Nigerian Journal of Fisheries and Aquaculture 3(1&2):55 – 61, 2015

Department of Fisheries, Faculty of Agriculture, University of Maiduguri, Nigeria
Available on line: http://www.unimaid.edu.ng
ISSN-2350-1537

Replacement Value of Neem (*Azadirachta indica*) Seed Cake for Soya Bean Meal in the Diet of African Catfish *Clarias gariepinus* Fingerlings

*[1]Hassan, M., [2]Auta, J. and [2]Abdulllahi, S. A.
[1,] Department of Biological Sciences, Ahmadu Bello University, Zaria
[2] Department of Fisheries, University of Maiduguri

Received, August 1, 2015 Accepted August 30, 2015

ABSTRACT
One of the major problems facing aquaculture development in Nigeria is the non-availability of quality and affordable fish feeds. This study investigated the nutritional suitability of *Azadirachta indica* seed cake as alternative protein source to soya bean meal in the diet of African catfish (*Clarias gariepinus).* The neem seed cake was processed using water washed method to reduce the bitterness and toxicity of the cake. Different concentrations of neem seed cake meals (0, 25, 50 and 75%, designated as DI, DII, DIII and DIV, respectively), were incorporated into 40 % crude protein diet. *C. gariepinus* (6.00±0.93- 6.06±0.87g weight and 8.03±0.83 - 8.46±0.21cm long) fingerlings were stocked in fifty litre plastic trough in replicates and feed for a period of eight (8) weeks. Growth performance and nutrient utilization indicated that the control and fish fed 25% name seed cake (NSC) recorded the best weight gain (23.24g and 20.8g), respectively. Growth significantly ($P<0.05$) decreased by 17.55% and 28.58% levels of inclusion. A similar trend was observed in specific growth rate and feed intake. However, feed conversion ratio, protein efficiency ratio and net protein utilization were not significantly different ($P>0.05$). The result suggests that the Neem seed cake can be incorporated at 9.62% in the diet of the African catfish (*Clarias gariepinus)* without compromising fish growth and nutrient utilization. Therefore the use of this unconventional ingredient can be encouraged in this era of competing demand for conventional ingredients.

Key Word: Replacement, *Neem Seed cake, Clarias gariepinus, Fingerlings Growth Performance*

INTRODUCTION

With the global population expansion, the demand for high quality animal protein is rising dramatically (Yisa*et al*., 2006). Increased aquaculture production is clearly needed to meet this demand, because capture fisheries are showing serious declines due to over fishing, aquatic habitat destruction and pollution (FAO, 2004).

African catfish *Clarias gariepinus* is one of the major aquaculture fish in Nigeria and it commands high commercial value (Ogwuba, 2003). This may not be unconnected with its accessory breathing organs which enable it to tolerate low dissolved oxygen and other adverse aquatic environmental conditions where other culturable fish species cannot survive. Unfortunately, the successful production of this important aquaculture candidate in Nigeria has been hampered by the high cost of feeds. Aniebo*et al*. (2009) reported that in Nigeria, the bulk of feed used for fish production is imported. Protein is the main constituents of feed for aquaculture animals. The cost of protein rich feed makes up a large percentage of the total expenses of aquaculture feeds. EI-Sayed (1999) reported that Soya bean meal (SBM) has been the most common source of plant protein use as a fish meal replacer in aquaculture feeds, because of its high protein content, relatively well balanced amino acid profiles, reasonable price and steady supply. However, wider utilization

*Corresponding Author email: warodizaks@yahoo.com

and availability of this conventional source for fish feed is limited by increasing demand for human consumption and by other animal feed industries (Siddhuraju and Becker, 2001).

Neem is a multipurpose tree. People consume its fruits, raw or cooked, and sometimes eat the young twigs and flowers as vegetables (Orwa*et al*., 2009). Neem leaves, bark and seed extracts have been used for centuries in India in ethnomedicine and ethnoveterinary medicine (Biswas *et al*., 2002; Subapriya*et al*., 2005). Neem *(Azadirachta indica)* seed cake (NSC), a by-product of Neem oil, is one of the non-conventional fish feed ingredients showing great potential for livestock feeding (Gowda *et al.*, 1998; Bawa*et al.* 2005). The Neem seed cake has been noted as a rich protein source. Rao (1987) reported that *A. indica* seed cakecontain 45–59% crude protein, 3-9.10% fats, 2-9.50%, 3.0 – 8.60% crude fibre and 7 – 9% moisture. While Djenontin*et al*. (2012) reported 31.0, 5.3, 6.4, 4.0 and 4.6 % crude protein, ash, fat, crude fibre and moisture respectively in neem seed cake. The neem seed cake also consists of all the essential amino and non-essential amino acid, except Valine and tryptophan. According to Rao (1987), the amino acid content of neem seed cake is comparable to that of groundnut cake. It has good keeping quality and does not spoil on storage or attack by fungi (WNC, 2012). However, its inclusion in animal feeds have been discouraged due to the inherent bitter and toxic principles (Paul *et al*., 1996).

