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Performance of African Catfish (*Clarias gariepinus)* juvenile fed *Morinda lucida* (Oruwo leaf)

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Target Audience: *Researchers, Animal Scientists, Fish farmers, Fish breeders*

Abstract

Many medicinal plants including Morinda lucida (Oruwo leaf) have shown promising findings when included in aquaculture rations as a feed additive. Growth and survival of Clarias gariepinus juveniles fed with a Morinda lucida feed were observed for 8 weeks. The proximate composition, growth performance, economics of the feeds and water quality of the cultured tanks were assessed. Fish fed with T2 (25% Morinda lucida) showed significant (P<0.05) higher weight increase, specific growth rate, protein efficiency ratio and low feed conversion ratio than other groups. Significant (P<0.05) higher mortality was recorded in fish fed with T5 (100% Morinda lucida) inclusion. The growth performance was a reflection of the proximate composition of the feeds with T5 (100% Morinda lucida) feed having low crude protein (15.14%), ether extract (3.64%) and ash (2.31%) when compared to T2 (25% Morinda lucida) which had 25.11% crude protein, 5.77% ether extract and 4.87 % ash with protein being most significant. Carbohydrate (61.00%) and crude fibre (6.16%) were higher in T5 feed than in T2 with an imbalance in carbohydrate and lipid ratio. Mortality was attributed to stress resulting from the poor quality of the feed. Cost of feeding with T5 (100% Morinda lucida) inclusion to a weight gain of 48.20g was ₦2000, while the cost of feeding with T2(25% Morinda lucida) to a weight gain of 84.87g was ₦2350. It was concluded that Morinda lucida inclusion could be incorporated into the diet of Clarias gariepinus up to 25% without posing health hazard on the fish.

Key words: *Morinda lucida, Clarias gariepinus juveniles, Growth Performance, feed per grain*

Description of Problem

Catfish (*Clarias gariepinus*) are the major commercially cultured species in Nigeria for good market reasons. Since 2000 there has been a rapid expansion in urban aquaculture and a significant development in high density catfish culture. As a result of this intensification in catfish production, the aqua feed industry grew and moved from the days of research in fish nutrition and fish diet started at NIOMR (Nigerian Institute for Oceanography and Marine Research) where a laboratory size pellet mill was established for that purpose to a thriving industry of about 12 commercial aqua feed producers in Nigeria and companies who import high quality floating feeds from Netherlands, United State American and Europe. As such there is currently in the market an assortment of both imported and locally manufactured pelleted floating catfish feed brands. Fish feed alone accounts for at least 60% of the total cost of fish production and the nutrient composition of feed influences feed utilization and ultimately the growth of fish. Given that feed is the highest recurring cost in catfish culture, the catfish farmer who is now besieged with different catfish brand options will benefit from information gathered through feed trials which indicate the more efficient brands in terms of optimum growth in minimum time. Feed trials have been carried out on *Clarias gariepinus* to evaluate their growth response to different readily available local plant and animal protein sources (1; 2).

The aquaculture industry has been globally recognized as the fastest growing food producing industry (3). However, the major challenge with the growth recorded in aquaculture is the overdependence of aquafeeds on fish meal. Fish meal is one of the most expensive ingredients of aquaculture diets. Diet supplementation is an important aspect in aquaculture management especially in intensive or in semi-intensive fish culture and is promising for increasing fish production. EL-Haroun (4) pointed out that diet is often the single largest operating cost item that represent over 50% of the operating costs in intensive aquaculture. Traditional use of antibiotics and other chemotherapeutics in fish culture has been criticized because of the potential development of multiple antibiotic resistant bacteria, environmental pollution and the accumulation of residues in fish (5). Scientists have intensified efforts to identify and develop safe dietary supplements and additives that enhance the life activity, health and immune system of farmed fish (6).

