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Reproductive Success of Intra-specific Hybridization between Two Strains of *Clarias gariepinus* from Katsina-ala and Gboko, North East, Nigeria

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Authors' contributions

This work was carried out in collaboration between both authors. Both authors read and approved the final manuscript.

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ABSTRACT

The intra-specific hybridization investigation was carried out at the Federal University Wukari, Fisheries and Aquaculture research farm. *Clarias gariepinus* broodstock was obtained from Katsina-Ala and Gboko in order to investigate the reproductive success among the inbred and crossbred individuals. Data on reproductive success (fertilization, hatchability, and survival), and the frequency of shooters were acquired. The result showed that the crossbreds of ♂G x ♀K had a greater percentage fertilization of 66.3% than ♂K x ♀G (62.03%). Inbred of ♂K x ♀K and ♂G x ♀G showed higher hatchability of 80.24% and 73.47%, respectively, whereas their crosses showed no significant difference. Because there is no statistically significant difference in survival at initial feeding between inbred and crossbred, the inbred performed better at the advanced fry stage. The frequency of shooters was higher in inbred of ♂G x ♀G with 4% and crossbred of ♂K x ♀G with

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3.6%, while the inbred of ♂K x ♀K has a lower value of 3.4% and crossbred of ♂G x ♀K with 3.0%. Katsina-Ala (♂K x ♀K) inbred produced the best results. As a result, broodstock selection and management are critical in avoiding the prevalence of unfavorable traits in *C. gariepinus* for Aquaculture.

Keywords: Clarias gariepinus; Gboko; hybridization; intra-specific; Katsina-Ala; North East.

1. INTRODUCTION

In the world diet, fish is frequently referred to as an indispensable source of animal-based protein. Due to a number of characteristics, including their capacity for rapid growth, tolerance of relatively low water quality, and ability to consume a variety of agricultural byproducts (They are omnivorous), catfish (Family *Clariidae*) are the most widely cultivated fish in Nigeria. They had an advantage due to their great disease resistance and tolerance to draught. The primary goal of fish farming is to meet the increasing demand of the human population, which relies on it as a major source protein [1]. This fish has a seasonal gonadal maturation that coincides with the wet season.

Clarias gariepinus maturation processes in nature are mainly regulated by annual fluctuations in water temperature and photo-periodicity, and the final triggering of spawning is frequently triggered by an increase in water level owing to rainfall. If kept in ponds with water temperature above 22^0C the female African catfish has a fully developed ovary that retains "ripe" eggs all year. A "ripe" female eggs account for 15-20% of her body weight. The African Catfish does not spawn spontaneously in captivity because the environmental variable such as the rising water levels and inundation of shallow areas do not occur on the fish farms. Various authors have reported reduced hatching rates for *C. gariepinus* under natural spawning condition. Fertilization, hatching and early larval survival are critical for effective African catfish aquaculture.

The growing human population and reports of enormous numbers of undernourished or malnourished people, particularly in developing nations, have made food production a major global concern. The maximum sustainable fishing limit has been reached in the majority of our lakes, rivers and oceans. As a result, Aquaculture will be required to overcome the fish supply shortfall. Hence, other cultural methods are required [2,3,4]. Induced breeding via hormone treatment and artificial incubation of fertilized egg has the advantages of faster fertilization and hatching, better conditions for larva growth and survival to fingerling, and better protection of larvae from unfavorable environmental conditions and predators [5].

Fish hybridization has been practiced for centuries in Africa. Hybridization has occurred in the African catfishes; *C. gariepinus*, *C. anguillaris*, *Heterobranchus bidorsalis* and *H. longifilis* [6,7]. The crossing or mating of fish of the same species and genus but from different places, regions and biological zones is known as Intra-specific hybridization. Intra-specific hybridization of fish has been proposed as a method of combining beneficial features from two or more species to produce hybrids that outperform both parents' species. Because hybrids among fishes are sterile, the primary purpose of Intra-specific hybridization is to top cross hybrid for species of different genotypes purely for production [8]. Many farmers prize fast-growing species resulting from these hybrids.

As a result, the purpose of this study is to examine the reproductive indices (i.e; fertilization, hatchability and the survival rate) and frequency of shooters among *C. gariepinus* strains and their reciprocal crosses derived from Gboko and Katsina Ala.

2. MATERIALS AND METHODS

2.1 Description of the Experimental Site

The experiment was carried out in the Fisheries Research Unit, Department of Fisheries and Aquaculture, Federal University Wukari, Taraba State, which is located at latitudes $7^0$52' 17.00" N and longitudes 9^0 46' 40.30" E of the equator and has an elevation of 189m above level.