Ruminants and rabbits have been reported to occasionally forage on neem leave and the use of the neem products for livestock feed is limited (Heuze*et al*., 2015). The Neem seed cake is a protein-rich ingredient and its use by farmers has been recorded, for instance in Southern India (Christopher, 1970). However, its unpalatability and toxicity have prevented its widespread use in livestock feeding. This product has been described as a promising potential feed ingredient since the 1970s and there have been a considerable number of attempts at rendering it suitable for livestock (Heuze*et al*., 2015). Oil has been extracted from neem seed. The resulting from oil extraction of whole seeds (neem seed cake), or decorticated seeds (neem seed kernel cake), is usually considered as a non-edible oil cake only to be used as organic nitrogenous fertilizer (Ramachandran *et al*., 2007).

Interestingly, attempts have been made over the years to detoxify and reduce the bitterness of the neem cake for animal use, through extraction, water washing, alkali soaking, urea ammunition and autoclaving (Gowda *et al*., 1998; Uko and Kamalu, 2006; Bawa*et al*., 2007), with a view to eliminating the growth depression reported in raw neem seed cake. Rational use of this nutritive ingredient for fish production can reduce the high price of feedstuffs and enhance profitability of fish farming. The search for alternative plant proteins to replace soybean meal has gained increasing interest in the research on aquaculture feedstuff. There is a paucity of information on the utilization of the neem seed cake as a dietary ingredient for *C. gariepinus* in Nigeria. The objective of this study therefore is to evaluate the effects of replacing soybean meal with neem seed cake as the dietary protein source in the diet of *C. gariepinus* fingerlings.

MATERIALS AND METHODS

Experimental site

The experiment was conducted in the Fisheries Research Laboratory at the Department of Biological Sciences, Ahmadu Bello University Zaria, Nigeria.

Experimental fish

A total number of 120 fingerlings of *Clarias gariepinus* **(**6.00±0.93- 6.06±0.87g weight and 8.03±0.83 - 8.46±0.21cm length) were procured from a commercial fish hatchery (Miracle Fish Farm) Zaria. The experimental fish were acclimated for two weeks in a 50 litre plastic trough, during which they were fed commercial feed (40%**CP**) at a daily rate of 5% of biomass with half of the daily ration fed in the morning (8:00am) and the other half in the evening (5:00pm). Thereafter, batch weighing and length measurement of fish was done to ascertain their initial mean weight in grams and initial mean length in centimetres, using

top-load weighing balance (Meter Tolardo567) and fish measuring board, respectively. This was done before the commencement of the experiment and subsequently after every two weeks.

Processing of the *A. indica* seed cake

The neem seed cake were was purchased from local neem seed oil extractor in Potiskum town, Yobe State Nigeria. The neem seed cake was processed according the method described by Smita *et al*. (2005). The cake was soaked and wash in tap water several times and late sun-dried. The sun-dried neem seed cake was in a black polythene bag at room temperature.

Feed formulation and compounding

All the dietary (fish meal, yellow maize, and soybean) ingredients were separately processed and milled to fine particle size. The soybean was toasted for 15 minutes at 50-60^{o}C using local toasting pan. The fish meal, toasted soybeans and yellow maize were separately milled into powder using hammer miller.

Four (4) nitrogenous diets were formulated according to the nutritional requirements of the experimental fish, i.e. 40% crude protein (Table1). The protein level was chosen on the basis of some previous studies Fagbenro *et al*. (1992), where 40% protein in formulating feed demonstrated better growth of Catfish. The dry ingredients were weighed out according to the formulation (Pearson Square). The milled ingredients were mixed until uniformly blended and kept in an airtight container until required.

