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Research Article

Determination of Sex-Reversal Rate and Growth Performance in Diallel Hybrids of Nile Tilapia (*Oreochromis niloticus*) and Blue Tilapia (*Oreochromis aureus*)

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Abstract: A total of 200 fish samples (100 *Oreochromis niloticus* (Nile tilapia) and 100 *Oreochromis aureus* (Blue tilapia)) were used for this research aimed to produce a population of tilapia species that is skewed toward the male population. Three mating periods were carried out in total. The males were removed from the hapa after each round of egg production to allow repercussion. Fries from the experimental units were collected, counted, and stocked in a tank with a dimension 3 x 1 x 4 m^2 and were fed to satiation while maintaining the basic water quality. The sex ratio data were subjected to student t-test analysis, while that of growth parameters were examined by ANOVA on Spss version 25. Results obtained showed a true hybridization between Nile tilapia and Blue tilapia that produced offspring that skewed toward the male population. A male skewed population was produced through interspecific crossing of two related strains of tilapia. This pattern repeated itself throughout the times when mating pairing was initiated. At the end of the trials, the hybrids and their reciprocals produced a higher number of male fish compared to the female. Hybrid 1 produced 93% male, while hybrid 2 produced 71% male. Furthermore, better performance was recorded for the hybrids compared to their pure strain despite the lower feed intake. Conclusively, a male skewed population is best produced through interspecific crossing of two related strains of hybrid tilapia.

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1. Introduction

Tilapia is regarded as an aquatic chicken because of its prolific nature (Menaga and Fitzsimmons, 2017). They are the third most important aquaculture product, just behind salmon and carp (Naylor et al., 2021). With the genus Oreochromis consisting of *O. niloticus*, *O. aureus,* and *O. mossambicus,* they remain one of the most widely cultured. Tilapia culture is popular because of its ability to thrive in most aquaculture systems (Nwachi and Esa, 2016). Khaw et al. (2016) infer that tilapia has almost all the traits that are associated with a fish with excellent culturable traits. Despite the fact that tilapia has the potential to do well in culture, its production in Africa remains at a minimal level (Goni et al., 2020; Ovharhe et al., 2020).

Tilapia could be cultured in different environments because of its ability to adapt to different environments. It is of note that strains of tilapia could be found in every body of water ranging from fresh water to marine environments. They are easy to culture, while their early maturation makes them it a choice for stock improvement. Although the aforementioned made it clear that tilapia is a fish that is farmed because of its desirable characteristics, drawbacks do exist while culturing this fish (Ng and Romano, 2013). Tilapia is known to quickly reproduce and populate the water body that it is introduced to (Oladimeji et al., 2015). This gives rise to a population of mini fish that are of little or no economic value.

Proliferation implies that there isn't enough food to go around for the army of mini fish that are produced. At a certain time in Southeast Asia, tilapia is regarded as weed fish (fish to be eliminated) before stocking the folk's fish as a result of the fish not reaching market size in culture (Nwachi et al., 2020). However, the ability of the male fish to reach market size on time made it possible for breeders to focus on raising monosex tilapia with a focus on the male fish because tilapia exhibit sexual dimorphism (Novelo et al., 2020).

A study by Nwachi and Esa (2016) infers that hybridization is a process of combining different strains with the purpose of producing a hybrid. It also involves mating fish with traits of interest, especially if they could be inherited. Strain or line crossing could be initiated depending on the need. Production of monosex tilapia has been a success because fertilization is external, making it possible to produce fish with a different number of chromosomes (Lago et al., 2016). Haque et al. (2016); Lozano et al. (2014) reported the use of interspecific breeding to produce hybrids of interest, which is in line with the way in which tilapia can easily accept related strains. A number of crosses have been used to produce male skewed sex individuals (Lahav and Lahav 1990; Rosenstein and Hulata 1994; Wohlfarth 1994; Verdegem el al., 1997; El-Hawarry 2012; Felix et al., 2019).

Despite some success recorded by the crossing of interspecific strains to achieve a population that is skewed toward males, proper documentation on the use of reciprocal mating and the growth rate achieved has not been fully exploited. Hence, there is a need to carry out diallel pairing of pure strains of *O. niloticus* to *O. aureus* to produce a high male sex ratio and a male of *O. aureus* to *O. niloticus to* produce a reciprocal.

The general objective was to produce a population of fish skewed towards a male population, with the specific objective of the pairing of male *O. aureus* (Blue tilapia) to female *O. niloticus* and the reciprocal. Also, evaluated the growth rate and colour variation of both the pure strain and the hybrid.