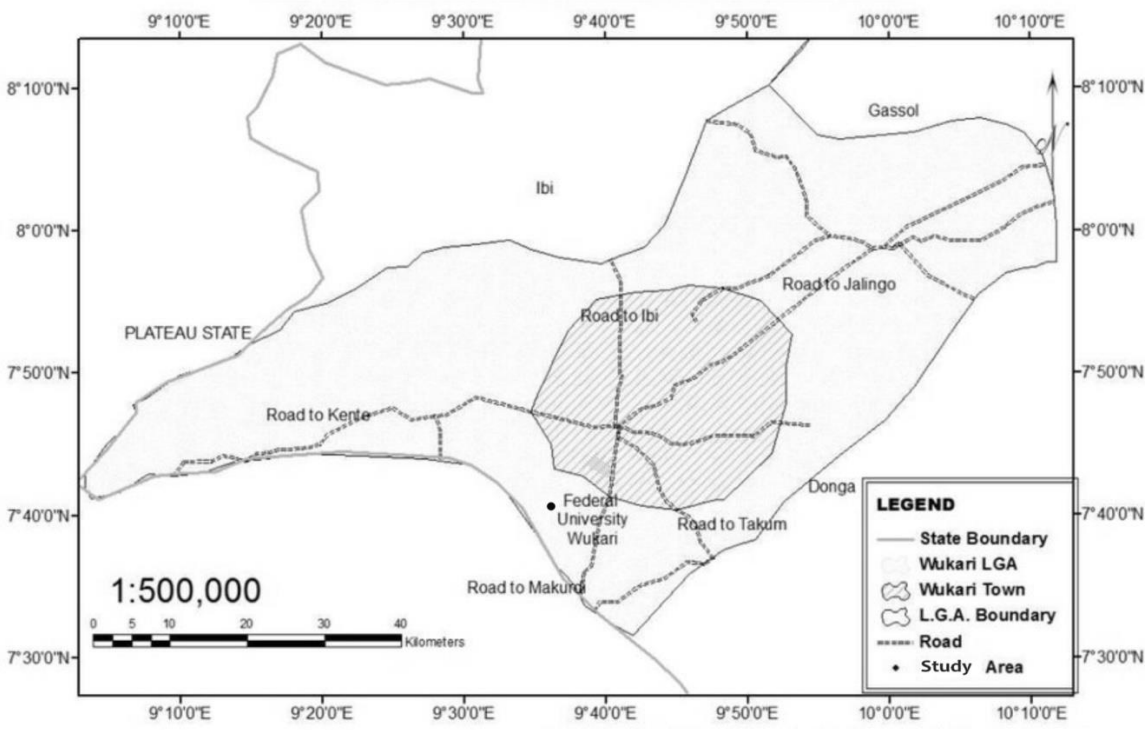

Fig. 1. A Map of the experimental site
Source: Ministry of land and survey Jalingo

2.2 Spawning Materials Acquisition

For this investigation, eight (8) mature (brood stock) African catfish (four males and four females) were purchased from Katsina-Ala college of Agriculture and Anointed farm Gboko. The hormone was obtained from the Makurdi Agro-service centre in Benue State. The broodstocks employed weighed between 800 g – 1000 g on average.

2.3 Broodstock Selection

The exterior morphological criteria specified by [5] were used to select all brood fish.

2.4 Procured Spawner Transportation and Acclimation

C. gariepinus broodstocks were transported in two (2) fifty litre (50Ltrs) jerricans to the Fisheries and Aquaculture research farm and acclimatized in fibre reinforced holding water tanks for 24 hours before artificially inducing breeding was carryout.

2.5 Hypophysation and Artificial Hybridization

The fish were sexed and classified as males or females based on the location of their genital organ. The gravid female's weight was assessed and induced intramuscularly at 0.5ml/kg of fish below the dorsal fin, towards the skull at a 45º angle. To prevent retraction or leakage of the synthetic hormone, the injected site were finger robbed. For ten (10) hours, the injected fish were conditioned in several mobile holding tanks.

2.6 Milt Collection

The milt was harvested after ten (10) hours of latency period by sacrificing the male base on the experimental cross combination. The male's testes lobes were removed, thoroughly cleaned with tissue papers, and stored in a labeled container for fertilization.