World Health Organization encourages use of medicinal herbs and plants to substitute or minimize the use of chemicals through the global trend to go back to nature. With the shifting of attention from synthetic drugs to natural plant products, the use of plant extracts for enhancing growth performance in animals is now on the increase. Plants that were once considered of no value are now being investigated, evaluated and developed into drugs with little or no side effects, one of such plant is *Morinda lucida* (Oruwo leaf). Aquaculturists have therefore, begun to evaluate alternative diet ingredients to replace fish meal with readily available inexpensive plant sources. Two important members of this family *Clarias gariepinus* and *Heterobrachus bidorsalis* are prominent in African aquaculture due to their fast growth rate, resistance to diseases tolerance to high density culture, ability to grow on a wide range of natural and low-cost artificial feeds and ability to withstand low oxygen and pH levels. There is scarcity of information regarding the utilization of *Morinda leaves* in the diet of African catfish (*C. gariepinus*). *Morinda lucida* leaves are recognized as remedies against different types of fever, including yellow fever, malaria, trypanosomiasis, and feverish condition during childbirth. The plants in some cases, is employed in treating diabetes, hypertension, cerebral congestion, dysentery, stomach ache, ulcers, leprosy and gonorrhea (7). Therefore, the aim of this study was to determine the effects of dietary *Morinda lucida* supplementation on the growth and nutritional performances of African catfish (*Class gariepinus*) Juvenile and to know whether *Morinda lucida* can be used as source of plant protein in order to reduce cost of fish production.

Materials and Methods

Experimental site

An 8weeks completely randomized design (CRD) feeding trial was conducted at the site of Agricultural Technology Department, Federal Polytechnic, Ado-Ekiti, Ekiti State, Nigeria. The state is located in south western part of the country, Ekiti State covers a land area of 6,353km square (2453sqmi) with a population estimated in 2005 to be 2,737,186. It enjoys tropical climate with two distinct seasons, these are rainy season (April to October) and dry season (November to March). Ado-Ekiti has a temperature ranges between 21 degree Celsius and 28 degrees Celsius with high humidity, the South westerly wind and the North east trade which blows in the raining season and dry (harmattan) season respectively, the tropical forest exist in the south of Ekiti State while savannah occupies the northern peripheries

Experimental design

Healthy and similar sized of One hundred and fifty juveniles of *Clarias gariepinus* were purchased from a reputable farm in Ado, Ekiti State of Nigeria. The fish were acclimatized in 50litres plastic bowls for two weeks. During the period of acclimatization, the fish were fed *ad libitum* as described by Anibeze and Eze. (8) at 5% body weight twice daily.

At the end of the acclimatization period, the fish were randomly selected and assigned to five dietary groups of 0% (control), 25%, 50%, 75% and 100% concentrations of *Morinda lucida* inclusion i.e.

Group 1: T1- Normal control (NC) – 0% *Morinda lucida* inclusion

Group 2: T2- 25% *Morinda lucida* inclusion

Group 3: T3- 50% *Morinda lucida* inclusion

Group 4: T4- 75% *Morinda lucida* inclusion

Group 5: T5- 100% *Morinda lucida* inclusion

Fifteen plastic aquaria, each measuring 40cm x 27 cm x 27cm with each aquarium holding 10 fish were set up to maintain three replicates per treatment. Feeding was suspended 24hours before the feeding trial to increase appetite and reception of the new diet.

Experimental diet

Feed ingredients were sourced from Feed Ingredients supply stores in Ado Ekiti and were used to formulate basal diets. All ingredients were ground into powdery form using a mechanical grinder, and then mixed with mineral mixture and vegetable oil. The ingredients and proximate chemical composition of basal feed was estimated by the methods described by the AOAC (9). Five different diets with or without substitution of *Morinda lucida inclusion* represented five dietary variants (Table 1) were then prepared by mixing *Morinda lucida* powder into basal diet at levels of 25%, 50%, 75% and 100%. The ingredients were then thoroughly mixed together by hand. Warm water was added to the premixed ingredients and homogenized to a dough like paste. The diets were pelletized using 0.2 mm pellet press and sun-dried for 4 days and labelled appropriately and stored in airtight containers throughout the experimental period.

Experimental procedure

Feeds were offered twice in a day (8:00am and 4:00pm) at 5% body weight throughout the experimental period of 56 days. Feeding allowance was adjusted in accordance with increase in body weight and diet allotments were increased weekly after the weight determination. Dead fish were picked daily and recorded.

