

Survivality of Mono-Sex Tilapia (*Oreochromis niloticus*) Fry Using 17- α Methyltestosterone in a Commercial Hatchery of Chittagong, Bangladesh

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Abstract: The research was undertaken to develop the sex reversal technique of tilapia by using 17- α methyltestosterone. Locally available materials were used to prepare some hatchery equipments. Fry were treated with four different hormone doses of 15, 30, 60 and 120 mg kg⁻¹ of feed in hapa I-IV, respectively in the ponds. The stocking density of fry was maintained 3000 m⁻² in the experimental hapas. Physico-chemical parameters varied from as temperature 26.5-29°C, pH 6-8, DO 4.5-7.5 mg L⁻¹, water transparency 16-35 cm, alkalinity 48-78 mg L⁻¹, hardness 50.7-155 mg L⁻¹. The sex reversal efficiency was found 88.33, 90.83, 96 and 78.33% in hapa I-IV, respectively. The highest sex reversal efficiency was obtained in hapa III where fry were treated with 60 mg 17- α methyltestosterone kg⁻¹ feed. The survival rate (%) were recorded 92.45, 95.41, 93.75 and 89% in hapa I-IV, respectively. The highest survival rate (95.41%) was found in hapa II.

Key words: Sex reversal, 17- α methyltestosterone, hapa, feed, stocking density, survival rate

INTRODUCTION

Bangladesh is uniquely rich and diverse in water resources. It has innumerable water bodies including ponds, lakes, rivers, haors, baors, beels, tanks, estuaries and inundated paddy fields. Due to favorable climatic condition, the water bodies of Bangladesh are highly productive and aquaculture is an important commercially viable activity. The fisheries sub-sector in Bangladesh contributes significantly to nutrition, employment, household income and foreign exchange earnings. Fish provides 63% of the animal protein intake in Bangladesh and the annual per capita fish consumption is 14 kg. About 1.2 million people are engaged full-time and 12 million people part-time in the aquatic production sector. Aquatic products are the country's 2nd largest export commodity contributing 10% of annual export earning, 5.2% of national GDP and 20% of the agriculture GDP (Shah, 2003).

One of the most important herbivorous fish species reared in aquaculture systems is the tilapia. Tilapia is more easily grown than other fowl fish species for either commercial or non-profit enterprises. Tilapia are naturally accustomed to eating plant ingredient and typically

considered strict herbivores, once they reach maturity (Arrignon, 1998). By production volume, tilapia (*Oreochromis niloticus*) culture is one of the largest freshwater aquaculture species worldwide and is mostly produced using semi-intensive systems in developing countries (Thomas and Michael, 1999). During the last decades, Nile tilapia, endemic to Africa became an important culture species in many Asian countries including China, Indonesia, Bangladesh, Malaysia, the Philippines, Thailand, Vietnam, Myanmar and Sri Lanka. UNICEF introduced the current stock of Nile tilapia into Bangladesh in 1974. In 1987, the Bangladesh Fisheries Research Institute (BFRI) introduced another stock of Nile tilapia from Thailand (Gupta *et al.*, 1992). Nile tilapia is a preferred culture species because of desirable features like adaptation to a wide range of environments including shallow or seasonal water bodies and ditches, good taste, fast growth, easy reproduction and versatile feeding behavior. It grows well on a vegetarian diet in part due to the possession of a long intestine and a stomach pH <2.0 (Getachew, 1989).

Tilapia have been the subject of intensive investigation since their introduction for aquaculture beginning in the 1950's. The various species of

commercial value have been introduced throughout tropical regions of the world. The widespread utilization of the hormone Methyl-testosterone (MT) to achieve the production of all male fry has been a significant factor in permitting development of Tilapia as an aquaculture commodity on global markets.

The use of hormones to alter the sex ratios of fish was first demonstrated in species other than tilapia. Yamamoto (1951) concluded that sex hormones in addition to modification of secondary sex characteristics also affect the gonads.

Androgen induced masculinization and estrogen resulted in femmization. Production of all male population through administration of androgen (17- α methyl-testosterone) is considered to be the most effective and economically feasible method for obtaining all male tilapia populations (Guerrero and Guerrero, 1988).

