IMPROVING BREEDING PERFORMANCE OF FINGERLINGS OF CLARIAS GARIEPINUS THROUGH INTRASPECIFIC HYBRIDIZATION

M. IWALEWA*, AKINWALE, M-M. ADEWOLE, E. OKHIRIA, B. EBONWU, E.B. IHEANACHO, O.C. BEDE

Nigerian Institute for Oceanography and Marine Research, Victoria Island, Lagos

*Correspondence Author : *iwalewamegbowon@yahoo.com*

ABSTRACT

The scarcity of genetically improved fish seed is a major constraint to rapid development of aquaculture in Nigeria due to inbreeding. The study was conducted to monitor breeding indices from intra-specific mating combinations of wild stocks of *Clarias gariepinus*. Broodstock of *C. gariepinus* were collected from Lokoja (LK), Makurdi (MK), Agenebode (AG) and Onitsha (ON) and compared with domesticated strain. 16 mating combinations of parental and intraspecific mating combinations were conducted in triplicates. Results were subjected to one way analysis of variance (ANOVA) (α = 0.05). The results showed collection from Onitsha had highest values of fertilization (95.2%), hatchability (87.6 %) and survival (66.0 %), followed by crosses from female from Onitsha : ♀ON X ♂LK, ♀ON X ♂ MK and ♀ ON X ♂ AG having 95.8 %, 91.1 % and 88.8 % (fertilization), 68.7 %, 61.9 % and 60.7 % (hatchability) and 61 %, 57 % and 63 % (survival) respectively. Domesticated strain had least values (81.9 %, 59.4 % and 51 %) fertilization, hatchability and survival respectively. ANOVA (α < 0.05) showed variation in fertilization, hatchability and survival. The better performing strains could be explored for genetic development. However, protein and gene profiling of these natural populations may be necessary to identify the proteins and genes responsible for these variations in breeding performances.

***Key Words*:** Breeding, performance, *Clarias gariepinus*, hybridization

RESUME

AMELIORATION DES PERFORMANCES D'ALEVINS DE CLARIAS GARIEPINUS PAR HYBRIDATION INTRASPECIFIQUE

La rareté des semences génétiquement améliorées de poisson est un obstacle majeur au développement rapide de l'aquaculture au Nigeria en raison de la consanguinité. L'étude a été menée afin de suivre les indices de reproduction de couples intraspécifiques de stocks sauvages de C. gariepinus*. Les géniteurs de* C. gariepinus *ont été collectés à Lokoja (LK), Makurdi (MK), Agenebode (AG) et Onitsha (ON) et comparée avec la souche domestiquée. Au total 16 combinaisons de croisements parentaux et intraspécifiques ont été effectuées en triplicat. Les résultats ont été soumis à une analyse de variance d'ANOVA au seuil de α = 0,05. Les résultats ont montré que le croisement issu d'Onitsha a les valeurs les plus élevées en termes de taux de fertilisation (95,2 %), d'éclosion (87,6 %) et de survie (66,0 %), suivis par les croisements avec les femelles d'Onitsha : ♀ON X ♂LK, ♀ON X ♂MK et ♀ ON X ♂ AG avec 95,8 % ; 91,1 % et 88,8 % de taux de fécondation, 68,7 % ; 61,9 % et 60,7 % de taux d'éclosion et 61 % ; 57 % et 63 % de taux de survie respectivement. La souche domestiquée a les plus faibles valeurs de taux de fécondation, d'éclosion et de survie respectivement 81,9 % ; 59,4 % et 51 %). L'analyse d'ANOVA (α < 0.05) a montré une variation du taux de fécondation, d'éclosion et de survie. Les souches plus performantes pourraient être explorées pour le développement génétique. Cependant, les protéines et le profil génétique de ces populations naturelles doivent être nécessairement identifiés de même que les gènes responsables de ces variations dans les performances de reproduction.*

Mots clés*: Elevage, performance,* Clarias gariepinus*, hybridation*

INTRODUCTION

The clariid catfish, *Clarias gariepinus* is a remarkable fish species in Nigeria where it is the leading aquatic crop. It has the credentials of fast growth, resistance to disease and handling stress. It has air-breathing structure and can therefore tolerate very low oxygen levels in any aquatic environment. Increased productivity of fry and fingerlings with attributes of fast growth and better environmental tolerance is *sine qua non* to ensuring food security in Africa. The scarcity of genetically improved fish seed is one of the major constraints to the rapid development of aquaculture industry and stock management in Nigeria. A good supply of high quality fingerlings is essential for successful aquaculture production. Many fish hatcheries in Nigeria use catfish of the same parentage resulting in inbreeding depression over several generations, leading to reduction in overall production (Olaleye, 2005).