Monitoring of water quality

Partial water exchange (half of the tank) was carried out as described for pre-experimental period before fish were fed, while water quality parameters (temperature, dissolved oxygen, total dissolved solids) were monitored before water exchange. The final weight of fish in each tank was determined at the end of the experiment. Other parameters such as number of fish survived, mean weight gain, specific growth rate, food conversation ratio, protein efficiency ratio and cost of using each feed in growing 1kg of fish were calculated as described by (10). The nutrient composition of the feed samples was determined using the standard procedures of Association of Official Analytical Chemists (9).

Data collection

Fish in each tank were batch weighed at the commencement of the study and weekly thereafter with digital electronic weighing balance to the nearest gram. The ration was adjusted every week when new mean weights of fish for the various experimental units were determined. Data on performance such as body weight changes, relative weight gain, specific growth rate, feed conversion ratio, condition factors and survival percentage were determined using the following formulae:

(a) Weight gain (g) = Final weight of fish - Initial weight of fish

(b) Relative weight gain (RWG, %) = $\frac{\text{Weight gain x 100}}{\text{Initial Weight}}$

(c) Specific growth rate (SGR) was calculated as:

$$SGR = \frac{\text{In (WF – W1) x 100}}{N}$$

(d) % survival Rate = $\frac{\text{Number remaining}}{\text{Number stock}}$ x100

(e) Feed conversion ratio = $\frac{\text{Total dry Feed fed (g)}}{\text{Total weight by fish (g)}}$

(f) Protein efficiency ratio = $\frac{\text{Wet weight gain by fish (g)}}{\text{Amount of protein in feed (g)}}$

Statistical analysis

The data obtained from the growth performance of *Clarias gariepinus* (catfish) fed graded inclusion levels of *Morinda lucida* (T1- T5) was subjected to standard methods of statistical analysis using windows-based SPSS, Version 20.0 (11). The analysis of variance (ANOVA) test was used and Level of significance was set at p<0.05.

Results

Proximate composition of feed samples

The results of the proximate analysis of the experimental feeds were presented in Table 2. T1 with 0% *Morinda lucida inclusion* (26.61%) and T2 with 25% *Morinda lucida inclusion* (25.11%) were found to be significantly (P< 0.5) higher in crude protein. T5 with 100% *Morinda lucida inclusion* had the highest crude fibre content (6.16%) while ether extract was found to be highest in T2 with 25% *Morinda lucida* (5.77%). T5 with 100% *Morinda lucida* had the highest carbohydrate content (61.00%), T2 with 25% *Morinda lucida* had the highest ash and moisture content (4.87% and 12.16%) respectively. The water quality parameters monitored in plastic aquaria tanks under laboratory conditions, as indicated in Table 3 were fairly stable in all the treatments. Water quality parameters were not significantly different (P>0.05) between treatments and were within the recommended ranges for the culture of *Clarias gariepinus* (12).

Table 4 showed the growth performance of the *Clarias gariepinus* fed graded levels of *Morinda lucida* inclusion, the Initial weight of T1 (control) was (35.30±0.06g); T2 (35.80±0.06g); T3 (35.63±0.07g); T4 (37.17±0.60g) and T5 (37.47±0.22). Fish fed with T1 (162.00±2.51g) and T2 (120.67±4.37g) recorded high values in final weight, weight gain T1 (126.70±2.45 g) and T2 (84.87±4.31g) and specific growth rate T1 (78.21%/day) and T2 (70.33%/day) as well as low values in feed conversion ratio T1 (6.03±1.29) and T2 (7.19±0.72) than those fed with T3 to T5. Percentage survival rate for T2 (100%) was the highest. Estimate cost of feeding was highest in T1 and T2.

