

Sex Reversal of *Sarotherodon melanotheron* and Evaluation of Growth Performance in Fresh Water Cultured System

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Abstract: A total of 2400 *Sarotherodon melanotheron* fry were reared in six (6) plastic (High density polyethylene) tanks for three months. Each tank was stocked with 400 fry of which three tanks T1₁, T1₂ and T1₃, served as treatment tanks while the other three tanks T0₁, T0₂ and T0₃, served as control. The treatment tanks were administered with 17 α -methyl testosterone hormone, which functioned as a sex reversal hormone for 30 days and further reared for 60 days in which physico-chemical parameters were taken and growth performance was evaluated. The results show that temperature in the broodstock ponds was 29.5°C \pm 1.2°C, Dissolve Oxygen (DO) of 6.222mg/L \pm 1.502 mg/L and hydrogen ion concentration (pH) value of 9.8 \pm 0.476. This did not alter spawning ability in fresh water. Water parameters for control were 29.5 \pm 1.2°C, 8.4 mg/L and 6.8 \pm 0.3 for Temperature, DO and pH respectively while treatment tank has temperature of 27.8 \pm 0.7°C, DO of 8.46 \pm 0.05 mg/L and pH values of 6.53 \pm 0.28. Sex reversal was however successful as spawning did not occur after treatment. Growth parameters were better in treatment tanks compared to control tanks, as growth was doubled in treatment tanks compared to control tanks, treated fish however has the better Mean Weight Gain (MWG), Specific Growth Rate (SGR) and also less but better Feed Conversion Ratio (FCR) with a value of 10.86 \pm 0.18g, 17.58 \pm 1.55g and 15.04 \pm 0.26g respectively. While control has lower MWG, SGR and higher but worst FCR value of 5.56 \pm 0.20g, 10.31 \pm 1.79g and 29.44 \pm 1.10g. The results showed that *S. melanotheron* treated with 17 α -methyl testosterone hormone achieved more growth in the same rearing time with the same quantity of feed. This result promotes the use of Methyl-testosterone in fish farming of *S. melanotheron* (brackish water species) as more weight, length and hence growth is achieved in a lesser time with less feed consumption.

Key words: Sex Reversal % Growth Performance % Cultured System % *Sarotherodon melanotheron*

INTRODUCTION

The ever increasing demand for fish protein in Nigeria has motivated speedy development of aquaculture sector. The importance of fish farming becomes obvious when viewed against the background of fish demand and supply in Nigeria; According to Ayoola [1] in 2008 total domestic fish production were 579,500 tonnes and fish demand currently estimated at 1.80 million metric tonnes and the most cultured fish species remains *C. gariepinus*. Hence the need to introduce other culturable and profitable fish is important to meet the increasing demand for fish protein.

Tilapias are fish originating from Africa and the Middle East. The main species cultured in ponds, cages and pens is the Nile tilapia. The problem with these fish is their quick maturation at two to three months of age and their ability to start breeding at a rate of once a month [2]. Members of the genus *Tilapia* (Family *Cichlidae*)

have been an important source of food for man in tropical and sub-tropical Africa and have entrenched as one of the world's most important fish by the end of the twentieth century, but with greatly increased emphasis on fish culture in this century plus the advent of modern facilities and transportation, *Tilapia* became even more valuable to man. The *Tilapia* is a group of fish species that exhibits parental care. In general, there are about 800 species, most native to West Africa and Asia.

They inhabit brackish water. It occurs both in open and closed lagoon. *Sarotherodon* species are among the commercially important inland fishes of Africa and investigations on several aspects of the biology of the various species have been confined mainly to East Africa. In pond culture, *Tilapia* species are known to exhibit stunted growth as a result of their rapid reproduction rate as female utilize most of their energy in egg production and in mouth brooding [4].

These characteristics result in the overpopulation of stocked tilapia ponds and the stunting of growth being cause by the crowding of the fish. Another problem associated with a mix of males and females is the sizes of the fish for harvest varying from small to large due to the faster growth of males. This makes it more difficult to establish uniformity of product. For producers wanting high yields of large-sized fish in three to four months, all male fry are preferred [5].