Experimental design

The neem seed cake was incorporated into the 40% crude protein diet at 0 (control), 25, 50, and 75% designated as treatments DI, DII, DIII and DIV, respectively (Table 1). The feed ingredients were thoroughly mixed until a homogenous product was obtained. Water was added (20%-30%) slowly to the mixture with continuous stirring until the dough was formed.

Table: 1 Gross and proximate composition of experimental diets

Ingredients	***A. indica* inclusion levels (%)**			
	DI	**DII**	**DIII**	**DIV**
Maize	24.25	23.69	25.51	22.49
Soya bean	36.37	27.44	17.55	9.52
Fish meal	36.37	36.37	36.37	36.37
Neem cake	-	9.62	17.55	28.58
Premixes	1.0	1.0	1.0	1.0
Binder	0.5	0.5	0.5	0.5
Lysine	0.5	0.5	0.5	0.5
Methionin	0.5	0.5	0.5	0.5
Salt	0.5	0.5	0.5	0.5
Fish oil	0.5	0.5	0.5	0.5
Proximate Composition				
DM	91.36	92.04	91.90	91.11
CP	41.12	40.50	40.51	40.56
CF	5.15	4.23	3.12	3.89
EE	6.39	6.08	7.00	6.99
Ash	7.62	7.90	6.17	6.17
NFE	39.75	41.41	43.33	42.41
Kcal/kg	380.99	382.38	398.32	394.79

This was followed by pelleting using manual meat mincer through 0.2mm die. The pelleted feeds were sun dried and later packaged in two layers of plastic bags stored in a well ventilated room under ambient

temperature. Before the commencement of the experiment, proximate analysis of experimental diets were conducted according to (AOAC, 1990). The fingerlings were stocked in 50 litre capacity plastic trough at density of 10fish/trough and were randomly allocated to the experimental name seed cake based diets in replicates. The fingerlings were fed on the experimental diets for eight (8) weeks. At the end of the 8 weeks rearing, final weight (g), final length (cm) total feed intake (g) and mortality of the fish were recorded. The following indices were used to determine growth performance and nutrient utilization:

i. Weight gain (WG) = $W_2 - W_1$, where, W_2 is mean final body weight (g), W_1 is mean initial mean body weight (g).
ii. Specific growth rate (SGR) = Ln $W_2 - W_1$/t (days) x 100, where Ln = log., t = culture period
iii. Survival rates of fish (SR) = number survived/initial number stock x 100
iv. Condition factor (K) = W/L^3 x 100, Where: W = Weight of fish. L = Standard length of fish
v. Feed conversion ratio (FCR) = feed consumed/weight gain
vi. Protein Efficiency Ratio (PER) = weight gain (g)/Crude protein fed (%)
vii. Net Protein Utilization (NPU) = Retained protein in tissue/Dietary protein feed x 100

Determination of water quality parameters

Water quality parameters such as Temperature, Dissolved Oxygen and pH were monitored daily using mercury -in- glass Thermometer and automatic DO/pH Analyser (Model JPB 607) respectively.

Statistical analysis

The data obtained were subjected to one-way analysis of variance (ANOVA). The Duncan multiple range test was applied to rank treatment means ($p< 0.05$) with the aid of SPSS Statistical package version 20.0

RESULTS

Growth performance

The growth performance of *Clarias gariepinus* fed experimental diets as presented in Table 2 shows that mean final weight gain ranged from 16.63g to 23.24g ($P<0.05$). The SGR ranged from 0.021g/ fish/day in 0.24g/day ($p>0.05$).

Table2: Mean ± SEM) growth performance and nutrient utilisation of *Clarias gariepinus*neen seed cake