2.7 Eggs Extraction and Artificial Fertilization

The female brooders were swept dry with a towel to avoid the egg coming in touch with water,

which could shut up the micropyle. Gently Pressing the abdomen of the injected female brooder allowed ovulated eggs to seep out of the virginal orifice, and the eggs were collected in stainless steel dishes with labels and weighed individually, while the males testes were cut open with scissors and the milt was squeezed out, then 0.9% saline solution (Nacl) was added to activate the milt and facilitate fertilization, and the milt was poured into stripped eggs in labeled bowls.

2.8 Crosses for Experimentation

The generic combinations listed below were tested;

Inbreds	Crossbreds
♂K x ♀K	♂K x ♀G
♂G x ♀G	♂G x ♀K

Note: ♂K x ♀K - Katsina Ala Male X Katsina Ala Female; ♂G x ♀G - Gboko Male X Gboko Female; ♂K x ♀G - Katsina Ala Male X Gboko Female; ♂G x ♀K - Gboko Male X Katsina Ala Female

2.9 Eggs Incubation

The fertilized eggs were incubated in fiber tanks. Both the parental and the intra-specific crossovers were reproduced. The fertilized eggs were equally placed in a single layer on a net suspended in water. The hatching rate was tracked during 24 to 36 hours. In each replication, the number of live larvae hatching was counted.

2.10 Indoor Experiment Setup and Daily Hatchling Survival

After calculating the pooled weight, 500 hatchlings were gathered and deposited in each flow through container. For a period of eight (8) weeks, the survival of fries in each container per treatment was measured daily and Weekly, while pooled weight, pooled length, and final survival were measured on the 56th day. Each treatment is done in threes.

2.11 Feeding of Larvae

Following hatching and yolk absorption, fry were fed Artemia 0.2mm for 14 days followed by other artificial feed for 6 weeks. During these times, weight and length of fries were measured on a weekly basis.

2.12 Data Collection

The following formulas were used to collect data on the number of fertilized eggs, hatched eggs, survival rate, and frequency of shooters: [9,10].

$$\% \text{ Hatchability} = \frac{\text{Number of hatchlings}}{\text{Number of fertilized eggs}} \times 100$$

$$\%\text{Survival rate (SR)} = \frac{Ni}{No} \times 100$$

Where: Ni is the total number of fry at the end of the experiment; N_0 is total number of fry at the beginning of the experiment

The percentages of shooters were computed using [11] methods.

$$\%\text{Shooters} = \frac{\text{Number of shooters}}{\text{Number of fry}} \times 100$$

2.13 Parameters of Water Quality

Because water is used in fish culture, its quality deteriorates quickly and necessitates intensive upkeep. The filthy water was drained and refilled with cleaned water to allow for aeration. Temperature, pH value, dissolved oxygen, ammonium level and other factors all have an impact on water quality. They were measured *in situ* using a thermometer to determine temperature and a pH reagent to determine pH. Other parameters were measured by immersing the measuring instrument probes in the water at a depth of about 4cm in the middle of the cultured water and taking readings from the meters when equilibrium was reached.

2.14 Data Analysis

The data from each treatment were analyzed using one-way analysis of variance (ANOVA). Duncan (1985) employed the Duncan Multiple Range Test (DMRT), to determine mean difference (p0.05) using SPSS version 20.0.

3. RESULTS

3.1 Mean Fertilization, Hatchability and Survival at Day 3 of Intra-specific Hybridization between Katsina Ala and Gboko Strains of *C. gariepinus*

Table 1 display the mean fertilization, hatchability and survival of *C. gariepinus* strains Katsina Ala and Gboko on day 3 of intra-specific

hybridization. Fertilization (66.30%), hatchability (80.24%) and day 3 survival (683) were highest in ♂G x ♀K, ♂K x ♀K and ♂K x ♀K respectively. There were significant variations in fertilization rate, hatchability rate and survival at day 3 between inbreds and crossbreds (P0.05).

3.2 The Survival Rate of *C. gariepinus* Fry

Fig. 2 depicts the weekly survival of inbreds and crossbreeds. The highest values of weekly survival over an 8-week period were achieved in ♂K x ♀K (62) and ♂G x ♀G (53) whereas the lowest were obtained in ♂K x ♀G (49) and ♂G x ♀K (42).

Table 2 display the percentage survival rate after eight weeks for inbreds and crossbreds, as well as frequency of shooters. The highest percentage survival rate was reported in ♂K x ♀K (12.4%), followed by ♂G x ♀G (10.6%), and the lowest percentage survival rate was recorded in ♂K x ♀G (9.8%) and ♂G x ♀K (8.4%).

