7.93	6.77	2.60	12.68	8.41
	±0.72*,a	±0.46*	±0.32*	±0.79	±1.48	±0.27*,a,b	±0.96*,a	±0.50	±1.63	±1.95b
M_{30}	5.99	8.19	10.16	6.44	12.55	9.00	6.94	3.02	13.67	9.93
	±0.80*,a	±0.13*,a	±0.39*	±1.01	±0.99a	±0.48*,a	±0.89*,a	±0.45	±0.98	±4.43

Values are expressed as mean ±SEM of 3 determinants
* = significantly different from C_1 at $p<0.05$
a = significantly different from C_2 at $p<0.05$
b = significantly different from M_0 at $p<0.05$

4. Discussion

The relatively poorer tank water quality of the MLM groups (significantly lower pH and dissolved oxygen, and higher CO_2) obtained in this study is attributable to the fact that the formulated feeds being non-floating, would normally sink to the bottom of the tank when not eaten immediately by the fish. The resulting water contamination is capable of compromising growth performance of the fish [29] and may also have contributed to 13 – 23% mortality observed among the MLM groups. However, the sinking nature of the MLM diets seemed not to have affected the feed intake of the fishes as there was no significant difference in this parameter among the groups when compared to the foreign commercial feed group.

The poor growth performance of the fish fed with MLM and shrimp meal (20%) supplemented diets used in this study is attributable to low crude protein and high crude fiber content of the diets [24]. The protein contents of non-conventional plant feedstuff in aquaculture have been reported to be generally low [30, 31]. Significantly high crude fiber and anti-nutrient content in diets result in slower growth rate and general poor performance of cultured species [32]. This is similar to an earlier observation of a significant decline in final weight gain with increasing level of MLM in the diet of broiler chickens [6]. High fibre content reduces digestibility, palatability and nutritive value of feeds [33, 34], all of which eventually compromise growth performance of fish.

Growth of fish is dependent upon the nutrient quality and quantity of ingredients in feeds. [35, 36] An average crude protein of 35% in diets is recommended for optimum

performance of *C. gariepinus* fingerlings [5, 37]. The MLM and shrimp meal formulated diets used in this study had crude protein levels of 12 - 14%, a far cry from > 50% provided for in the formulation [24]. The high fibre (from MLM) and chitin (from shrimp meal) are reported to limit protein availability in fish diets [24, 38]. Adding MLM extract and further digestion of shrimp meal [21, 39] may be a way to reduce the fibre and chitin levels respectively, in the diets thus making for better protein nutrient availability.

The results of our study show that locally manufactured commercial feeds compete favorably with foreign commercial feeds and can therefore serve as an adequate alternative in catfish nutrition, giving similar performance with high cost benefit. This is in agreement with previous studies. [33, 34]

The mortality level reported in this study is greater than 10% for the MLM groups. It increased with increase in MLM supplementation. This may not be unconnected with increasing fibre and anti-nutritional factors present in MLM. [40, 41] Increased mortality with higher inclusion levels of cassava leaf meal in the diet of *C. gariepinus* has also been reported [42].

The results of our study also show that 10% MLM supplementation in fish diet provided optimum growth performance when compared with higher levels. This is in agreement with other studies [7, 43] which also encouraged MLM supplementation in local fish feed manufacturing.

The nutrient quality of the fish (flesh) fed with MLM and shrimp meal supplemented diets are comparable with those raised with commercial feeds; similar in crude fat and protein, and higher in carbohydrate and crude fibre. The levels of essential amino acids (all except Alanine) obtained in this study in the MLM fish groups are quite impressive. Of the ten amino acids, only Methionine was found to be significantly lower in the MLM groups than the commercial feed groups. This is in spite of the 0.2% supplementation in the feed formulation [24]. Again M_{10} had the best nutrient profile among the MLM groups. This study recommends the consumption of MLM fed fish for dietary management of cardiovascular disease risk factors in human subjects as intake of omega-3 (n-3) fatty acids and high fibre diets are known to improve high density lipoprotein cholesterol (HDL-C) and control diabetes mellitus [44].