Production of all male tilapia can be accomplished by such techniques as separating the males and females manually, hybridization which mates two species to produce all-male offspring, or by artificial sex reversal [6]. The most efficient and least expensive method is sex reversal with the use of methyl testosterone to artificially create sex reversal, the physical sex direction of the fish is manipulated by the feeding of Methyl-Testosterone prior to and during the early sexless stage of the fry. This technique was first developed in Japan in the 1950s for sex reversal of aquarium fish and species of carp. It was demonstrated as commercially feasible in the 1970s. Fish raised in this manner grow bigger quicker because they do not need to expend energy in developing reproductive organs, production of eggs and parental care. If the hormone is properly applied, the sex reversal treatment can be 98 to 100% effective [2].

Use of the hormone Methyl-Testosterone (MT) to induce sex reversal in farmed tilapias has become a common practice in many parts of the world. MT is a simple and reliable way to produce all-male tilapia stocks, which consistently grow to a larger/more uniform size than mixed sex or all-female stocks [4]. Thus, MT usage in tilapia farming is expected to continue to increase rapidly as the global demand for large whole tilapia and tilapia fillets grows. Currently, tilapia is farmed in at least 85 countries, making it the most widely farmed finfish worldwide and second in volume only to carps [7].

MT reduces stunting but increases growth and can be effectively applied in fresh water culture system for production of male only tilapia which reduces reproduction and over population in culture tanks. This study is to determine the effectiveness of sex reversal of *Sarotherodon melanotheron* (black jaw tilapia) in fresh water cultured system using androgen hormone (Methyl-testosterone) and evaluate growth performance of cultured *Sarotherodon melanotheron* (black jaw tilapia) when fed with artificial feed.

MATERIALS AND METHOD

Eighty broodstocks of *Sarotherodon melanotheron* were collected from a floating cage of Department of Marine Sciences Mariculture site in the Lagos lagoon when salinity was about 20ppt. The fishes were transported in an open 25L white plastic container to the Marine Research Laboratory of the University of Lagos in a water of about 20ppt salinity.

The broodstocks were transferred using a scoop net and acclimatized for 14 days in a concrete pond with a dimension of 1 X 1 X 5m. Physico-chemical parameters were taken weekly.

The water was changed once in two weeks to allow production of natural food (production of plankton and zooplankton) in the pond (*scenedesmos spp*, *microcystis spp*, *chroococcus turgidus*, *euglena spp*, *paramecium spp* and mosquito larva) which was supplemented with artificial feed, the water was left to be turbid in order to allow the environment suitable for breeding. Spawning however occur after 41 days.

The fry were immediately collected with a scoop net and transferred into 6 plastic tanks of dimension 1 X 1 X 1.2m. Were they were acclimatized for 3 days before the experiment. The project lasted for 90 days, during which the fry are acclimatized, the sex reversed, growth performance is monitored and physico-chemical parameters were taken.

Experimental Set up: Six plastic tanks each with a dimension of 1 X 1 X 1.2m were used for the experiment. Each of the tanks were disinfected and filled with dechlorinated tap water were stocked with 400 fry. Water was filled to 2/3 of the volume of each tank. The mean weight gain of the specimen in each of the experiment tanks were obtained at the end of every week.

Preparation of Feed with Hormone and Feeding Regime: Methyl testosterone 60mg was dissolved in ethanol and diluted with water until there is no precipitate, the solution was mixed intensively with one kg of coppers (0.5mm-0.8mm) with 56 % protein, The mixture is then dried at a temperature of 60-80°C, The fry were fed at least three times daily from the third day after hatching with 5 percent body weight to 30days [8, 9]. The tanks labeled T₀₁, T₀₂ and T₀₃ were not treated with the hormone while tanks T₁₁, T₁₂ and were treated with the hormone.

Parameters Measurement: The mean standard weight of the fish in each tank was determined at the beginning of the experiment and at every 1 week. The weight of all the fish in each tank was measured using a sensitive scale and mean value was calculated.

DO was measured using appropriate digital instruments (Horiba U-10), pH was measured using a pH meter and temperature was measured using a mercury in glass thermometer.

