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Preliminary Observations on Effect of Basal Feed and 17-α Methyltestosterone on Producing Monosex Culture of *Oreochromis niloticus*

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ABSTRACT. *Two trials were conducted to study the effect of basal diet (chicken egg yolk or fish meal) with or without a hormone (17-α methyltestosterone) on the production and growth rate of monosex fry of Oreochromis niloticus. The hormone was mixed at the rates of 0.125 or 0.075 mg per 25 g feed. Artificially hatched day old post-larvae were fed with one of the diets at 10% of body weight, thrice a day, for forty days. The amount of feed given was adjusted weekly according to the body weights of the fish fry. On the forty first day, fish were transferred to cemented tanks and fed a normal fish diet which contained 40% crude protein. At the end of the third month, fish were sexed visually. The percentage of males of the surviving fish fed with the hormone added diet was 100%. Growth rates for egg and fish meal were approximately 3.0±0.7 g per day and 2.3±0.6 g per day, respectively. These values were not significantly different (P>0.05). The results suggest that 17-α methyltestosterone could be used to produce an all-male population, but basal diet had no influence on hormonal effect.*

INTRODUCTION

Fresh water aquaculture in a developing country like Sri Lanka offers a promising solution to improve protein deficiency (Galapitage, 1981). Since Nile tilapia fish has desirable characters, such as easiness in domestication, efficient utilization of available feed and high proliferation rate, it is a potential source to increase the output from fresh water aquaculture in Sri Lanka.

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Nile tilapia (*Oreochromis niloticus*) when cultured in ponds, reach sexual maturity while they are still very small in size (Dunham, 1990), leading to overpopulation. This causes stunting due to lack of space, resulting in a significant number of tilapia having a liveweight lower than the edible size of 100 g (Edirisinghe, 1986).

Overpopulation could not be effectively controlled by the use of predator fish. To this respect, several methods have been tried, and monosex culture seems to be the one of the best solutions available. Male fish should be selected for this purpose because they grow faster compared to females, and the absence of females prevent breeding (Peggy, 1993) allowing male tilapia to grow faster.

Hand sexing, a possible way of obtaining monosex culture is time consuming, labourious and expensive (Ogalale and Indukar, 1991). Hybrid culture is a genetic manipulating method which requires the maintenance of pure stocks (Lahav and Lahav, 1990; Varadaraj and Pandlan, 1991). This cannot be effective in Sri Lanka due to unavailability of pure tilapia species.

Hormonal treatment is a suitable method to obtain monosex culture of tilapia fish in Srl Lanka. This is a process of converting genetic females into functional males (Nandeesha *et al.*, 1990; Pandian and Varadaraj, 1990). This conversion might be influenced by the basal diet in which the hormone is introduced. The objective of this study was to quantify the influence of the basal diet on the ability of 17-α Methyltestosterone to produce monosex tilapia fish culture.

MATERIALS AND METHODS

Experiments were conducted at the Department of Animal Science, Faculty of Agriculture, University of Peradeniya. A total of 8 male and 24 female Nile tilapia were removed from mature parent fish ponds and introduced to separate ponds. After feeding for two weeks in these ponds, they were re-stocked and allowed to breed in a spawning pond. Eggs were collected and artificially hatched in indoor tanks.

Fingerlings were fed with one of the four different types of diets. Diet I was prepared by mixing 25 g of powdered egg yolk (obtained by boiling hens' eggs at 100°C for 5 min) with 50 ml of ethanol containing 0.125 mg of 17-α methyltestosterone. This hormone added diet was kept at room temperature until ethanol was evaporated and then refrigerated at -4°C.

Diet II was prepared by mixing ground carcasses (after removing the viscera and head) of dried tilapia fish (35% in the diet) and ground shrimp heads (23.5% in the diet). This was mixed with 41.5% other ingredients (Gunawardane *et al.*, 1994). Finally, 17-α methyltestosterone was added as per diet I.

Diet III consisted of 25 g powdered egg yolk mixed with 0.075 mg 17-α methyltestosterone.