2. Material and Methods

The work was carried out at the research center of the Department of Aquaculture and Fisheries Delta State University Abraka, Nigeria. The candidate fish was collected from notable tilapia cage culture farms in River Benue and Ase creek. A total of 100 of each strain of *Oreochromis niloticus* (Nile tilapia) and *Oreochromis aureus* (Blue tilapia) were collected, respectively. The weight and length of the 12 months old female broodstocks range from 48.70 g to 168.50 g at 13.6 cm – 21.40 cm, while the 14 months old male range from 50.60 g to 190.60 g at 14.40 cm – 23.50 cm.

A pure strain of this stock was required; hence, prior to purchase for the research, they were subjected to further identification to species level with the aid of microsatellite by a group of experts at the African Bioscience Laboratory Ibadan Nigeria and Inqaba, West Africa after which a total of 36 females and 12 males that were identified as the pure strain was selected. A man-made lake of 100 sq.m was chosen for the work. Hapa of $1x1x1m^2$ was made from micro mesh size net that could retain fry but allow free flow of water. Water quality parameters such as temperature, pH, and dissolved oxygen were constantly monitored to ensure they were within the acceptable levels 27 °C, 7.5, and 6.9 mg/L^{-1}, respectively, as reported by Mohammadi et al. (2021).

Paring was at a ratio of 1:3 for male and female broodstocks. Mouth clipping was carried out on the male fish to reduce the incidence of cannibalism. The female fish is stocked for five days in the hapa before the introduction of the male fish. The male fish is crossed with the female fish at random to produce half-sib so that phenotypic and heritability correlation could be carried out. Mating of male to female *O. niloticus* (pure strain 1) and male to female *O. aureus* (pure strain 2) made up treatments 1 and 2, while male *O. aureus* to female *O. niloticus* (hybrid 1) and male *O. niloticus* to female *O. aureus* (hybrid 2) made up treatments 3 and 4. All treatments were in triplicates.

2.1. Spawning and fry collection

The paired broodstocks were fed to satiation twice a day at 07:00 and 04:00 GMT with commercial feed (Stretches) of size 3 mm at 25% crude protein. Free swimming fries were scoped out of the water. The mouths of the brooders were also examined for fries and fertilized eggs that were collected and transferred to the hatchery for completion of the hatching process. A total of three mating periods were carried out. The male fish was removed from the hapa after each round of egg production to allow repercussion.

2.2. Fry rearing and sexing

Fries from the experimental units were counted and stocked in a tank of $3x1x4m^2$. They were fed to satiation while maintaining the basic water quality as described by Boyd and Lichtkpper (2002). The fries were fed with artemia and green algae from the pond in a progressive manner and were subsequently fed with various sizes of commercial feed to satiation. Three different culture tanks were used in raising each set that was produced at the same time to reduce the incidence of cannibalism. After a period of twelve weeks, methylene blue and hand lenses were used in separating the sexes. The male fish has only one opening through which both milt and urine pass, while the females have different openings for eggs, and urine exists.

2.3. Data analysis

The sex ratio was calculated with the aid of the Independent Groups t-test, while growth analysis was done by Anova on IBM SPSS v25 while Duncan Multiple Range was used to differentiate the means (Irabor et al., 2021).

3. Results

In Table 1, the result of interspecific breeding infers that a true hybridization between Nile tilapia and Blue tilapia produces a population that is skewed toward the male population. The mating of the pure strain produced both populations; mating between male and female pure 1 produced 126 males and 174 females in the first trial. At the end of the last trial, 470 males and 592 females were produced from a total of 1062. Pure 2 produced 414 males from a total of 1103 after the third trial. Similarly, the trial produces a 44% male population for pure 1 and a 37% for pure 2. Hybrid 1 produces a total of 925 males and 70 females from a total of 995. This makes it 93%, while the hybrid 2 produces 71% from a total of 993, making it 663 males and 270 females.

Table 1. Number of male fish from the trials

Trails	Pure strain 1	Pure strain 2	Hybrid 1	Hybrid 2
1	126(300)	115(318)	206(220)	198(225)
2	146(362)	143(355)	339(355)	215(310)
3	198(400)	156(430)	380(420)	250(398)
Average	470(1062)	414(1103)	925(995)	663(933)
Percentage	44	37	93	71

Values without superscripts are the same.

The mating of Nile tilapia to blue tilapia gives strains at a different number of days. It takes a mean of 24.16 days for the first trail to produce fries for pure 1 and 24.67 days for pure 2. The average number of days for pure 1 and pure 2 was 15.97 and 16.13, respectively. In the first mating, the hybrid recorded success after 28.21 and 28.47 days, respectively, for the hybrids 1 and 2. However, the average number of days for success to be recorded was 19.21 for pure 1 and 19.6 for pure 2. Although the numbers of successful days for the two pure strains were the same at the first mating, they were significantly different from the hybrids (Table 2). This trend repeated itself throughout the times that pairing for mating was initiated (Table 3).