The eggs of a « ripe » female make up 15 - 20 % of the body weight. In captivity the African Catfish does not spawn spontaneously since the environmental factors such as the rise in water level and inundation of shallow areas do not occur on the fish farms. Under natural condition of spawning, lower hatching rates have been reported for *Clarias gariepinus* by various authors. de Graaf *et al.* (1995) reported an average rate of 59.1 % in the rainy season for *C. gariepinus* in the Republic of Congo, while Macharia *et al.* (2005) reported a rate as low as 4 % for *C. gariepinus* eggs incubated on a nylon substrate. Fertilization, hatching and early survival of larvae are vital for successful aquaculture of the African catfishes (Ataguba *et al.*, 2009).

Cultured fish are being improved for a multitude of traits including growth rate, feed conversion efficiency, disease resistance, tolerance of low water quality, cold tolerance, body shape, dress out percentage, carcass quality, fish quality, fertility and reproduction and harvestibility (Dunham, 2001). Fish hybridization is one of the major breeding strategies that have been used to improve production characteristics in aquatic organisms. Improving growth rate will decrease the time it takes to grow a fish to marketable size which is advantageous for fish farmers. This will increase production efficiency, fish production and farmer's income. The objective of the study is to improve the breeding performance of catfish fingerlings through intraspecific hybridization in an indoor hatchery.

MATERIAL AND METHODS

INTRASPECIFIC HYBRIDIZATION OF C. GARIEPINUS FROM DIFFERENT WATER BODIES

The research study was carried out at Nigerian Institute for Oceanography and Marine Research, Badore station, Lagos. Broodstock of *C. gariepinus* were collected in natural populations from Lokoja (LK), Makurdi (MK), Agenebode (AG), and Onitsha (ON). The Lagos domesticated fish (LA), was used as control. All the broodstocks were kept in 12m^3 at the rate of 4 fishes per m^3, fed locally formulated diet (Crude protein, 35 %) for 3 months for gonadal development. The mature females were selected based on their swollen, reddish vent, well distended soft abdomen and extrusion of few eggs on gentle pressure of finger on the abdomen. Ripe males were also selected based on their reddish urinogenital Papilla. The males and females of each combination were weighed and females injected with synthetic ovaprim hormone. The ovaprim was administered intra-muscularly at the rate of 0.5 ml per kilogram of fish.

DISSECTION OF MALE BROODSTOCK FOR GONAD REMOVAL

The male broodstocks, after observing the latency period of 11 - 12 hours were removed from their respective holding troughs. They were placed dorsally on a wet towel, held firmly down to ensure careful removal of the testis. Using a sharp scapel, the abdominal cavity of the fish was dissected and testes were carefully removed at the ventral wall of the abdominal cavity. The testes were removed whole and cleaned with fluffy material.

STRIPPING AND EGG FERTILIZATION

After observing the 11 - 12 hours latency period, the female broodstock was removed from the trough carefully and held firmly with a wet towel at both ends, the abdomen was then pressed carefully to extrude the eggs into a dry bowl, and the stripping was done towards the fish vent. After stripping, the spent females were carefully returned into their respective troughs. About 0.6

g each of the egg mass was weighed out and the content of the testes (milt) was spilled on the eggs for fertilization (a drop of milt on each treatment), the eggs were then spread on 8 cm X 8 cm incubation tray placed in labeled spawning troughs already containing 1litre of water, and the fertilized egg was left in the spawning troughs, placed in 500 l of hatchery tough with flow-through water system, at water temperature between 26 and 31 °C.

EXPERIMENTAL CROSSES

The following generic combinations were carried out:

Parental crosses

♀ *Clarias gariepinus* (LL) X ♂ *larias gariepinus* (LL)

♀ *Clarias gariepinus* (AG) X ♂ *Clarias gariepinus* (AG)

♀ *Clarias gariepinus (*OT) X ♂ *Clarias gariepinus (*OT)

♀ *Clarias gariepinus* (MK) X ♂ *Clarias gariepinus* (MK)

♀ *Clarias gariepinus* (LA) X ♂ *Clarias gariepinus* LA (Control)

Intra-specific crosses

♀ *Clarias gariepinus* (LK) X ♂ *Clarias gariepinus* (MK)

♀ *Clarias gariepinus* (LK) X ♂ *Clarias gariepinus* (ON)

♀ *Clarias gariepinus* (LK) X ♂ *Clarias gariepinus* (AG)

♀ *Clarias gariepinus* (MK) X ♂ *Clarias gariepinus* (LK)