Table 1. Fertilization, hatchability and survival rate at day 3 of intra-specific hybridization between *C. gariepinus* strains of Katsina Ala and Gboko

Genetic Crosses	% Fertilization	% Hatchability	Survival at Day 3
♂K x ♀K	59.96±0.00^{a}	80.24±10.00^{a}	683±50.00^{b}
♂G x ♀G	58.83±5.00^{a}	73.47±0.00^{a}	624±0.00^{ab}
♂K x ♀G	62.03±7.00^{a}	72.04±2.00^{a}	593±3.00^{ab}
♂G x ♀K	66.3±0.00^{a}	70.24±0.00^{a}	545±0.00^{a}

Means with the same superscript letter in the same column are not significantly different at P<0.05
Note: ♂K= Katsina Ala male; ♀K= Katsina Ala female; ♂G= Gboko male; ♀G= Gboko female

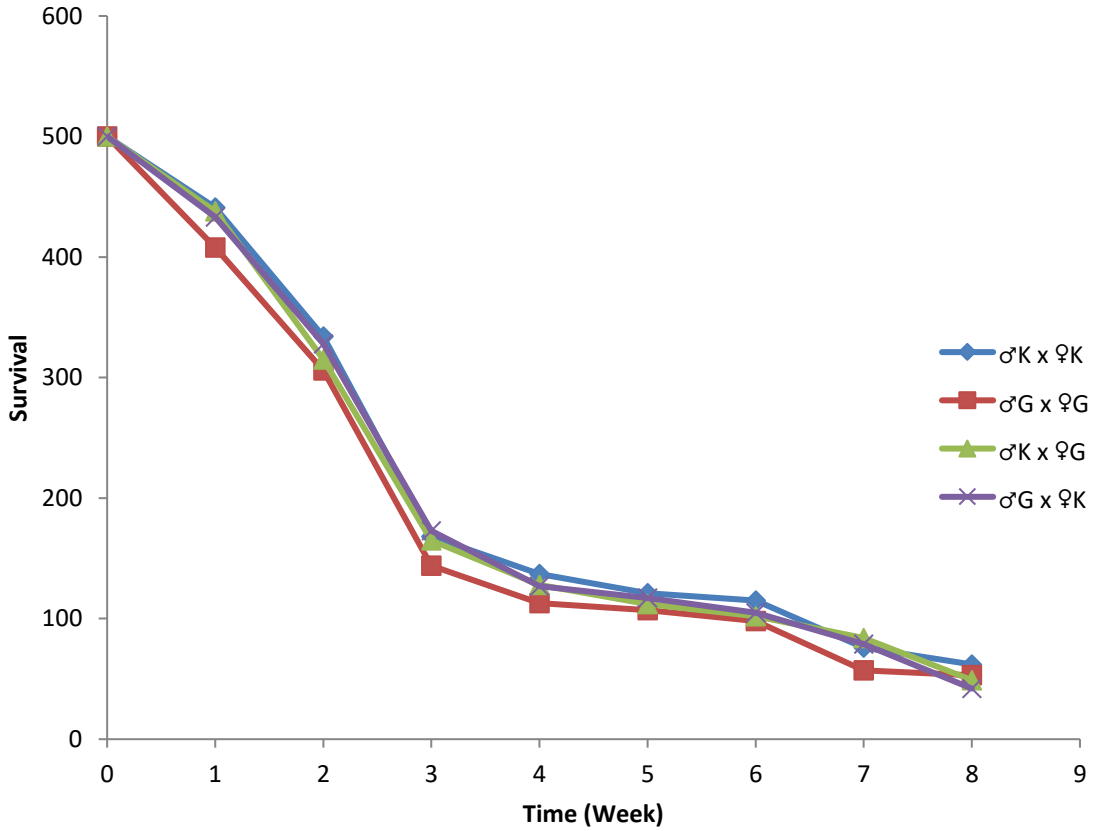

Fig. 2. Weekly Survival of inbreds and crossbreds *C. gariepinus* during eight weeks of rearing

Table 2. Percentage survival and Frequency of shooters after eight weeks of rearing

Genetic Crosses	Number of fish	Final number of survival	%Survival rate	%Frequency of shooters
♂K x ♀K	500	62	12.4	3.4
♂G x ♀G	500	53	10.6	4.0
♂K x ♀G	500	49	9.8	3.6
♂G x ♀K	500	42	8.4	3.0