4. Conclusion

The results of this study have shown that dietary inclusion of 10% *M. oleifera* leaf meal (plus 20% shrimp meal) offered optimum growth performance and excellent nutrient quality to the flesh of African catfish, *Clarias gariepinus.* The study recommends further processing of MLM and shrimp meal before incorporation into fish meals to enhance protein availability. This way, the widespread availability of *Moringa oleifera* and shrimp meal (from artisanal fishing and sales) will be taken advantage of in the Nigerian aquaculture industry.

5. Acknowledgment

We are grateful to Mr. Michael Ategwu of Cross River State School of Health Technology, Calabar, and Mr. Igiri I. Igiri of Faculty of Agriculture, University of Calabar, for technical assistance.

6. Authors' Contribution

David-Oku and Anani conceived and designed the study; Ntaji and Edide developed the methodology; Anani and Edide performed the bench work; Obiajunwa-Otteh analyzed and interpreted the data; David-Oku and Anani wrote and reviewed the manuscript; while Ene-Obong revised the draft manuscript. Overall study supervision was by David-Oku.

7. References

1. Adeogun OA, Ogunbadejo HK, Ayinla OA, Oresegun A, Oguntade OR, Tanko A *et al*. Urban Aquaculture: producer perceptions and practices in Lagos state, Nigeria. Middle East Journal of Science Research. 2007; 2(1):21-27.
2. Bard J, De Kimpe PJ, Lazard J, Lamasson J, Lessent P. Handbook of tropical fish culture centre. Technique Forestier Tropical, France. 1976, 128.
3. Anon R. The state of the Worlds fisheries and aquaculture. FAO fisheries Technical, 2006, 500.
4. Aderolu AZ, Seriki BM, Apatira AL, Ajaegbo CU. Effects of feeding frequency on growth, feed efficiency and economic viability of rearing African catfish (Clarias gariepinus, Burchell 1822) fingerlings and juveniles. African Journal of Food Science. 2010; 4(5):286-290.
5. Pandey G. Feed formulation and feeding technology for fishes. International Research Journal of Pharmacy. 2013; 4(3):23-30.
6. Zanu HK, Asiedu P, Tampuori M, Abada M, Asante I. Possibilities of using moringa (*Moringa oleifera*) leaf meal as partial substitute for fish meal in broiler chicken diets. Online Journal of Animal Feed Research. 2012; 2(1):70-75.
7. Oludoyi IO, Toye AA. Effect of early feeding of moringa on performance of broilers and pullet chicks. Agricultural Research. 2012; 12(2):160-17.
8. Egwui PC, Mgbenka BO, Ezeonyejiaku CD. Moringa plant and its use as feed in aquaculture development: a review. Journal of Animal Research international. 2013; 10(1):1672-1680.
9. Santos AFS, Paiva1 PMG, Coelho LCBB. Moringa oleifera: a multipurpose tree with water coagulant seed proteins. In: Advances in Environmental Research, Daniels. J. A. (ed). Nova Science Publishers, Inc. 2012, 17
10. https://www.researchgate.net/publication/262829037_Moringa_oleifera_a_multipurpose_tree_with_water_coagulant_seed_proteins [accessed Aug 07 2018].
11. Afuang W, Siddhuranju P, Becker K. Comparative Nutritional Evaluation of Raw Methanol extract residues and methanol extract of Moringa (*Moringa oleifera* Lam) leaves on growth performance and feed utilization in Nile Tilapia (*Orcochromis noloticus* L.). Journal of Aquaculture Research. 2003; 34:1147-1159.
12. Gakuya DW, Mbugua PN, Kavoi B, Kiama SG. Effect of supplementation of *Moringa oleifera* leaf meal in broiler chicken feed. International Journal of Poultry Science. 2014; 13(4):208-213.
13. Richter N, Siddhrurayu A, Becker K. Evaluation of nutritional quality of moringa (*Moringa oleifera* Lam). Leaves as alternate protein source of tilapia (*Oreochromis Niloticus* L.). Aquaculture. 2003; 217:599-611.
14. Abo-state H, Hammounda Y, El-nadi A, Abozaid H. Evaluation of feeding raw moringa (*Moringa oleifera* Lam) leaves meal in Nile Tilapia fingerlings (*Oreochromis niloticus*) diets. Global Veterinary. 2014; 13(1):105-111.