Table 1: Gross composition of experimental diets for Catfish (*Clarias gariepinus*)

Ingredients	T1(g) (0%)	T2(g) (25%)	T3(g) (50%)	T4(g) (75%)	T5(g) (100%)
Maize	31	31	31	31	31
Morinda leaf	____	5.25	10.5	15.75	21
Ground nut cake	23.4	23.4	23.4	23.4	23.4
Soy bean meal	21	15.75	10.5	5.25	____
Fish meal	20	20	20	20	20
Oil	2.0	2.0	2.0	2.0	2.0
Lysine	0.5	0.5	0.5	0.5	0.5
Vitamin premix	0.1	0.1	0.1	0.1	0.1
Salt	0.25	0.25	0.25	0.25	0.25
Bone meal	1.5	1.5	1.5	1.5	1.5
Methionine	0.25	0.25	0.25	0.25	0.25
Total	**100**	**100**	**100**	**100**	**100**

Table 2: Proximate composition of the experimental diets

Ingredients	T1 0%	T2 25%	T3 50%	T4 75%	T5 100%
Protein	26.61	25.11	22.01	21.37	15.14
Carbohydrate	51.10	53.11	59.10	57.19	61.00
Moisture content	10.79	12.16	11.66	10.70	10.73
Ether Extract	3.90	5.77	4.91	3.64	3.64
Crude Fiber	3.80	4.07	4.13	3.56	6.16
Ash	3.21	4.87	3.47	3.41	2.31

Table 3: Water quality parameters during the experimental trial

Treatment	T1 0%	T2 25%	T3 50%	T4 75%	T5 100%
Dissolved Oxygen (mg/L)	4.42 ± 0.61^a	4.29 ± 0.33^a	4.26 ± 0.57^a	4.22 ± 0.58^a	4.12 ± 0.61^a
Temp(0C)	28.03 ± 0.28^a	28.00 ± 0.30^a	27.97 ± 0.33^a	27.97 ± 0.33^a	28.00 ± 0.30^a
Ph	7.10 ± 0.0^a	7.10 ± 0.0^a	7.10 ± 0.0^a	7.10 ± 0.0^a	7.07 ± 0.03^a

Mean value with the same superscript letter in the same row are not significantly different (p>0.05).

Table 4: Growth performance of *Clarias gariepinus* juvenile fed graded levels of *Morinda lucida* inclusion

Parameters	T1 0%	T2 25%	T3 50%	T4 75%	T5 100%
Initial Weight(g)	35.30±0.06^{a}	35.80±0.06^{a}	35.63±0.07^{a}	37.17±0.60^{b}	37.47±0.22^{b}
Final Weight(g)	162.00±2.51^{a}	120.67±4.37^{b}	84.67±9.17^{d}	98.00±6.43^{c}	85.67±2.33^{d}
Weight gain(g)	126.70±2.45^{a}	84.87±4.31^{b}	49.04±9.10^{d}	60.83±5.83^{c}	48.20±2.11^{d}
Specific Growth Rate (%/day)	78.21^{a}	70.33^{a}	57.92^{c}	62.07^{b}	56.26^{c}
Protein efficiency ratio	6.15^{a}	3.38^{bc}	2.23^{c}	4.02^{b}	2.26^{d}
Survival Rate (%)	96^{a}	100^{a}	86^{b}	84^{b}	80^{c}
FCR	6.03±1.29^{a}	7.19±0.72^{ab}	10.14±0.72^{b}	9.03±0.17^{c}	10.96±1.12^{b}
Cost of feeding (N/kg)	2500	2350	2215	2108	2009

^{abc} Values in the same column for each parameter with superscript different are significantly different at ($p< 0.05$).

Discussion

Growth data parameters, survival and mortality are great tools for evaluating the effect of feed and its value composition on fish species. This study showed that medicinal plant used in this research (like most other ingredient sold and used for feeding *C. gariepinus* and other fish species in Nigeria) affect growth response in *C. gariepinus* as compared to the use of soy bean meal feed ingredient. The poor growth response as recorded from the weight gained, low SGR and PER and high FCR is attributable to the proximate composition of the feed. According to De Silva and Anderson (13), the quality of a feed is a function of how well that feed meets the nutrient requirement of a fish. The good growth performance of fish fed with soy bean meal is an indication that the feed contained well balanced nutrients as seen in the proximate composition of the feed as well as its high digestibility and nutrient utilization. The very low percentage composition of crude protein, lipid and ash and very high percentage composition of carbohydrate and crude fiber in T5(100%) *Morinda lucida* feed were responsible for the poor growth performance of *C. gariepinus* when fed with the feed with protein being most significant and limiting the growth. It had been shown by various workers that fish growth is significantly influenced by the level of protein in the feed (14); (15) with 40% dietary protein promoting maximum growth of *C. gariepinus* (16) The crude protein in T4 and T5 feed was far less than the acceptable range recommended for commercial fish (17).