Tilapia (*Oreochromis niloticus*) was first introduced in Bangladesh in 1954 which was not succeeded at that time. Later in 1974, UNICEF again introduced *O. niloticus* from Thailand. This attempt for tilapia culture also did not flourish as its biology, behavior and culture technology were not known. In recent years, tilapia culture has become very popular among the fish farmers. There is a great potential for successful tilapia culture in numerous ponds of Bangladesh.

The tilapia (GIFT and Mono-sex) has been recognized in the last few years as a species with high potential for aquaculture. The present research was an attempt to determine the hatching percentage of eggs, survivability of *Oreochromis niloticus* fry and sex reversal efficiency by the application of different doses of 17- α methyltestosterone.

MATERIALS AND METHODS

The present study was carried out at Rawzan Rupali Prawn Hatchery Ltd. Bainna Pukur Par, Rawzan Municipality, Rawzan upazila which is located in the northern part of Chittagong (Fig. 1). Total area of the hatchery is 10 acres consisting of six grow out ponds and brood stock ponds; eight nursery ponds, two indoor brine chambers, an overhead tank, a laboratory, a staff quarter, a hatchery unit for *Macrobachium rosenbergii* and another hatchery unit for Mono sex tilapia fry production. It has a deep tube well for fresh water source.

The tilapia hatchery unit has various facilities for Mono-sex tilapia fry production like a cemented filtration unit and 20 cemented feeding trays. The research study was carried out from December 2009 to April 2010. In this study, one pond was used for broodstock management and four separate ponds were used for nursing the



Fig. 1: Geographical location of the study area (Banglapedia, 2004)

fingerling with hormone treatment. The areas of the broodstock ponds were 79 and 56 decimal where as the nursing ponds were 40, 45, 62 and 53 decimal, respectively. Water filling and fertilization was done in order to encourage a healthy plankton bloom because a healthy plankton bloom shades the pond bottom and prevents the growth of harmful benthic algae, reduces fluctuation in water temperature, provides oxygen utilize nitrogenous and phosphate wastes with in the pond and provides a suitable environment where the tilapia feels comfort.

In case of water intake each pond followed a simple method of water screening through 350 micron mesh screens to avoid the entrance of larvae or juveniles of undesirable species. Water depth was maintained at 15 cm for 4 days and gradually raised up to 30 cm at a rate of 5 cm day⁻¹. The blooms were judged by watercolor and turbidity. In the following week, water level was gradually increased up to 1.2 m. Organic fertilizers (dried cow dung and poultry liter at wet condition) were applied in the ponds at a rate of 3 ton/ha/per month. Inorganic fertilizers (TSP and Urea) were applied at a rate of 100 kg/ha/per month. After raising the soil pH at a desired level, the ponds were treated by lime (Ca MgCO₃) at a rate of 125 kg ha⁻¹ to stabilize the soil pH of the ponds as well as to support the fertility of the ponds. Two sand gravel

filters were used for water filtration. The rectangular filter was made up of cement. The dimensions of the unit were 1 m in width, 10 m in length and 1 m in height. It consisted of two chambers-water filtration chamber and water storage chamber. Low cost hatching jar and hatching tray were prepared for the fry hatching management in the hatchery. Fine mesh net (1.6 mm) was used to make the hapa in which the four top and bottom corners were tied to bamboo stakes. The seams were sewn typically at a rectangular in shape with a nylon thread and double stitched to prevent splitting. Hapas were then installed in broodstock ponds and nursery ponds in such a way that the open part of the hapas was two feet above the water surface. Four bamboo poles were used for the placement of each hapa. Hapas were designed to allow the fish to be crowded to one end for collection so that brooders could be examined for egg and any diseased brood, sac fry, swim-up fry could be removed easily.

The area of the brood stock hapa was 32 m² (8×4 m) and the area of the nursery hapa was 2 m² (2×1 m). The height of the both types hapa was 1 m. Soil samples were collected from the pond bottom and dikes at a depth of 0-10 cm. Five representative samples were collected from four corners and middle of the ponds. pH was determined by a soil pH meter. All the experimental ponds were sun-dried until the soil cracked. After drying, ponds were tilled entirely to a depth of 5-10 cm for two times. Followed by tilling, reconditioning of pond bottom trench, leveling, pond repair, etc., were done. As the soil pH of the ponds were found to be slightly acidic (5.1-6.2), agricultural lime (CaCO₃) was applied to the ponds at a rate of 1,200 kg ha⁻¹. Lime was first crushed into dust and broadcasted all over the ponds bottom and dikes. Adult GIFT tilapia weighting, 100-150 g were collected from brood stock pond of the hatchery at January 28, 2010. After collection, male and female were sorted out by manual sexing. The broods were treated with 50 ppm formalin for 10 min. The selected male and female broods were kept separately in two separate hapas which were setup in broodstock pond. Stocking density of broods in the hapa was 4 indivs m⁻².