♀ *Clarias gariepinus* (MK) X ♂ *Clarias gariepinus* (ON)

♀ *Clarias gariepinus* (MK) X ♂ *Clarias gariepinus* (AG)

♀ *Clarias gariepinus* (AG) X ♂ *Clarias gariepinus* (LK)

♀ *Clarias gariepinus* (AG) X ♂ *Clarias gariepinus* (ON)

♀ *Clarias gariepinus* (AG) X ♂ *Clarias gariepinus* (MK)

♀ *Clarias gariepinus* (ON) X ♂ *Clarias gariepinus* (LK)

♀ *Clarias gariepinus* (ON) X ♂ *Clarias gariepinus* (MK)

♀ *Clarias gariepinus* (ON) X ♂ *Clarias gariepinus* (AG)

INCUBATION AND HATCHING

The fertilized egg masses were incubated in the spawning bowls for a period of 24 to 30 hours. After this period, most of the larvae emerged and some eggs did not hatch at all.

SURVIVAL

After hatching and determination of hatching rate, the un-hatched eggs were siphoned out of the spawning bowls in order to ensure the survival of the hatched ones and the water was partially changed with utmost care. 100 hatchlings were taken per replicate in each treatment and kept in 1L of water in triplicate experiment. The water was daily exchanged with daily reading of basic water quality parameters. The larvae were daily observed after hatching for 10 days to determine the rate of survival by estimating the dead and live fry.

The percentage fertilization, hatching and survival rates of each cross were calculated according to Oguntuase and Adebayo (2013) as follows :

Fertilization Rate (%) = [Number of fertilized eggs / Total number of eggs] x 100

Hatching Rate (%) = [Number of eggs hatched / Total number of eggs incubated] x 100

Survival Rate (%) = [Number of live hatchlings to larval stage / Total number of hatchlings] x 100

RESULTS

The results of water quality are presented in Table 1. The results showed that water quality parameter met optimum requirement for hatchery rearing of catfish. Results of the percentage fertilization, hatchability, and survival (10 days) of fingerlings of the crosses are as shown in Figure 1. The result revealed that collection from Onitsha had the highest values for fertilization (95.2 %), hatchability (87.6 %) and survival (66 %). This was followed by other crosses obtained from female of Onitsha strain namely ; ♀ ON X ♂ LK, ♀ON X ♂ MK and ♀ON X ♂ AG which had 95.8 %, 91.1 % and 88.8 % fertilization, 68.7 %, 61.9 % and 60.7 % hatchability and 61 %, 57 % and 63 % survival respectively. The domesticated strain had the least of these indices which were 81.9 %, 59.4 % and 51 % fertilization, hatchability and survival respectively. It was equally observed that the egg mass of the Onitsha strain was dark-brown while others were dirty green. Analysis of variance showed a significant difference ($\alpha < 0.05$) in the hatchability and survival of the fertilized eggs.

Table 1 : Water quality parameters of the various treatments during the experiment (minimum-maximum)

Paramètres de qualité de l'eau des différents traitements au cours de l'expérience (minimum-maximum)