The low protein content in the T4(75%) and T5(100%) *Morinda lucida* feed was responsible for the high FCR recorded in the fish. This showed that low value of medicinal plant would be required to produce the fish to table size, thereby making the production of the fish more expensive as compared to expensive soy bean meal feed which requires less feed-to-feed fish to table size. Sawhney and Gandotra (18) found that food conversion efficiencies in fish increased with increasing protein in the diet. This study showed that *C. gariepinus* T5(100%) *Morinda lucida* did not utilize high amount of carbohydrates for growth, but had high protein content. This was in agreement with Mollah and Alam (19) who reported negative effect of carbohydrate on growth of *C. batrachus* fry when fed with more than 15% in the diet. Similarly, Tan *et al.* (20) reported that carbohydrate in the diet of *Clarias* species should not exceed 20%, if it thus, FCR and PER begins to decrease. The imbalance in carbohydrate value (CHO) in diet T4 and T5 was another reason why the fish fed with diet T5 showed very poor growth response to the feed. Ali and Jauncey (21) observed that CHO value ranging from 1.70 produced significant improved growth performance and feed utilization in *C. gariepinus*. Erfanullah and Jaffri (22) showed that imbalance with respect to non-protein energy sources and their inclusion levels may have direct effect on the growth, feed conversion, nutrient retention and body composition, with fish fed lowest or highest CHO value produced lower growth and feed conversion efficiencies.

High FCR on account of reduction in feed intake observed in the tanks gave rise to a lot of uneaten feed thereby deteriorating the water quality with water pH becoming low (acidic) and carbon dioxide increased and dissolved oxygen decreased. Similar scenario has been reported by Tan *et al.* (20). Another possible cause of the slow growth performance in fish fed with T5(100% *Morinda lucida* was the high percentage composition of fiber in the feed. This could be due to the inability of the fish digesting and utilizing the high fiber content in the feed. High level of fiber content in feed has been observed to slow the growth of *C. gariepinus* fingerlings (23; 24). In the results of Agokei *et al.* (25) significant highest growth performance of *C. gariepinus* juveniles was found in the diet that contained <2% fiber content. The low ash content found in the local feed could also be responsible for the poor growth performance of the fish in T3, T4 and T5 tanks. This occurred as a result of necessary mineral elements such as calcium and phosphorous that promotes growth in fish lacking in the diets. Ali and Jauncey (21) noted a better growth performance of *C. gariepinus* on diet containing 9.3% ash content, while Alam *et al.* (26) opined that ash content in the feed of *C. gariepinus* should not be less than 8%. High ash content of >12% in feed has been reported to produce better growth performance in *Clarias* species (27).

The proximate analysis of the control feed done in this research agreed with that of Agokei *et al.* (25), Ekanem *et al.* (28) and Ayuba and Iorkohol (29). The lack of balanced ration in the feed gave rise to large amount of uneaten food which subsequently decomposed leading to the deteriorating water quality in the tanks. All these caused stress to the fish leading to their poor growth and mortality. Huntingford *et al.* (30) stressed that lack of essential and balanced nutrients in feed could lead to poor growth and mortality, while Mustapha *et al.* (31) observed that stressful conditions in culture tanks of *C. gariepinus* juveniles always lead to their mortality. The inability of the fish species in T3, T4 and T5 tanks to completely eat the feed could be linked to the less fishy odor of the feed since *C. gariepinus* uses olfactory senses during feeding. The hardy nature and ability of the *C. gariepinus* to tolerate poor water quality made the mortality to be minimal considering the period of the experiment (56 days). The water quality parameters in the five tanks were within the tolerable limits for the culture of the species.