Parameters	**A. *indica* inculsion levels (%)**			
	DI	**DII**	**DIII**	**DIV**
Initial Length (cm)	8.03±0.83	8.17±0.62	8.22±0.94	8.46±0.21
Final Length (cm)	13.95±0.31	12.79±0.31	11.80±0.30	12.89±0.60
Initial Weight (g)	6.00±0.93^a	5.97±0.88^a	6.06±0.16^a	6.06±0.87^a
Mean Final Weight (g)	29.75±0.96^a	26.83±0.99^{ab}	23.90±2.00^b	22.70±0.72^b
Weight Gain (g)	23.24±0.96^a	20.80±0.98^{ab}	17.83±2.00^b	16.63±0.05^b
Specific Growth Rate (g)	0.024±0.001^a	0.023±0.001^{ab}	0.022±0.001^b	0.021±0.003^b
Condition Factor	1.24±0.22^a	1.04±0.13^a	1.45±0.18^a	1.07±0.01^a
Survival Rate (%)	90.00±5.77^a	90.00±5.77^a	83.33±0.57^a	90.00±5.77^a
Feed Intake (g)	30.72±0.54^a	29.77±1.10^a	26.85±0.93^{ab}	25.66±0.59^b
Feed Conversion Ratio	1.29±0.06^a	1.43±0.08^a	1.53±0.15^a	1.53±0.04^a
Net Protein Utilization	99.00±1.52^a	96.66±3.71^a	87.00±3.08^a	90.33±2.40^a
Protein Efficiency Ratio	1.86±0.09^a	1.72±0.10^a	1.64±0.14^a	1.59±0.02^a

Means with same letter in the same column are not significantly different ($p>0.05$).

The control diets recorded the highest specific growth rate while DIV (50%NSC) had the lowest specific growth rate. The condition factor ranged from 1.07 to 1.45 ($p>0.05$). The survival rate ranged from 83.33% to 92%. The mean total feed intake ranged from 29.77g to 30.72g. ($p<0.05$). The PER ranged from 1.6 to 1.88 ($p>0.05$). The net protein utilization ranged from 87 to 100 ($p>0.05$). The FCR ranged from 1.29 to 1.53 ($p>0.05$).

Physico-chemical parameters

Table 3 shows the physic-chemical parameters recorded during the study. Temperature ranged from 26.0 to 26.45ºC ($p>0.05$), pH ranged from 7.27 – 7.96 ($p>0.05$) while dissolved ranged from 5.0 – 5.56 mg/l).

Table 4: Water quality parameters during the experiment

A. *indica* inculsion levels (%)	**Temp (ºC)**	**pH**	**Dissolved Oxygen (mg/l)**
DI	26.33±0.33	7.23 ± 0.14	5.46 ± 0.27
DII	26.00 ± 0.57	7.96 ± 0.06	5.37 ± 0.15
DIII	26.33 ± 0.33	7.96 ± 0.03	5.00 ± 0.33
DIV	26.45 ± 0.23	7.38± 0.03	5.10 ± 0.23

DISCUSSION

The growth performance of *Clarias gariepinus* fed water washed neem seed cake indicates that mean initial weight (5.97g -- 6.0g) was not significantly different ($p>0.05$) among the experimental treatments, showing uniformity in size at the onset of the experiment. Mean final weight gain generally decreased with increasing levels of neem seed cake in the diets. The control diet recorded the best weight gain (23.24g) which was not significantly different ($p>0.05$) from 20.8g recorded in fish fed diet containing 25 % Neem seed cake. Similarly, the specific growth rate (SGR) also displayed a decreasing trend with increasing levels of Neem seed cake in the diets. The best SGR value (0.024g/day) was obtained in fish fed the control diet (0%nsc) followed by fish fed 25% NSC diet (0.023g/day) while the lowest value (0.021g/day) was recorded in fish fed on a 75 % NSC diet. The decreasing trend in growths may be due to residual anti nutritional factors inherent in the cake.

The better growth performances recorded in DIV (75 % NSC) may be due to lower levels of inclusion which enhanced its nutritional composition, palatability and bioavailability. In a related experiment Jegede and Fagbenro (2008) did not observe any significant variation in growth parameters and feed conversion ratio of mixed sex *tilapia zilli* fed neem leaf meal supplemented diets. It is also in agreement with the report of Smita *et al*. (2005). The welfare (CF) of the fish in this experiment ranged from 1.07–1.45 ($P>0.05$) revealed that dietary inclusion of neem seed cake has no effect on the condition of the fish.

The decreasing trend recorded in feed intake may be as a result of the decreased palatability of the diets as the results of higher level of inclusion. The control diets and the diets containing 25% neem seed cake were generally more acceptable since fish were observed to be actively feeding and the activity ceased in less than 5 minutes and no left over feed was observed. The other diet was less readily accepted by fish, taking longer periods (about 5-15 minutes). Similar observations were made by Wasanthakumar *et al*. (1999) and Gowda and Sastry (2000). Another observation was made by Wasanthakumar *et al*. (1999) in an experiment with terrestrial animal where he reported lowered feed intake, nutrient digestibility and growth in rabbits fed 20% raw neem seed meal.