Collection, sorting and stocking of broods were completed within 9:00 a.m. to avoid temperature stress. After rearing of 20 days the broods were transferred to new hapa for mating at the ratio of 1:4 (M: F). Supplementary feeds were used for proper nutrition. The feeding rate was 3% of total body weight. The total amount of feed was divided into two equal rations and fed in the early morning and in late evening (at 7:00 and 18:00 h). Dough type of feeds were administered on feeding tray made of polyethylene sheet fixed on bamboo frame. To maintain sufficient natural feeds in the pond

fertilizers were applied at a reduced rate (urea 35 kg ha⁻¹ and TSP 20 kg ha⁻¹) at 15 days interval. Sometimes organic manure was applied at the rate of 2 ton ha⁻¹. Water transparency (secchi depth) was checked regularly and maintained in between 20-35 cm to ensure the existence of sufficient natural feed.

Females were checked with a minimum disturbance to determine which one was holding eggs. The eggs were rinsed from the mouth of female and collected eggs were taken in a plastic bowl. Then the females were returned to the hapa for next spawn. At that time, broods with poor condition were removed. The prepared jars were setup on the filtration chamber by using wooden frame to ensure a continuous moving base aeration and water flow. Eggs were then put into the incubation jar at the rate of 250 g per jar. Underground water was first stored in the overhead tank.

Then a constant and regulated downward water flow was provided on the incubation jar by pipe line. The water flow on incubation jar was maintained at the rate of 8 L min⁻¹. So, the eggs were rolled vigorously in the round bottomed incubators. When the eggs were turned from light-yellow to dark orange in color (just ready to hatch), these were transferred into the hand made hatching tray. In hatching tray, the eggs were kept at suspended and movable condition by a continuous horizontal flow of water through pipe line. Here, the flow of water was maintained at 4 L min⁻¹ which was well enough to keep the eggs movable and stress free. In order to inspect any disorder, standby monitoring was carrying on and dead egg cells; dead sac fry which were attached along the net of tray were removed by using brush. Within 24 h, the yolk-sac fry were hatched out. The eggs which were not responded were removed from the tray. The swim-up fry were kept here at same condition without feeding until they absorbed their yolk-sac.

After yolk-sac absorption, hormone mixed feed was fed to newly hatched swimup fry for 21 days for sex reversal. 17- α Methyltestosterone (MT) hormone was purchased from the local distributor (Allwells Marketing Ltd. Cox's Bazar) of ARGENT Chemical Laboratories, USA. Four different (15, 30, 60 and 120 mg) concentrated dose of stock solution was prepared to apply in different hapa. A highly palatable feed was needed to obtain an active feed response and effective sex reversal. A commercial Nursery-I feed (<40% protein) was collected from NIRIBILI Feed Co. Ltd. The feed were ground and passed through a 0.6 mm mesh screen to get a powder like feed. Then already prepared stock solution was sprayed over the feed at the rate of 15, 30, 60 and 120 mg hormone concentrated solution per kg feed and mixed properly. After their absorption of yolk-sac fry were collected in a

bowl from the tray through siphoning by using plastic tube. Prior to stocking in hapa fries were counted by visual estimation. According to the method, the fry were first counted individually into a selected bucket adding enough fry with water that gave uniform distribution throughout the container. Thus, a standard was prepared using 1000 fry per bucket.

Then by enumerating through visual distribution, the buckets of same color and size were filled with 5-10 cm water and fry were added until the fish density in all the buckets appeared the same as the standard one. During visual estimation, healthy fry were counted in the container and replaced them if they became stressed. At the same time, unwanted materials (if any) were removed carefully. Then the fry were stocked in four nursery hapas which were installed in four separate ponds. In each hapa (2×1×1 m), 4000 fries were stocked maintaining the stocking density of 2000 indvs m⁻².