Growth and Nutrient Utilization Parameters: The following indices were used to determine the biological evaluation of growth performance and nutrient utilization of the experimental fish.

Mean Weight Gain: The weight gain per week was calculated using the formula below.

$$\text{Final weight (g)} - \text{initial weight (g)}$$

Percentage Mean Weight Gain: This was calculated using the formula below.

$$\frac{\text{Mean weight gain (g)}}{\text{Initial mean weight (g)}} \times 100$$

Percentages Mean Weight Gain per Week: This was calculated using the formula below.

$$\frac{\text{Mean weight gain per week (g)}}{\text{Initial mean weight (g)}} \times 100$$

Specific Growth Rate (SGR) is the percentage rate of change in the logarithmic body weight. The SGR was calculated using the formula below.

$$\text{SGR} = \frac{\text{Log}_e W_f - \text{Log}_e W_i \times 100}{\text{Times (in days)}}$$

Where W_f = Final body weight and W_i = initial body weight

Feed Conversion Ratio (FCR): Is the amount of unit weight of food that the fish were able to convert into unit muscle.

$$\text{FCR} = \frac{\text{Feed intake (g)}}{\text{Total weight gain (g)}}$$

Daily Rate of Feeding (DRF) is determined by subtracting the remaining feed after feeding from the initial weight of the feed.

Weekly Rate of Feeding: Is calculated by multiplying DRF by seven.

Gross Efficiency of Food Conversion (GEFC)

$$\text{GEFC} = \frac{\text{Daily rate of growth}}{\text{Daily rate of feeding}}$$

Protein Efficiency Ratio (PER) Is calculated from the relationship between the increment in the weight of (i.e. weight gain) of fish and protein consumed.

$$\text{PER} = \frac{\text{Mean weight gain (g)}}{\text{Protein intake}}$$

Statistical Analysis: The data obtained were statistically evaluated using T test. Data obtained were subjected to one way analysis of variance. Data are represented as mean \pm standard deviation. While the means was compared for significant differences using Duncan's multiple range test using Statistical Package for the Social Sciences (SPSS) 10 packet programs.

RESULTS

Viability of Culturing *Sarotherodon Melanotheron* (Black Jaw Tilapia) in Fresh Water: The experimental fish were successfully reared in fresh water system throughout the course of the experiment.

According to the result Temperature in the broodstock pond ranged from 28°C to 31°C having an average value of 29.5°C \pm 1.2°C, dissolved oxygen values ranged from 5.10mg/l to 8.40mg/l with an average value of 6.222 \pm 1.502mg/L, Hydrogen ion concentration (pH) values ranged from 9.3 to 10.3 with an average value of 9.8 \pm 0.476.

Sarotherodon melanotheron (black jaw tilapia) was successfully cultured in fresh water system. The physico-chemical parameters of fresh water did not alter the reproductive ability of the broodstocks and the fries were successfully cultured in fresh water system.

Sex Reversal and Growth Performance: At the end of 90 days the sex of all the female in the tanks T1₁, T1₂ and T1₃ Which were fed with imported feed treated with hormone (Methyl-Testosterone) were all reversed to male this was also obvious in the fast growth of the fish in tanks treated with hormone when compared to growth rate of untreated fries, initial and final weight of fry is shown in Table 1 below,

Growth and nutrient utilization parameters of the fish in different treatments are presented in Table 2.

Table 1: Mean weight, percentage mean weight gain per week and percentage mean weight gained in different tanks (90days)

Tanks	Initial mean weight (g)	Final mean weight (g)	Mean weight gained (g)	% mean weight gained (g)	% mean weight gain per week (g)
T0 ₁	0.62	6.41	5.79	933.90	77.83
T0 ₂	0.88	6.26	5.38	611.43	50.95
T0 ₃	1.32	6.83	5.51	417.42	34.85
T1 ₁	0.56	11.56	11.00	1964.35	163.70
T1 ₂	0.90	11.55	10.65	1183.33	98.61
T1 ₃	0.62	11.57	10.95	1766.13	147.18

Table 2: Growth indices and nutrient parameters of the studied fish in different tanks