The nutrient utilization parameters revealed ($p>0.05$) for the feed conversion ratio (FCR), Protein efficiency ratio (PER) and Net Protein Utilization among the experimental diets compared to the control. The feed conversion ratio recorded in this experiment (1.29 – 1.53) in which the control diet had the lowest (best) and DIII and DIV had the highest (poor) was similar to the value on cottonseed meal (1.55) reported by Jabeen*et al*. (2004). Similarly, Smita *et al*. (2005) reported a significant increase ($p > 0.05$) in feed

conversion ratio, decreased specific growth rate and protein efficiency ratio in fish fed water washed neem seed cake supplemented diets.

This experiment suggests that even though growth depression occurred with fish fed water processed neem seed cake diet, the processed Neem seed cake was still adequate and can meet the basic physiological needs of the experimental fish. Another reason for this appreciable nutrient utilization may be due to the good composition of essential and non-essential amino acids in Neem cake reported by Gowda and Sastry (2000) as complete in its amino acid compositions.

Conclusion

The result of this experiment revealed that water washing can be a reliable method of de-bitterizing and detoxification of neem seed, suggesting that WNSC can be used to partially replace soya beans in *C. gariepinus* diet

Acknowledgement

The authors wish to acknowledge Mr. M. Steven and Mr. S. Luka of Department of Biological Sciences of the Ahmadu Bello University, Zaria for their assistance during the experiment. The contribution of Mr. Kwano Ibrahim of Department of Animal Science, ABU for proximate analysis of the experimental feed is also acknowledged.

REFERENCES:

Aniebo, A. O., Erondu, E. S. and Owen, O. J. (2009). Replacement of fish meal with maggot meal in African catfish (*Clarias gariepinus*) diets. *Revistacientifica,* 9(3): 666-671.

AOAC (1990). *Official Method of* Analysis. Helrich, K. (ed.) 15th Ed. Association of Analytical Chemist, Arlington, 684 Pp.

Ayoola, O. A. and Fredrick, O. A. (2012). Effects of the shape of culture tanks on production of the African catfish *Clarias gariepinus* juveniles. *Journal of Agriculture and Social Research*, 12(1):130-142.

Bairagi, A. S., Ghosh, K.S., and Ray, A. K., (2004). Evaluation of the nutritive value of *Leucaena leucocephala* leaf meal, inoculated with fish intestinal bacteria *Bacillus subtilis and Bacillus circulans*in formulated diets for Rohu, *Labeo rohit*a *(Hamilton)* fingerlings. *Aquaculture Research*, 35:436-446.

Bawa, G. S., Orunmuyi, M. and Onabanjo, O.A. (2005). Effects of dietary inclusion levels of mechanically extracted neem seed cake on performance of young rabbits. *Nigerian Journal of Animal Production*, 32: 233-239.

Bawa, G.S., Orunmuyi, M., Agbaji, A. S., Landam, Z., and Okekeifi, U. O. (2007). Effect of different methods of processing neem (*Azadirachta indica*) seeds on performance of young Rabbits. *Pakistan Journal of Nutrition*, 6 (3):213-216.

Biswas, K., Chattopadhyay, I., Banerjee, R. K. and Bandyopadhyay, U. (2002). Biological activities and Medicinal properties of neem (*Azadirachta indica*). *Current Sci.*, 82 (11): 1336-1345.

Christopher, J. (1970). Neem-seed cake is also good for cattle. *Indian Farming*. Available on: http://www.cabdirect.org/abstracts/19711403922.html, 20: 38.

Djenontin, T. S., Wotto, V. D., Avlessi, F., Lozano, P., Sohounhloué, D. K.C. and Pioch, D. (2012). Composition of Azadirachta indica and *Carapaprocera* (Meliaceae) seed oils and cakes obtained after oil extraction. *Industrial Crops and Products*, 38:39– 45.

El-Sayed, A.F.M. (1999). Alternative dietary protein sources for farmed tilapia, *Oreochromis* spp. *Aquaculture*, 179:149-168.

Fagbenro, O.A. Balogun, A.M. and Anyanwu, C. N. (1992). Optimal dietary protein level for *Heterobranchus bidorsalis* fingerlings fed compound diets. *Isr. J. Aquaculture Bamidgeh,* 44: 84-92.