In the total duration of 21 days of hormone treatment, hormone mixed feed was given 20% of their body weight/day for the 1st week, 15% of their body weight/day for the 2nd week, 10% of their body weight/day for the 3rd week. The fries were fed 3 times day⁻¹ for best growth. In order to inspect any disorder or to prevent sudden fluctuation during the nursing period some major water quality parameters of the ponds were recorded with great care and accuracy. If any abnormalities were found then necessary steps were undertaken. The observed parameters were: water temperature, water pH, dissolved oxygen, alkalinity, hardness and secchi depth.

Water temperature was measured by using a mercury thermometer, water pH was recorded by pH meter, secchi disk visibility of water was measured by lowering a secchi disk of 30 cm diameter into the water. The DO of pond water was determined by following Winkler's method (Barnes, 1959). The alkalinity of pond water was determined by following standard method (Barnes, 1959). Hardness of the experimental pond was analyzed by the standard method (APHA, 1995).

After the hormone, treatment efficacy was evaluated through a detailed examination of the gonads of the representative sample of fish. Fry were collected at the end of the hormone treatment period by crowding the fish together and collecting a random sample. A representative sample of ≥100 fish was selected for gonadal examination and preserved in 10% formalin. For gonadal examination, dissecting equipment was needed along with a microscope slide and a stain (hemotoxin). Fry were preserved a minimum of 10 days in formalin before gonadal examination. Because the gonadal tissue of fish preserved for <10 days would remain elastic and often

break when being removed. Fry were dissected by making a cut near the anus to below the base of the pectoral fin. The entire gonad, located on the dorsal portion of the peritoneal lining was removed carefully beginning ventrally and going forward. Thick gonads were needed to be sliced longitudinally before they were examined properly. For efficient use of supplies, about 4-5 sets of gonads were placed on a microscope slide and each given a drop of dye. Another slide was placed on top and the gonads were gently rolled or squashed. And finally, the gonads were determined microscopically on the basis of their gonadal tissue.

RESULTS AND DISCUSSION

Total 100 males and 400 females weighing 150-200 g were selected. They were conditioned for 20 days for better spawning performance. After conditioning, they were transferred to four broodstock hapas at a ratio of 1:4. After 15 days, eggs were collected from hapas which were found to be in good condition. The fishes were highly fecund. Around 300 eggs were collected from each female. In this present study, four low-cost hatching jars were used with a continuous downward water flow. In each jar, 25000 eggs were given. During incubation, light yellow colored eggs were turned in to dark orange eggs. Then the eggs were transferred to hatching tray. Yolk-sac fry were hatched out within 24 h. They were kept in tray until their yolk sac was totally absorbed. The hatching rates of fertilized eggs were 76% in tray 1, 84% in tray 2, 82% in tray 3 and 72% in tray 4 (Fig. 2). The highest hatching rate was 84% in tray 2.

After 2 days when the yolk-sac was absorbed, the survival rates of fry were 94.38% in tray 1, 90.55% in tray 2, 95.12% in tray 3 and 92.47% in tray 4 (Fig. 3). After yolk-sac absorption swim up fry were stocked in four hapas installed in four separate ponds. In each hapa, 6000 fry were stocked and treated with different doses of hormone. The fry were fed 15, 30, 60 and 120 mg kg⁻¹ feed in hapa I-IV, respectively (Table 1-2).

In the present study, water quality parameters of the nursing hapas such as temperature, pH, dissolved oxygen, alkalinity and hardness secchi depth were measured every three days interval. During the study

Table 1: Amount of 17- α methyltestosterone application in experimental hapas

Hapa	Applied hormone dose (mg kg ⁻¹ feed)
I	15
II	30
III	60
IV	120

Table 2 : Growth of fry in different hapa with different dose of 17- α methyltestosterone mixed feed

Sampling periods	Hapa I (15 mg kg ⁻¹ dose of hormone)		Hapa II (30 mg kg ⁻¹ dose of hormone)		Hapa III (60 mg kg ⁻¹ dose of hormone)		Hapa IV (120 mg kg ⁻¹ dose of hormone)	
	Fry length (mm)	Weight (g) (1000 indivs.)	Fry length (mm)	Weight (g) (1000 indivs.)	Fry length (mm)	Weight (g) (1000 indivs.)	Fry length (mm)	Weight (g) (1000 indivs.)
03.03.10	10.8	26.5	10.6	25.5	10.5	27.7	12.6	25.8
06.03.10	11.9	35.7	11.7	32.6	12.5	35.0	13.0	35.2
09.03.10	12.8	41.9	12.5	42.5	13.1	45.2	13.4	41.3
12.03.10	14.3	59.5	14.1	60.3	15.9	58.5	15.5	61.6
15.03.10	16.7	75.4	17.5	75.1	17.3	76.8	17.3	79.3
18.03.10	19.4	94.5	20.7	95.8	29.2	90.3	21.8	94.5
21.03.10	22.5	124.3	23.3	132.2	22.9	128.0	23.2	134.2
24.03.10	25.4	136.5	25.5	139.0	24.5	134.0	26.0	137.0