The lack of balanced nutrients and poor proximate composition of the local feed which reflected in the high FCR when fed to fish was also seen in the economics of the feeding and feeding. ₦ 2000 was used in feeding 30 fish for 56 days as opposed to ₦

2350 used in feeding 10 fish with 25% soy bean meal 84.87g body weight in 56days while ₦ 2000 used in feeding 10 fish with 100% soy bean meal 48.20g body weight in 56days. This is a case of pennywise pound foolish for the farmer who thinks the medicinal plants was cheaper and did not consider the FCR, PER, SGR and weight gain of the fish fed with medicinal plant as compared to those fed feed with soy bean meal. The need to understand the roles of medicinal plant in aquaculture has led to various investigation of different herbal plants in aquaculture feed. To date a variety of herbs and spices have been successfully used in fish culture as growth promoters and immune stimulants (32). When medicinal plants are used in fish diets, one of the common problems encountered is the acceptability of the feed by fish, and this frequently relates to the palatability of the diet.

In this present investigation, the experimental diet variants were accepted by *Clarias gariepinus* juvenile, indicating that the levels of incorporation of herbal plant used did not affect the palatability of the diets. This might be attributed to the processing technique which involved air drying techniques that might have reduced the anti-nutrient factors that may be present in this plant thereby not affecting the palatability of the diets. This observation corroborated the works of Francis *et al.* (33). These workers reported that reduction in anti-nutrient by different processing like soaking, and drying techniques resulted in better palatability and growth in fish. Medicinal plants have received increasing attention as spices for human and additives in diets for animals. However, only few studies had been done on the use of feed additives in fish nutrition (34) and more so medicinal plants as feed ingredient substitution in rearing of African catfish in this part of the world. The results in this experiment suggested that dietary feed ingredient of *Morinda lucida* had effect on growth of *Clarias gariepinus* juvenile as evidenced by similar body weight gain among treatment groups fed supplemented diet. However, T2 (25% *Morinda lucida)* showed improved weight gain, Feed Conversion Ratio and higher survival rate (SR) compared to other groups. Moreover, final fish weights were higher than the initial weights in all the groups, this indicated no negative growth as a result of diet supplementation. The low weight gains recorded in T5 (100% *Morinda lucida*) in this study might be attributed also to the fact that fish were fed exclusively on the formulated feeds without having access to natural feed as may be found in pond or riverine conditions. The African catfish is omnivorous and feeds from a wide array of organisms under natural conditions.

Conclusion and Applications

1. This study had shown that 25% *Morinda lucida* (Oruwo leaf) inclusion in fish feeds enhanced growth of *Clarias gariepinus*, however the substitution rate at 25% proved to be more beneficial than other substitution rates used in the experiment. Variations in growth parameters observed in the present study implied that catfish farmers should look for other plant protein feed ingredient for formulating feeds for their fish in order to reduce cost of production.
2. Further research was recommended to investigate toxicity of this plant at varying substitution levels of 25% to 100% in the *Clarias gariepinus* culturing to prove the safety of this plant. Likewise, variations in growth parameters and estimated cost of feeding observed in the present study

implied that catfish farmers should exercise great caution in formulating feeds for their fish since these feeds produce different growth effects on fish.

3. It is also recommended that feeds producers should formulate a ration that would give high profits which would determine the types of ingredients used and consequently the nutrient compositions of the feeds.
4. So, it is essential that every feed manufacturer formulate a nutritionally balanced-ration that would produce optimum fish growth.
5. Research-based evidence on nutritive values of feeds should also be provided by feed millers before they are certified for use. This becomes necessary in order to avert non-nutritious feeds with questionable nutrient compositions that can adversely affect local production of *Clarias gariepinus.*