Note: ♂K= Katsina Ala male; ♀K= Katsina Ala female; ♂G= Gboko male; ♀G= Gboko female

Table 3. Water quality parameters

Genetic crosses	Temp	pH	DO	NH_3	NO_2	NO_3
♂K x ♀K	28.08	8	5.3	1.2	0.02	1.3
♂G x ♀G	28.64	8	5.3	1.5	0.02	1.26
♂K x ♀G	27.66	8	5.3	1.2	0.02	1.43
♂G x ♀K	27.9	8	5.3	1.5	0.02	1.36

Note: ♂K= Katsina Ala male; ♀K= Katsina Ala female; ♂G= Gboko male; ♀G= Gboko female

3.3 Water Quality Parameters of the Rearing Medium

Table 3 displays the mean water quality parameters during the course of the 8-week investigation. Temperatures varied from 27.66 to 28.64 degrees Celsius, pH was 8.0, Dissolve oxygen was 5.3, Ammonia was 1.2 - 1.5, Nitrite was 0.02, and Nitrate was 1.26 - 1.43.

4. DISCUSSION

4.1 Fertilization and Hatchability

Catfish hybridization has enormous future potential; a better catfish hybrid is equating to a better catfish aquaculture, as well a contribution to world food security. Higher fertilization rates of 66.30% and 62.03% were observed in crossbreds of ♂G x ♀K and ♂K x ♀G when compared to inbred ♂K x ♀K and ♂G x ♀G, which are lower than the findings of Shah et al. [12] who reported lower fertilization rates of 86-89% when Jamuna Padma were crossed with hatchery strains, whereas pure Jamuna strain which showed the highest fertilization rate (95%). There are no statistically significant differences in percentage fertilization. The greater fertilization rate in crossbreds could be attributed to changes in their genetic make-up. However, when compared to the crosses ♂K x ♀G (72.04%) and ♂G x ♀K (70.24), the inbred of ♂K x ♀K and ♂G x ♀G had hatchability rates of 80.24% and 73.47%, respectively, which agrees with the findings of Olufeagba et al. [13] who obtained a low hatchability rate among the reciprocal hybrids (41.0%) when compared with the parental crosses (94.0%). Tilahun et al. [14] and Sayeed [15] noticed similar tendencies. Variations in seasons can also lead to differences in hatching rates, as Shah et al. [12] and Ochokwu et al. [7] correctly found.

4.2 Survival of *C. gariepinus*

During eight (8) weeks of upbringing the survival rate of Katsina-Ala and Gboko inbreds was higher than that of their crossbred of ♂K x ♀G and ♂G x ♀K (Table 1). This could be ascribed to its greater adaptability to the environment when compared to crossbreds. This contradicts the findings of Tilahun et al. [14], who found that pure *C. gariepinus* and *C. batrachus* exhibited lower survival rates for 60 days than their hybrids.

The inbred of ♂K x ♀K had the highest percentage survival of 12.4%, while the crossbreds of ♂G x ♀K had the lowest percentage survival of 8.4%, both of which are lower than the findings of Adene et al. [16], who reported a percentage survival of 63.33% - 91.67%. This could be related to changes in environmental conditions. Similarly, Jothilakshmanan and Karal Marx [17] found lower survival (0.8 and 0.9%) in hybrids of *Hetropneustes longifilis* and *Clarias batrachus* due to high incidence of mortality of hatching during the transition from endogenous to external feeding. Legendre et al. [18] and Tilahun et al. [14] concluded that the survival of hybrids is heavily impacted by the maternal parent.

4.3 Shooter Frequency and Fry Size Variations

The main issue in catfish production schemes is growth heterogeneity and the cannibalistic inclination of faster growing individuals (Shooters) which has detrimental impact on yield and industry remuneration. Gamete variation has a considerable impact on shooter heterogeneity, frequency, and weight. The higher frequency of shooting in inbreds could be linked to cannibalism among the fry as well as environmental variables. However, the acquired result is lower than the findings of Oyebola et al. [11] who reported a frequency of shooters ranging from 5.9% to 19.5%.

4.4 Water Quality Parameters

Careful monitoring of the water quality parameters was required to keep circumstances below acceptable limit, as indicated by Emmanuel et al. [19]. The water parameters values for these observations are within the permissible levels for pisciculture.

5. CONCLUSION

The intra-specific hybridization investigation revealed information on various elements of reproduction indices; thus, the selection of inbreds for breeding programme employed in this study can improve the genetic make-up of *Clarias gariepinus*, increasing potentials for Aquaculture production.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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