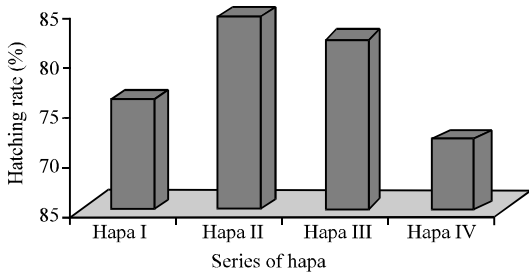


Fig. 2: Hatching rates of eggs in different hapas

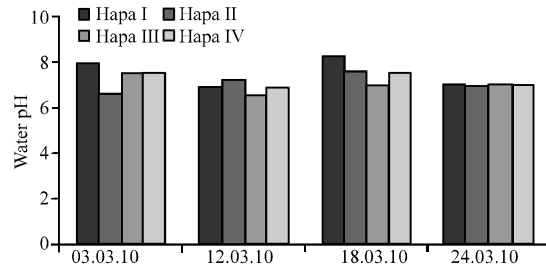


Fig. 5: Water pH in different hapa

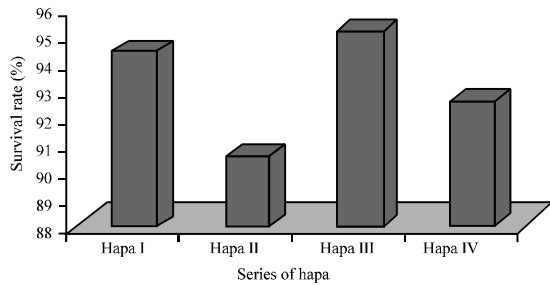


Fig. 3 : Survival rates (%) of fries in different hapas

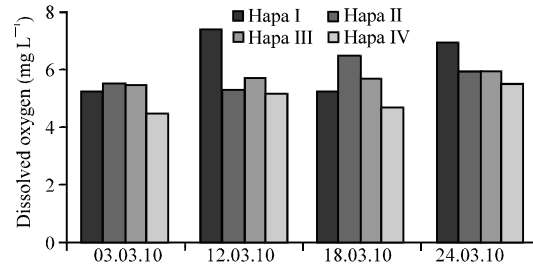


Fig. 6: Dissolved oxygen in different hapas

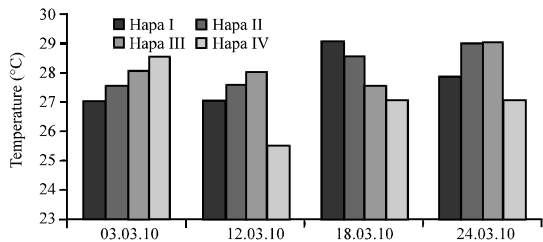


Fig. 4: Water temperature in different hapas

period, surface water temperature varied from 27-29, 27.5- 29, 27-29 and 27-28.6°C in hapa I-IV, respectively. The mean values of water temperature were detected as 27.8±0.607, 28.25±0.53, 27.91±0.76 and 27.45±0.3181°C in hapa I-IV, respectively. So, the average water temperature was maximum in hapa II and minimum in hapa IV (Fig. 4). The water pH was recorded from 6-8.5, 6.5-8.0, 6.5-7.5 and 6.4-7.68 in hapa I-IV, respectively. The mean values of