Parameter	T0 ₁	T0 ₂	T0 ₃	T1 ₁	T1 ₂	T1 ₃
MWG	5.79	5.38	5.51	11.00	10.65	10.95
%MWG	933.90	611.43	417.42	1964.35	1183.33	1766.13
%MWG/WK	77.83	50.95	34.85	163.70	98.61	147.18
SGR	8.56	12.17	10.19	18.78	15.83	18.15
FCR	28.24	30.41	29.67	14.85	15.35	14.93
DRF(g)	10.895	10.895	10.895	10.895	10.895	10.895
WRF(g)	76.895	76.895	76.895	76.895	76.895	76.895
GEFC	0.0354	0.0328	0.0337	0.0672	0.0651	0.0669
PER	0.1033	0.0960	0.0983	0.1964	0.1901	0.1955

Where: T0₁, T0₂, T0₃ being the control tanks and T1₁, T1₂, T1₃ being treatment tanks

DISCUSSION

The recorded water parameters did not alter spawning in the tilapia as spawning occurred in about 14 days of stocking, this recorded water quality parameters is in agreement with the work of [10, 11]. The physico-chemical parameters was also monitored in the experimental tanks as temperature remain uniform in all studied tanks, while dissolved oxygen and hydrogen ion concentration (pH) value varied in different tanks for culturing of *Sarotherodon melanotheron* and this is in accordance to [9, 12] findings, who stated that DO should be above 3mg/L, pH of 6.0- 9.5, temperature (28±1°C),

At the end of 90 days the sex of all the female in the experimental tanks which were fed with feed treated with Androgen hormone (Methyl-Testosterone) were all reversed to male this was obvious in the fast growth of the fish and inability to reproduce in the experimental tanks, with male population of 99.33±1.15% in treatment tank while the control has a population of 46.66±7.63% male this agreed with the findings of [4] which recorded about 95% success with the use of Methyl-Testosterone, [13, 14] both recorded 98% - 100%, [15] obtained 97%, [16] obtained 98% males at the dose rate of 60 mg kgG¹ MT, [17], also obtained 71.9% males at the dose rate of 120 mg kgG¹ MT of feed.

The feeding trial revealed that *Sarotherodon melanotheron* responded to imported feed (coppens 0.5mm – 0.8mm) with crude protein of 56 percent, *Sarotherodon melanotheron* was able to utilize imported feed for growth which was also successfully used to rear *Sarotherodon melanotheron* by [11]. The rate of feeding was the same in each tank and a daily feed of 5 percent body weight was given three times to each tank daily. Growth parameters were better in treated tanks compared to control tanks, as growth was doubled in treated tanks compared to control tanks. The higher mean weight was obtained in treated tanks as compared to the control tanks where lower mean weight gain was recorded, Specific growth rate (SGR) was better in treated tanks and control tanks having the lower SGR, FCR value was lower and better in treated tanks while control tanks have a higher and worse FCR. The significant difference in growth rate can be ascribed to the inclusion of 17 alpha-methyl testosterone in the feed of the fish in treatment tanks, Compared to the control tanks which were also fed with imported feed but not treated with 17 alpha-methyl testosterone this result is in line with [9] which recorded a maximum gain in body weight i.e., 11.8 g, of *Sarotherodon melanotheron* fed with 17 alpha-methyl testosterone in 90 days. Hanson *et al.* [18] reported that 10-60 ppm MT-treatment showed a better growth than control, these are also in accordance with Dan and

Little [19], who compared the culture performance of different strains of *Sarotherodon melanotheron* and found that considering all strains, MT treatment resulted in a final size of fish 10.7% larger than mixed sex fish. At the end of 90 days the fries treated with methyl-testosterone shows better growth than fries that are not treated with methyl-testosterone, sex of fries treated with MT were completely reversed from female to male this result helped in cutting the prolific spawning ability of Tilapia which normally results in stunted growth and over population.

This result will definitely promote the use of Methyl-testosterone in farming of *Sarotherodon melanotheron* (brackish water specie) as more weight, length and hence growth is achieved in a lesser time compared to ordinary feed. Grow out period of *Sarotherodon melanotheron* is reduced, less feed is consumed thereby increasing income for tilapia producers.

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