References

1. Otubusin, S. O., Ogunleye, F.O., Agbebi, O.T. (2009). Feeding trials using local protein sources to replace fishmeal in pelleted feeds in catfish *Clarias gariepinus* (Burchell 1822) Culture. *European Journal of Scientific Research,* 31(1): 142-174.
2. Sotolu, A. O. (2009). Comparative utilizations of fish waste meal with imported fishmeal by African catfish (Clarias gariepinus). *American-Eurasian Journal of Scientific Research*, 4(4): 285-289.
3. FAO (2010) The State of World Fisheries and Aquaculture. Pp.197.
4. EL Haroun E.R, Ma A, Goda S, Kabir Chowdhury M.A. (2006) Effect of dietary probiotic Biogens supplementation as growth promoter on growth performance and feed utilization of Nile tilapia, *Oreochromis niloticus* (L.). *Aquaculture Resources* 37: 1473-1480.
5. Ringo, E., Olsen, R. E., Gifstad, T. O., Damo, R. A., Amlund, H. (2010). Prebiotics in aquaculture: A review. *Aquacultural Nutrition* 16(2): 117-136.
6. Shim, S. M., Ferruzzi, M. G., Kim, Y. C., Janle, E. M., Santerre, C. R. (2009). Impact of Phytochemical-rich foods on bio-accessibility of mercury from fish. *Food Chemistry*, 112: 46-50.
7. Abu, N. E., Ezeomeke, S. I., Azegba, P., Davidson, G. I. (2016). Phytochemical, nutritional and anti-nutritional properties of leaves, stem bark and root of trees used in popular medicine for the treatment of malaria in south eastern Nigeria. *Journal of Medicinal Plant Research*. 10(38):662-668.
8. Anibeze, C. I. P.and Eze, A. (2000) Growth response of two African Catfishes (*Osteichthys: Clariidae*) in Homestead concrete ponds. *Journal of Aquacultural Sciences* 15: 55-58.
9. AOAC. (2005). Official methods of analysis. 18th ed. Association of Official Analytical Chemist. Arlington, V.A. pp. 806-842.
10. Limbu, S. M. **(**2015). The effect of floating and sinking diets on growth performance, feed conversion efficiency, yield and cost-effectiveness of African sharptooth catfish, *Clarias gariepinus* reared in earthen ponds. *International Journal of Fisheries and Aquaticultural Studies*, 2(5): 253-259
11. SPSS 20.0 (2014). Statistical Computer Software, SPSS Inc., Chicago, IL, USA. ISBN: 0-13-017902-7.
12. Viveen, W. J., Richter, C. J. J., Van Oordt, P. G., Janssen, J. A. L, Huisman, E. A. (1986). Practical manual for the African Catfish, *Clarias gariepinus*. Section for Research and Technology, Box 20061, 2500EB. The Haque, Netherlands Pp. 121.