water pH were 7.8±0.60, 7.21±0.22, 7.1±0.070 and 7.2±0.141 in hapa I-IV, respectively. So, the average value of pH was maximum in hapa I and minimum in hapa III (Fig. 5). Dissolved oxygen varied from 5.3-7.4, 5.3-6.5, 4.5-5.9 and 4.5-5.8 mg L⁻¹ in hapa I-IV, respectively (Fig. 6). The mean values of DO were 6.33±0.78, 5.73±0.191, 5.37±0.371 and 5.15±0.247mg L⁻¹ in hapa I-IV, respectively (Fig. 7). So, the average DO content of water little bit higher in hapa II. In the present study, range of water transparency were varied from 22-29, 24-29, 16-25 and 23-35 cm in hapa I-IV, respectively. The mean values of water transparency were 25.8±2.02, 26.25±0.88, 20.75±1.59 and 27.75±3.001 cm in hapa I-IV, respectively. So, the average water transparency was maximum in hapa IV and minimum in hapa III. The ranges of water alkalinity were varied from 48-78, 56-76, 48-72 and 55-68 mg L⁻¹ in hapa I-IV, respectively. The mean values of alkalinity were 66.57±9.38, 62.87±3.62, 60.37±3.27 and 61.0±4.50 mg L⁻¹ in hapa I-IV, respectively. So, the average alkalinity

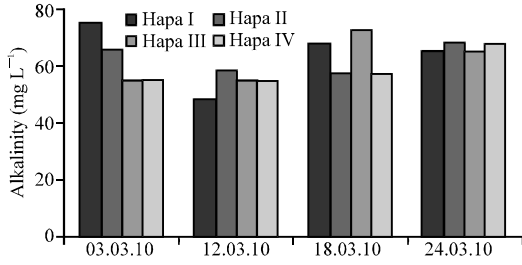


Fig. 7: Alkalinity in different hapas

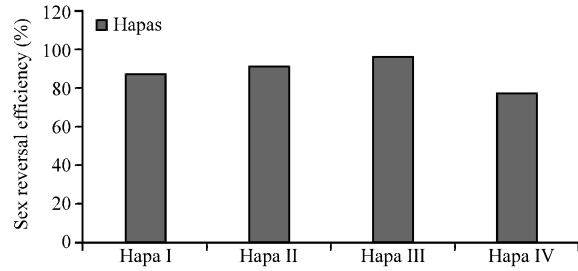


Fig. 10: Sex reversal efficiency of tilapia fry

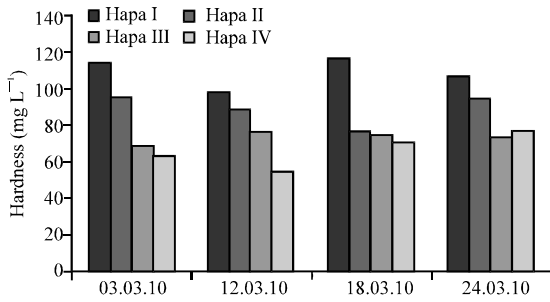


Fig. 8: Hardness of water different hapas

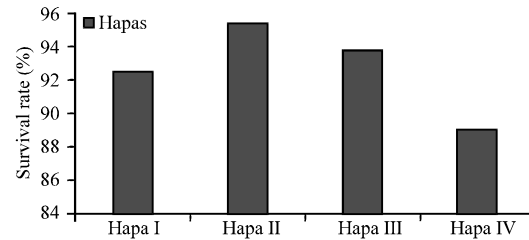


Fig. 11: Survival rate in response to hormone in feed

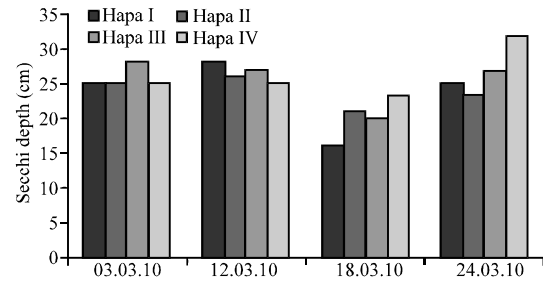


Fig. 9: Secchi depth of water in four hapas ponds

was maximum in hapa I and minimum in hapa III. The range of water hardness was varied from 108-155, 80-125, 50.7-85.5 and 55-78 mg L⁻¹ in hapa I-IV, respectively. The mean values of hardness were 118.5±16.20, 93.48±1.06, 72.58±0.99 and 67.37±7.51 mg L⁻¹ in hapa I-IV, respectively. So, the average hardness was maximum in hapa I and minimum in hapa IV (Fig. 8).