Table 2. Mean number of fries produced at each crossing

Mean (Days)	Pure strain 1	Pure strain 2	Hybrid 1	Hybrid 2
1	24.16 ± 0.33^{a}	24.67 ± 0.63^{a}	28.21 ± 0.54^{b}	28.47 ± 0.34^{b}
2	13.44 ± 0.14^{a}	13.87 ± 0.76^{a}	16.31 ± 0.65^{b}	16.63 ± 0.5^{b}
3	10.32 ± 0.23^{a}	9.85 ± 0.85^{a}	13.12 ± 0.11^{b}	13.71 ± 0.41^{b}
Average	15.97	16.13	19.21	19.6

The superscripts were used to show different averages, and values with the same superscripts are the same.

Table 3. Growth Performance of Interspecific hybridization

Mean (Days)	Pure strain 1	Pure strain 2	Hybrid 1	Hybrid 2
Initial weight (g)	48.01 ± 0.67	48.00 ± 0.75	48.02 ± 0.32	48.00 ± 0.41
Final live weight (g)	82.23 ± 0.23^{a}	77.19 ± 0.11^{a}	91.62 ± 1.54	89.29 ± 1.23
MWG (g)	34.22 ± 0.77^{a}	29.19 ± 0.65^{a}	43.60 ± 0.71^{b}	41.29 ± 0.12^{b}
MDWG (g/day)	0.36 ± 1.23^{a}	0.31 ± 1.41^{a}	0.46 ± 0.33^{b}	0.44 ± 0.51^{b}
PWG (%)	66.58 ± 0.71^{a}	57.73 ± 0.22^{a}	86.52 ± 0.62^{b}	82.51 ± 0.31^{b}
TFI (g)	114.72 ± 1.43^{a}	114.02 ± 1.67^{a}	117.19 ± 1.56^{b}	116.42 ± 1.89^{b}

The superscripts were used to show different averages, and values with the same superscripts are the same; MWG is the mean weight gain; MDWG is the mean weight gain per day; TFI is the total feed intake.

4. Discussion and Conclusion

The reports by Assis et al. (2017); Lago et al. (2016); Nwachi and Esa (2016) inferred that tilapia exhibits sexual dimorphism, making the male fish much more valuable than the female, and the practice of all-male fish, culture is an option to adapt to if the goal of the culturist is to produce food sources for human. A number of ways that could lead to the exclusive production of a population that is skewed toward the male proportions were evaluated.

However, in this study, the use of interspecific breeding by pairing Nile tilapia with Blue tilapia was examined. Pure strains of each of the base parents were crossed, and the resulting fries were examined. Nwachi et al. (2020) reported the sexual difference that was shown at the mating of reddish coloured fish to their wild counterparts, with the conclusion that difficulties (delayed) in producing fries take place because of differences in their colours, despite the fact that they are from the same strain. In this study, more variation was observed in the hybridization matches (produced from pairing the interspecific and pure strains) when compared to that of the pure strain.

It is of note that the interspecific strain produced a population with more males compared to the mating of the pure strains with each other. In Table 1, the mating of the hybrids produced 93 and 71% male proportion, while the crossing of the pure strain produced 37 and 44%. Eknath and Hulata (2009)**;** El-Zaeem et al., (2012) opined that a pure strain of Nile tilapia crossed with Blue tilapia produces a population that is skewed towards the male gender which is in agreement with the results of this study. Similarly, the time at which swim-up appears in the hatching hapa, which is an indication that fries have been produced for the pure strain, is shorter compared to the hybrids. In Table 2, after the three trials, the average length of time for spawning to take place is 15.97 and 16.13 days, respectfully, for the pure strains, while it takes the hybrid 19.21 and 19.60 days, respectively. This assertion was in line with the work of Nwachi et al. (2020) on the effect of colour on the diallel mating of wild tilapia to UPM red tilapia.

Similarly, the hybrid gave a better growth rate throughout the study, as shown in Table 3. The mean growth rate of the hybrid was higher compared to the pure strain despite lower total feed intake, and this was attributed to the genotype of the hybrid. Lugert et al. (2019); De Verdal et al. (2018) were of the opinion that hybrids performed better than their parents, even with a lower feed intake. The mean daily growth rate of 43 and 41g is higher than the 36 and 31g recorded in their parents.

Conclusively, a male skewed population was produced through the interspecific crossing of two related strains of tilapia. The hybrids and their reciprocals produced a higher number of male fish compared to the females. Similarly, better performance was recorded by the hybrids when compared with their pure strains despite the lower feed intake. This means that the production of the hybrid for commercial purposes will give more income.

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