13. De Silva, S. S., and Anderson, T. A. (1995): Fish nutrition in Aquaculture, Chapman & Hall, London. Duncan db, 1955: Multiple range and multiple, F-tests: *Biometrics ...aiep*. 2000: 1-2.
14. Keremah, R. I. and Beregha, O. (2014). Effect of varying dietary protein levels on growth and nutrient utilization of African catfish *Clarias gariepinus* fingerlings. *Journal of Experimental Biology and Agricultural Sciences,* 2(1): 13-18.
15. Corn'elio FHG, Cunha DA, Silveira J, Alexandre D, Silva C and Fracalossi DM (2014) Dietary protein requirement of juvenile Cachara Catfish, *Pseudoplatystoma reticulatum*. *Journal of the World Aquaculture Society* 45(1): 45-54.
16. Van Weerd, J. H. V. (1995) Nutrition and growth in *Clarias* species - a review. *Aquatic Living Resources* 8: 395- 401.
17. NRC (National Research Council) (2011). Nutrient Requirements of Fish and Shrimp. National Academic Press, Washington, DC, USA.
18. Sawhney, S. and Gandotra, R. (2010) Effects of photoperiod on growth feed conversion, efficiency and survival of fry and fingerlings of Mahseer, *Tor putitora. Israel Journal of Aquaculture*, *Bamidgeh* 62(4): 266-271.
19. Mollah, M. F. A. and Alam, M. S. (1990). Effects of different levels of dietary carbohydrate on growth and feed utilization of catfish (*Clarias batrachus* L.) fry. *Indian Journal of Fisheries,* 37(3): 243-249.
20. Tan Q, Xie S, Xhu X, Lei W and Yang Y (2007) Effect of carbohydrate to lipid ratios on growth and feed efficiency in Chinese long snout catfish (*Leiocasis longirostris*). *Journal of Applied Ichthyology* 23(5): 605-610.
21. Ali, M. Z. and Jauncey, K. (2004a) Effect of feeding regime and dietary protein on growth and body composition of Clarias gariepinus (Burchell, 1822).
22. Erfanullah and Jafri, A. K. (1998). Effect of dietary carbohydrate-to-lipid ratio on growth and body composition of walking catfish *Clarias batrachus*. *Aquaculture* 161(1-4): 159-168.
23. Adewolu, M. A., Ikenweiwe, N. B. and Mulero, S. M. (2010). Evaluation of an animal protein mixture as a replacement for fishmeal in practical diets for fingerlings of *Clarias gariepinus* (Burchell, 1822*). Israel Journal of Aquaculture-Bamidgeh*, 62(4): 237-244.
24. Agbabiaka LA, Okorie KC and Ezeafulukwe CF (2013) Plantain peels as dietary supplement in practical diets for African catfish (*Clarias gariepinus* Burchell 1822) fingerlings. *Agriculture and Biology Journal of North America* 4(2): 155-159.
25. Agokei, E. O., Oparah, C. A., Aranyo, A. and Apapa, U. (2011). Growth of *Clarias gariepinus* juveniles fed five commercial feeds. *Continental Journal of Fish and Aquatic Science*, 5(3): 1-5.
26. Alam, M. K., Habib, M. A. B. and Tahmid, M. S. (2012). A survey on commercial fish feed used at Fulpur area in Mymensingh District. *Journal of Bangladesh Agricultural University* 10(1): 175-178.
27. Kiriratnikom, S. and Kiriratnikom, A. (2012) Growth, feed utilization, survival and body composition of fingerlings of slender walking catfish, *Clarias nieuhofii*, fed diets containing different protein levels. *Songklanakarin Journal of Science and Technology,* 34(1): 37-43.
28. Ekanem, A. P., Eyo, V. S., Obiekezie, A. I., Enin, U. I. and Udo, P. J. (2012). comparative study of the growth performance and food utilization of the African catfish (*Clarias gariepinus*) fed Unical aqua feed and coppens

commercial feed. *Journal of Marine Biology and Oceanography* 1(2): 1-6.

29. Ayuba, V. O. and Iorkohol, E. K. (2012) Proximate composition of some commercial fish feeds sold in Nigeria. *Journal of Fisheries and Aquatic Science* 8(1): 248-252.
30. Huntingford, F, Adams, C., Braithwait, V. A., Kadri, S., Pottinger, T. G., Sandøe, P. and Turnbull, J. F. (2006). Current issues in fish welfare. *Journal of Fish Biology* 68(2): 332-372.
31. Mustapha, M. K., Akinware, B. F., Faseyi, C. A., Alade, A. A. (2014). Comparative effect of local and foreign commercial feeds on the growth and survival of *Clarias gariepinus* juveniles. *Journal of Fish*ery. 2(2):106-112.
32. Irkin, L. C., Yigit, M., Yilmaz, S., Maita, M. (2014). Toxicological Evaluation of Dietary Garlic (*Allium sativum*) Powder in European Sea Bass *Dicentrarchus labrax* Juveniles. *Food and Nutrition Sciences.* 5: 989-996.
33. Francis, G., Makkar, H. P. S., Becker, K. (2001) Antinutritional factors present in plant-derived alternate fish feed ingredients and their effects in fish. *Aquaculture.* 199: 197-227.

34 Lawhavinit, O., Sincharoenpokai, P., Sunthornandh, P. (2011) Effects of ethanol turmeric (*Curcuma longa* Linn.) extract against shrimp pathogenic *Vibrio spp*. and on growth performance and immune status of white shrimp (*Litopenaeus vannamei*). *Kasetsart Science, Journal of Natural sciences,* 45: 70-77.