The sex reversal efficiency was found 88.33, 90.83, 96 and 78.33% in hapa I-IV, respectively. The maximum sex reversal efficiency was in hapa III where fry were treated with 60 mg kg⁻¹ feed (Fig. 9 and 10). The survival rates were recorded 92.45, 95.41, 93.75, 89% in hapa I-IV, respectively.

The highest survival rate was found in hapa II. Fry were fed with hormone treated feed at 20% of body weight during 1st week, 15% of during 2nd week and 10% of during 3rd week (Fig. 11). Fry were sampled at 3 days interval to determine body length and weight (Table 2).

The significant positive correlations ($p < 0.05$) of water quality of the hapa based experimental ponds with length and weight of juvenile *Oreochromis niloticus* have been observed (for hapa-1, $r_{\text{water temperature} \times \text{fy length}} = 0.48914$, $r_{\text{water temperature} \times \text{fy weight}} = 0.47314$, $r_{\text{water pH} \times \text{fy length}} = 0.064$, $r_{\text{water pH} \times \text{fy weight}} = 0.0215$, $r_{\text{alkalinity} \times \text{fy length}} = 0.005$, $r_{\text{transparency} \times \text{fy length}} = 0.159$ and $r_{\text{transparency} \times \text{fy weight}} = 0.160$) stand as proof of interchange or exchange process between the water quality and *Oreochromis niloticus* (Table 3).

During the study period, surface water temperature varied from 27-29, 27.5-29, 27-29 and 27-28.6°C in hapa I-IV, respectively. So, the average water temperature was maximum in hapa II and minimum in hapa IV. According to Phelps *et al.* (1993) for sex reversal of tilapia optimum temperature is between 26-28°C. Temperatures <24°C significantly reduce growth and may result in some fish not having completed gonadal differentiation during the treatment.

Lower temperatures also favor more disease problems. Water pH varied 6-8.5, 6.5-8.0, 6.5-7.5 and 6.4-7.6 in hapa I-IV, respectively. So, the average value of pH was maximum in hapa I and minimum in hapa III. Dissolved oxygen varied from 5.3-7.4, 5.3-6.5, 4.5-5.9 and 4.5-5.8 mg L⁻¹ in hapa I-IV, respectively.

So, the average DO content of water little bit higher in hapa II. According to Pakrasi and Banerjee, 4-8 mg L⁻¹ of DO is favorable range for pond culture and Singh (1984) considered 5 mg L⁻¹ or more to be desirable range, the level of DO that was recorded in the present investigation was favorable for pond culture as it were

Table 3: Correlation coefficient (r) between water quality and sex reversal efficiency

Combinations	Values (r)			
	Hapa I	Hapa II	Hapa III	Hapa IV
Water temperature x fry length	0.489144	0.705621	0.255337	-0.113040
Water temperature x fry weight	0.473140	0.647831	0.255337	-0.113040
Water pH x fry length	0.064000	-0.192080	-0.405170	-0.395120
Water pH x fry weight	0.021516	0.561620	-0.497640	-0.424540
D.O. x fry length	-0.004100	0.204394	0.570841	0.622629
D.O. x fry weight	-0.006770	0.116167	0.542363	0.978397
Alkalinity x fry length	0.005662	-0.100360	0.826119	0.410586
Alkalinity x fry weight	-0.132690	0.795234	0.790072	0.453920
Hardness x fry length	-0.404090	-0.500520	0.597407	0.714362
Hardness x fry weight	-0.425400	-0.520710	0.769154	0.743996
Transparency x fry length	0.159951	-0.378240	0.769694	0.317475
Transparency x fry weight	0.160554	-0.402730	0.764685	0.296395

4.5-6.1 in three experimental ponds. In the present study, water transparency were varied from 22-29, 24-29, 16-25 and 23-35 cm in hapa I-IV, respectively. So, the average water transparency was maximum in hapa IV and minimum in hapa III.

There was no marked fluctuation of transparency inline present study. It ranged from 15-33 cm which may be compared with the studies of Duenas *et al.* (1983) reported 26-54 cm to be the suitable turbidity range for culture. Cange (1982) also reported the transparency of water ranging from 10-56 cm is suitable for the growth. Larkins (1995) stated that a low alkalinity would allow a greater fluctuation of pH values from a low in the morning to a high value in the afternoon.