CROSSES	pH	Temperature (^{0}C)	NH_3-N (mgL^{-1})	NO_2-N (mgL^{-1})	$CaCO_3$ (mgL^{-1})	DO (mgL^{-1})
♀ LK X ♂LK	7.9 – 8.6	27.8 – 30.2	0.4 – 0.6	0.05 – 0.2	98 - 100	4.0 – 8.0
♀ OT X ♂ OT	7.8 – 8.3	28.5 – 29.5	0.4 – 0.8	0.05 – 0.1	98 - 100	6.0 – 8.0
♀ MK X ♂ MK	7.9 – 8.3	28.5 – 30.0	0.4 – 1.5	0.05 – 0.2	60 - 98	4.0 – 8.0
♀ AG X ♂ AG	7.7 – 8.2	28.0 – 31.0	0.4 – 0.7	0.05 – 0.1	98 - 100	4.5 – 8.0
♀ LA X ♂ LA (Control)	7.0 – 8.2	28.2 – 30.0	0.5 – 0.8	0.05 – 0.1	70 - 98	4.5 – 8.0
♀ LK X ♂ MK	7.8 – 8.3	27.0 – 30.0	0.4 – 0.8	0.05	60 - 100	4.0 – 8.0
♀ LK X ♂ ON	7.9 – 8.4	27.5 – 30.5	0.4 – 0.6	0.05 – 0.1	60 - 100	3.0 – 6.0
♀ *LK* X ♂ AG	7.5 – 8.5	28.5 – 30.0	0.6 – 8.0	0.05 – 0.1	98 - 100	3.0 – 6.0
♀ *MK* X ♂ LK	7.9 – 8.4	27.0 – 30.5	0.6 – 0.8	0.05	60 - 100	4.0 – 8.0
♀ MK X ♂ ON	7.8 – 8.2	29.0 – 31.0	0.4 – 0.8	0.05 – 0.06	98 - 100	4.0 – 6.0
♀ MK X ♂ AG	6.1 – 8.3	28.0 – 30.5	0.6 – 1.5	0.05 – 0.2	80 - 100	3.5 – 6.0
♀ AG X ♂ LK	8.1 – 8.2	28.9 – 30.0	0.4 – 0.8	0.05 – 0.1	69 - 100	4.0 – 6.0
♀ AG X ♂ ON	8.0 – 8.8	28.0 – 29.5	0.4 – 1.5	0.05	60 - 100	4.0 – 8.0
♀ AG X ♂ MK	7.9 – 8.6	27.0 – 30.5	0.4 – 0.6	0.05 – 0.1	60 - 100	3.0 – 6.0
♀ ON X ♂ LK	7.9 – 8.1	28.0 – 30.5	0.4 – 0.8	0.05 – 0.8	68 - 100	4.0 – 6.8
♀ ON X ♂ MK	7.8 – 8.1	29.0 – 30.7	0.4 – 1.5	0.05 – 0.6	60 - 100	4.0 – 6.0
♀ ON X ♂ AG	7.9 – 8.3	27.5 – 39.0	0.4 – 0.8	0.05 – 0.1	96 - 100	4.5 – 6.0

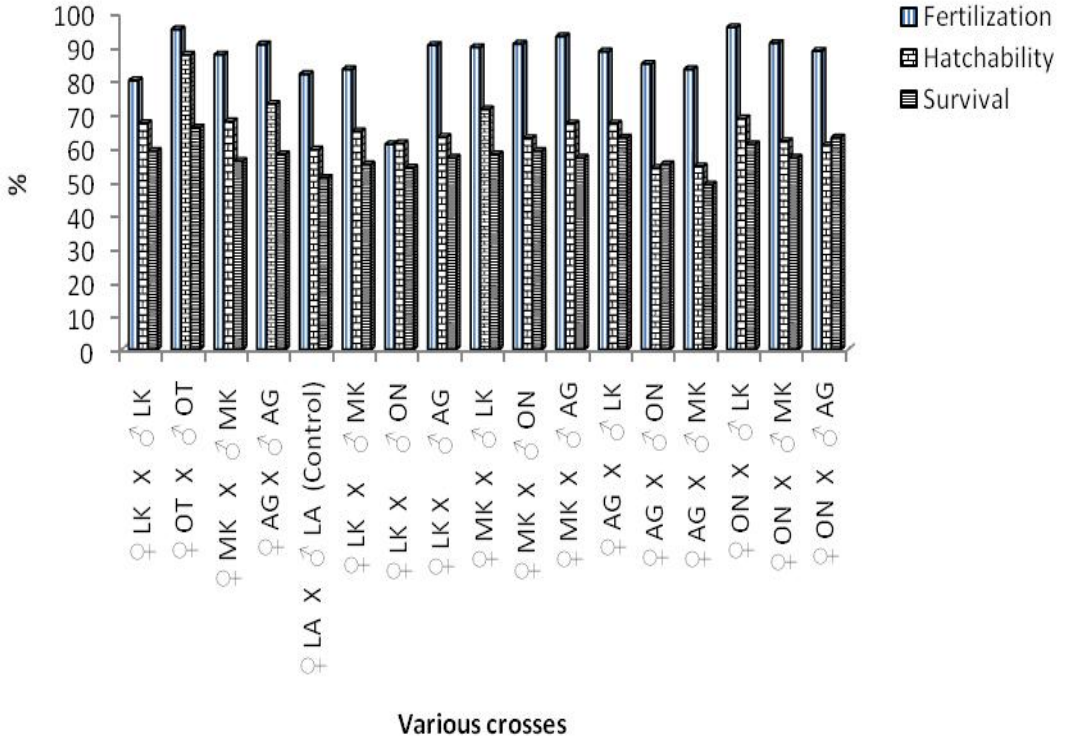

Figure 1 : Percentage fertilization, hatchability and survival of seventeen (17) intraspecific crosses of *Clarias gariepinus*

Pourcentage de fertilisation, éclosion et survie de dix-sept (17) croisements intraspécifiques de Clarias gariepinus