15. Olaniyi CO, Ajani NO, Adetomi MN. Growth performance and nutrient utilization of *Clarias gariepinus* fed *Moringa oleifera* leaf meal. Journal of National Sciences Research. 2013; 3(8):99-104
16. Bello NO, Nzeh GC. Effects of varying levels of Moringa oleifera leaf meal diet on growth performance, hematological indices and biochemical enzymes of African catfish *Clarias gariepinus* (Burchell 1822). Elixir Aquaculture. 2013; 47A:14459-14466.
17. Lush RH. Shrimp meal for milk production. American Society of Animal Production. 1936; 1:63-66.
18. Cobos MA, Pérez-Sato M, Piloni-Martini J, González SS, Barcena JR. Evaluation of diets containing shrimp shell waste and an inoculum of Streptococcus milleri on rumen bacteria and performance of lambs. Animal Feed Science and Technology. 2007; 132(3, 4):324-330.
19. Fanimo AO, Oduguwa OO, Onifade AO, Olatunde AO. Protein quality of shrimp waste meal. Bioiresource Technology. 2000; 72(2):185-188.
20. Rosenfeld DJ, Gernet AG, Marcano JD, Murillo JG, Lopez GH, Flores JA. The effect of using different levels of shrimp meal in broiler diets. Poultry Science. 1997; 76(4):581-587.
21. Hertramf JW, Piedad-Pascal F. Sunflower meal. International Handbook on ingredients for aquaculture feeds. Dordrecht Netherlands: Kluewer Academic Publishers, 2000, 69-79.
22. Nwanna LC. Nutritional value and digestibility of fermented shrimp head waste meal by African catfish *Clarias gariepinus*. Pakistan Journal of Nutrition. 2003; 2(6):339-345.
23. De Silva SS, Shim KF, Ong AK. An evaluation of the methods used in digestibility estimations of a dietary ingredient and comparisons on external and internal markers and time of faeces collection in digestibility studies in fish Oreochromis aureus (Steindachner). Journal of Reproduction and Nutritional Development. 1996; 30:215-226.
24. Rachmansyah LA, Ahmad T. The use of shrimp head meal as a substitute to fishmeal in diets for humpback grouper (*Cromileptes altivelis*). In: Advances in grouper aquaculture. Rimmer, M. A., McBride, S., Williams, K. C. (Eds.). 2004, 133-114.
25. David-Oku E, Anani EE, Ntaji OE, Edide RO, Obiajunwa JI. Proximate, mineral and vitamin composition of fish feed supplemented with *Moringa* oleifera leaf and shrimp meals as protein sources. European Journal of Pharmaceutical and Medical Research. 2018 (in press).
26. Cook JT, McNiven MA, Richardson GF, Sutterlin AM. Growth rate, body composition and feed digestibility, conversion of growth-enhanced transgenic Atlantic salmon (*Salmo salar*). Aquaculture. 2000; 188:15-32.
27. AOAC. Official methods of analysis.16th Edition. Association of Official Analytical Chemists. Washington, DC, 2005.
28. Ibitoye AA. Laboratory Manual on Basic Methods in Analytical Chemistry. Foladave Nig Ltd, Akure, 2010.
29. Neda I, Vlazan P, Pop RO, Sfarloaga P, Grozescu I, Segneanu A. Peptide and Amino Acids Separation and Identification from Natural Products. In: Analytical Chemistry (Krull, I. S., ed.) IntechOpen, London, 2012. DOI: 10.5772/51619.
30. Bhatnagar A, Devi P. Water quality guidelines for the management of pond fish culture. International Journal of Environmental Sciences. 2013; 3(6):1980-2009.
31. Ugwumba AAA, Ugwumba AO. Aquaculture options and the future of fish supply in Nigeria. The Zoologist. 2003; 2:96-122.