Gowda, S.K., Sastry, V.R.B. and Katiyar, R.C. (1998*).* Study on the utilization of neem kernel meal as a protein supplement for growing rabbits. *Indian Veterinary Journal*, 75: 281-282.

Heuze, V., Tran, G., Bastianelli, D., Lebas, F., Archimede, H. (2015). Neem *(Azadirachta indica)*.

Feedipedia,a programme by INRA, CIRAD, AFZ and FAO. http://feedipedia.org/node/182 *Last updated on July 29, 2014*, 16:36.

Jabeen, S., Salim, M. and Akhtar, P. (2004). Study on feed conversion ratio of major carp *Cirrhinusmrigala*fingerlings fed on cotton seed meal, fish meal and barley. *Pakistan Veterinary Journal*, 24(1): 42-46.

Jegede, T. and Fagbenro, O. (2008). Dietary neem (*Azadirachta indica*) leaf meal as reproduction inhibitor in redbelly *Tilapia*, *tilapia zillii8th International Symposium on Tilapia in Aquaculture*, 365-373.

Nath, K., S. Rajagopal and A.K. Garge, (1983). Water washed neem (*Azadirachta indica*) seed kernel in cattle feed. *Journal of Agricultural Science*, 101: 323-326.

Odunsi, A. A., Adegbile, S. A., Akande, T.O. and Olayeni, T. B. (2009). Neem (*Azadirachta indica*) seed cake in the diets of cockerel chickens, *International Journal of Poultry Science*, 8(1):47 – 51.

Ogwuba, A.O. (2003); Key note Address of the programme manager (OGADEP) Abeokuta at the *first annual fish farmers seminar and training workshop organized by fisheries society of Nigeria Ogun state chapter at Abeokuta on July 31, 2003*, 4Pp.

Orwa, C., Mutua, A., Kindt, R., Jamnadass, R., and Anthony, S. (2009). *Agroforestree Database*: a tree reference and selection guide version 4.0. World Agroforestry Centre, Kenya.

Paul, B.N., Haque, N. and Garg, A. K. (1996). Effect of feeding neem seed kernel cake (*Azadirachta indica*) on rumen enzyme of Buffalo. *Indian Veterinary Journal*, 73:720-728.

Ramachandran, S. Singh., S. K., Larroche, C., Soccol, C. R. and Pandey, A. (2007). Oil cakes and their biotechnological applications – A review. Biores. *Technol.*, 98 (10): 2000-2009.

Rao, P. U. (1987). Chemical composition and biological evaluation of de-bitterized and defatted neem (*Azadirachta indica*) seed kernel cake. *Journal of the American Oil Chemists Society (JAOCS)*, 64 (9): 1348-1351.

Siddhuraju, P. and Becker, K. (2001). Preliminary nutritional evaluation of mucuna seed meal (*Mucunaprurien*svar. *utilis*) in common carp (*Cyprinuscarpio*L.): An assessment by growth performance and feed utilization. *Aquaculture*, 196:105-123.

Smita L., Shiba, S. G. and Kumar, P. H. (2005) Performance of *Cyprinuscarpio* fingerlings fed on diets containing water washed neem ((*Azadirachta indica*) Seed Cake. *Conference on International Agricultural Research for Development Stuttgarnheim*, October 11-13.

Subapriya, R., and Nagini, S. (2005). Medicinal properties of neem leaves: a review. *Curr. Med. Chem. Anticancer Agents*, 5 (2):149Pp.

Uko, O. J. and Kamalu, T. N. (2001). The neem tree: Uses and potentials. *Nigerian Journal of Experimental and Applied Biology*, 2(2): 223-229.

Wasanthakumar, P., Sharma K., Sastry V.R.B. and Kumar, S. (1999). Effect of graded dietary levels of neem (*Azadirachta indica*) seed kernel cake on carcass characteristics of broiler rabbits. *World Rabbit Science*, 7(3):145-149.

W.N.C. (2012). Neem as animal feeds. *World neem conference Nagpur*. Available on:www.neemfoundation.org/neem-aticles.html. 23rd may, 2013, 4Pp.

Yisa, M. Lamai, S. Olufeogba, S. O. and Tsadu, S. M. (2006). Intra-specific hybridization studies on three strains of *Clarias gariepinus. Proceedings of Fisheries Society of Nigeria*, 229-231.