DISCUSSION

The feasibility of crosses of natural populations and domesticated strains *C. gariepinus* and its reciprocal cross-breeding was demonstrated in the present study. The results above indicated that cross made from Onitsha in Anambra state had the best breeding performance in terms of fertilization, hatchability and survival. Similarly, crosses obtained from female of Onitsha strain with other strains from Lokoja, Agenebode and Markudi equally perform well. The poor performance of domesticated strain with respect to these indices is expected, considering the degree of inbreeding depression found in catfish farmed in Nigeria. This artificial propagation through intraspecific hybridization will facilitate the production of superior strain of the Clariid catfish. This is in agreement with Akankali *et al*. (2011) who reported that apart from being able to obtain quality seed, the artificial propagation technique can also be used to develop strains superior to their ancestors by the methods of selective breeding and hybridization.

The results of the finding of the breeding performance revealed that natural populations of catfish performed better than domesticated strains cultured in Nigeria farms. Previous studies revealed that the cultured strains performed better than natural populations in terms of growth performance (Megbowon *et al*., 2012). This was probably due to the introduced Dutch strain brought to Nigeria from the Netherlands whose gene pool has mixed with our indigenous domesticated catfish. Although this has resulted in increased production of catfish in Nigeria, its genetic consequences may not be pleasant. This has led to inbreeding escape of the progeny into our water may have unforeseen consequences (FAO, 1996). There is therefore the need to explore the natural populations for genetic development through selective breeding, hybridization and gene transfer.

CONCLUSION

Although the strain from Onitsha showed the best breeding performance, the strain showed different phenotypic character in terms of egg colour. What could have accounted for this is unknown. There might be need to evaluate the

water, sediment and fish for heavy metals to see where the variation is coming from, whether genetic or environmental.

There is need to carry out protein and gene profiling of these natural population to identify the proteins and gene responsible for this variation in breeding performances if variations are traceable to genetic factors. Furthermore, we need to assemble strains from a number of other natural populations from other rivers and stream besides the River Niger/Benue system.

REFERENCES

Akankali J.A., Seiyaboh E.I., Abowei J.F.N. 2011. Fish Hatchery Management in Nigeria. Advance Journal of Food Science and Technology 3 (2) : 144 - 154.

Ataguba G.A, Annune P.A., Ogbe F.G. 2009. Induced breeding and early growth of progeny from crosses between two African clariid fishes, *Clarias gariepinus* (Burchell) and *Heterobranchus longifilis* under hatchery conditions. Journal of Applied Bios ciences 14 : 755 – 760.

de Graaf G.J., Galemoni F., Banzoussi B. 1995. The artificial reproduction and fingerling production of the African catfish *Clarias gariepinus* (Burchell 1822) in protected and unprotected ponds. Aquaculture Research 26 : 233 - 242.

Dunham R.A., Majumdar K., Hallerman E., Bartley D., Mair G., Hulata G., Liu Z., Pongthana N., Bakos J., Penman D., Gupta M., Rothlisberg P., Hoerstgen-Schwark G. 2001. Review of the status of aquaculture genetics. *In* : Aquaculture in the Third Millennium. Subasinghe R.P., Bueno P., Phillips M.J., Hough C., McGladdery S.E., Arthur J.R. (eds). Technical Proceedings of the Conference on Aquaculture in the Third Millennium, Bangkok, Thailand, 20 - 25 February 2000. NACA, Bangkok and FAO, Rome, pp. 137 - 166.

FAO. 1996 . Handbook on the artificial reproduction and pond rearing of the African catfish *Clarias gariepinus* in Sub-saharan Africa. FAO fisheries technical paper, 362 p.

Macharia S.K, Ngugi C.C, Rasowo J. 2005. Comparative Study of Hatching Rates of African Catfish (*Clarias gariepinus* Burchell 1822) Eggs on Different Substrates. NAGA, World Fish Center Quaterly 28 (3 & 4) : 23 - 26.

Megbowon I., Fashina-Bombata H.A., Akinwale M.M.-A., Hammed A.M., Okunade O.A., Mojekwu T.O. 2012. Breeding Performance of *Clarias gariepinus* Obtained From Nigerian Waters.

Oguntuase B.Gb., Adebayo O.T. 2013. Sperm Quality and reproductive performance of male *Clarias gariepinus* induced with synthetic hormones (Ovatide and Ovaprim). International Journal of Fisheries and Aquaculture 6 (1) : 9 - 15.

Olaleye V.F. 2005. A review of reproduction and gamete management in the African catfish, *Clarias gariepinus* (Burchell 1822). Ife Journal of Science 7 (1) : 63 - 70.