32. Gabriel UU, Akinrotimi OA, Bekibele DO, Onunkwo DN, Anyanwu PE. Locally produced fish feed, potentials for aquaculture development in sub-Saharan Africa. Journal of African Agricultural Research. 2007; 2(7):287-295.
33. Alegbeleye WO, Oresegun AO, Omitoyin O. Use of bambara groundnut (*Vigna subterranean*) meal in the diets of *Heteroclarias* fingerlings. Moor Journal of Agriculture Research. 2001; 2:54-59.
34. Agokei EO, Oparah C, Aranyo A, Apapa U. Growth of *Clarias gariepinus* juveniles fed five commercial feed continental. Journal of Fisheries and Aquaculture Science. 2011; 5(3):1-5
35. Auta J, Yashim YE, Dambo A, Tiamiyu BB. Growth responses of African catfish, *Clarias gariepinus*, fingerlings to imported and local feeds. International Journal of Applied Biological Research. 2013; 5(1):55-61.
36. Glencross BD, Booth M, Akam GL. A feed is only as good as its ingredients: A review of ingredients evaluation strategies for aquaculture feeds. Aquaculture Nutrition. 2007; 62:100-134.
37. Sogbesan AO, Ugwumba AAA. Nutritional evaluation of termite *Macrotermes subhyalinus.* Meal as animal protein supplements in the diets of *Heterobranchus longifilis* fingerlings. Journal of Fisheries and Aquatic science. 2008; 8:149-157.
38. Amisah S, Oteng MA, Ofori JK. Growth performance of the African catfish, *Clarias gariepinus*, fed varying inclusion levels of *Leucaena leucocephala* leaf meal. Journal of Applied Science and Environmental Management. 2009; 13(1):21-26.
39. Fox CJ, Blaw P, Brown JH, Watson I. The effects of various processing methods on the physical and biochemical properties of shrimp head meals and their utilization by juvenile Penaeus monodon Fab. Aquaculture. 1994; 122:209-226.
40. Bhuiyan AKM. Survey of potential fish feed ingredients of Bangladesh on the basis of their availability and biochemical composition. Fisheries Research Institute, Freshwater Station, Mymensingh, Bangladesh, Report, 1989; 1:70.
41. Oresegun A, Alegbeleye WO. Growth response and nutrient utilization of tilapia *Oreochromis niloticus* fed varying dietary levels of cassava peels based on rations supplemented with di-methionine Fish Nutrition and Fish Feed Technology in Nigeria. In: A. A. Eyo, (ed) Proceedings of the first National Symposium on Fish Nutrition and Fish Feed Technology Lagos NIOMR. 2001, 8-13.
42. Ibiyo LMO, Olowosegun T. The potential for improving profitability in Aquaculture In: P. A. Araoye (ed). Proceedings of the 19th Annual Conference of the Fisheries Society of Nigeria (FISON) ILORIN. 2004; 896:45-53.
43. Anyanwu DC, Umeh OI, Ugo CK, Maduka BC, Mbachu MU. Nutrient utilization and growth of catfish (*Clarias gariepinus*) fed dietary levels of cassava leaf meal. Pakistan Journal of Nutrition. 2012; 11(11):1037-1040.

44. Ozovehe BN. Growth performance, haematological indices and some biochemical enzymes of juveniles *Clarias gariepinus* (Burchell 1822) fed varying leaves of Moringa oleifera leaf meal diet. Aquatic Research and Development. 2013; 4:166.
45. Raatz SK, Silverstein JT, Jahns L, Picklo Sr, MJ. Issues of Fish Consumption for Cardiovascular Disease Risk Reduction. Nutrients. 2013; 5:1081-1097. Doi:10.3390/nu5041081.