He also stated that fluctuation in water pH would stress the larvae by causing fluctuations in the toxic forms of ammonia and hydrogen sulphide. The alkalinity range of 42-96 mg L⁻¹ (CaCO₃) in the ponds that recorded in the present investigation was congenial for coastal pond culture. During study period, the ranges of water alkalinity were varied from 48-78, 56-76, 48-72 and 55-68 mg L⁻¹ in hapa I-IV, respectively. So, the average alkalinity was maximum in hapa I and minimum in hapa III.

The duration of treatment must be adequate to allow all fish to complete gonadal differentiation during the treatment period. In the present study, fry were treated with 60 mg MT kg⁻¹ for 21 days and 87% males were produced. At the end, the fry were 23 mm in average. Bocek *et al.* (1992) produced 98% males feeding 60 mg MT kg⁻¹ for 30 days. At the end of treatment, the fish averaged 14.9 mm.

Phelps *et al.* (1993), feeding MT to *O. niloticus* for 28 days found that the effectiveness of hormone sex reversal was correlated to the number of days fry received hormone before reaching 18 mm. Starting with fish <11 mm that grew rapidly during treatment; fish received 14 days of hormone feeding before reaching 18 mm and were 95.7% males. Starting with 12-13 mm fish under the same

growth conditions, fish were >18 mm in 9 days and were only 87.3% males. In the present experiment, the stocking density was 3,000 m⁻² in out door nursing hapas which in close agreement with the statement of Cruz and Mair (1994). They compared stocking densities of 1,000, 3,000 and 5,000 fry m⁻² of hapa using *O. niloticus* and found best sex reversal at 3,000 and 5,000 m⁻² but lower survival at 5,000 m⁻². Popma and Green (1990) also stocked at densities of 3,000-5,000 m⁻² in hapa. High densities-help insure an active feeding response needed so all fish are consuming feed.

Pandian and Varadaraj (1987) observed that fry can establish a hierarchy in feeding order resulting in small fish not consuming adequate quantities of hormone treated feed for successful sex reversal. Stocking fry directly into earthen ponds has also been effective. Phelps *et al.* (1995) obtained >96% males when *O. niloticus* fry were stocked at 200-260 m⁻² into 215 m⁻² earthen ponds and fed MT treated feed for 28 days. In a 2nd trial, fish were stocked at only 75 m⁻², the percentage of males was 91.3%.

A highly palatable feed with high nutritional value is needed to obtain an active feed response and effective sex reversal. So, high protein, complete in vitamins and minerals with fish oil added to increase nutritional value and palatability in this study. Oral opening of fry is very small so the feed was ground to pass through a 0.6 mm mesh. Hormone was thoroughly mixed with feed to make the feed particles homogeneous.

Okoko (1996) fed *O. niloticus* at 15% body weight/day, diets containing 3.75, 7.5, 15, 30, 60, 120, 240, 480, 600 or 1200 mg MT kg⁻¹. At 3:75 mg he obtained 80.0% males and 19% intersex at 7.5 mg the results were 91.7 % males and 8.3% intersex and at 15 mg obtained 98.3% males and 1.75 inter-sexes. He calculated that daily MT intakes of 0.52-2.85: g g⁻¹ of fish gave >95% male populations. In the present study, fry were reared in hapa installed in earthen pond where natural food was

available. Sex reversal efficiency was high in 30 and 60 mg α MT kg⁻¹ as 87 and 93%. Similar result was reported by Phelps *et al.* (1995) in a clear water environment with no natural food available a diet containing 15-30 mg kg⁻¹ may be effective while in an outdoor setting with natural food available an optimum dose may be between 30-60 mg kg⁻¹ of diet given at 15% body weight or more.

CONCLUSION

The research study demonstrated that good management of Mono-sex tilapia (*Oreochromis niloticus*) depends on environmental parameters. Better production of the Mono-sex tilapia fry and survivability depend on the proper utilize of 17- α MT in hapas. During the study, it was found that different doses of 17- α MT have different effects on the production and survival of Mono-sex tilapia. In the whole study period, all the parameters and feed management were properly maintained.

This study provides a guide line for the proper management system of the brooders and proper utilization of hormone mixed with feed for better performance. To enrich the total aquaculture production, the culture of tilapia should be increased which can be ensured by producing sufficient Mono-sex tilapia fry of good quality. So for mass production of mono-sex tilapia fry good management system for the broodstocks is a must. Further, scientific experiments are to be needed to solve the problem of water quality management.

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