Diet IV (control), consisted of normal fish feed (broiler starter) without hormone. The crude protein content of broiler starter was 21%.

Artificially hatched tilapia post-larvae were collected on Day 8, once the yolk sac had completely disappeared. They were placed in indoor aquarium tanks (0.25 m^3) at the rate of 100 per tank and assigned to the 4 dietary treatments (2 tanks/diet).

Post-larvae were fed at 10% of body weight per feeding, at 0800, 1200 and 1600 hour each day. The amount of feed offered was adjusted weekly according to body weight. Feeding was continued for forty days. On the forty first day, the fish were weighed and transferred to an outdoor tank and reared for another two months on the same dietary treatments. At the end of the third month, fish were sexed by visual examination of gonads. Sex ratio was analyzed by using Chi-square method and the growth rate was estimated through regression analysis.

In trial I, to examine the effect of basal feed, percent survival, growth rate and sex ratio of fish resulted from Diet I and Diet II were compared with those of the control diet. In trial II, percent survival, growth rate and sex ratio of Diet III was compared with control diet in order to examine the effect of lower level of hormone incorporation.

RESULTS AND DISCUSSION

Percent survival and sex ratio of fish fed with Diet I, II and broiler starter (control) are given in Table 1, and that of Diet III and control are given in Table 2. Basal diet had no effect on survival rate. Addition of hormone changed the sex ratio (Tables 1 and 2). One hundred percent of the survived population obtained from Diet I and Diet II were males. In the control, the ratio between male and female was approximately 1:1 (Table 1). This observation suggest that the two basal diets did not have any significant

influence (P>0.05) on the effect of 17-α Methyltestosterone in converting genetic females into functional male. Reduction of hormone concentration did not affect sex ratio (Tables 1 and 2).

Table 1. Effect of 17-α Methyltestosterone on sex ratio and survival of fish culture with egg yolk (Diet I) or tilapia fish meal (Diet II) as basal feed.

Feed	No. post-larvae introduced	Sex Ratio (M:F)	Survival (%)
Diet I	100	54:0	54±4
Diet II	100	49:0	49±6
Control	100	22:25	47±3

Diet I - 25 g Egg yolk meal + 0.125mg of 17-alpha Methyltestosterone.
Diet II - 25 g Tilapia fish meal + 0.125 mg of 17 alpha Methyltestosterone.
Control - Normal fish feed (Broiler starter).

Table 2. Sex ratio and survival of tilapia fish culture with egg yolk containing lower level of 17-α Methyltestosterone.

Feed	No. post-larvae introduced	sex Ratio (M:F)	Survival (%)
Diet III	100	72:0	72±3
Control	100	32:37	69±5

Diet III - 25 g Egg yolk meal + 0.075 mg of 17-alpha Methyltestosterone.
Control - Normal fish feed (Broiler starter).

Weekly body weights obtained with Diet I, II and control diet are given in Table 3. The body weights obtained with Diet III and control diet are given in Table 4. The growth rate as calculated on the basis of changes in body weight (Tables 3 and 4), was lower with Diet II (2.3± 0.63 g per

day) than the growth rate obtained with Diet I (3.0±0.72 g per day) or control diet (2.9±0.65 g per day). But this difference was not statistically significant ($p>0.05$).

Table 3. **Mean weekly body weights (± SE) of tilapia fish fed with egg yolk or tilapia fish meal containing 17-α Methyltestosterone 0.125 mg per 25 g of feed.**

Week	Diet I (Egg yolk+hormone)	Diet II (Fish meal+hormone)	Control (broiler starter)
0	0.013±0.02	0.013±0.02	0.013±0.02
1	0.034±0.04	0.040±0.03	0.035±0.03
2	0.068±0.07	0.057±0.06	0.060±0.07
3	3.30 ±0.20	2.50 ±0.30	3.00 ±0.20
4	9.00 ±0.40	5.50 ±0.50	7.00 ±0.30
5	15.55 ±0.30	12.50 ±0.60	14.00 ±0.40

Table 4. **Mean weekly body weights (± SE) of tilapia fish fed with egg yolk containing 17-α Methyltestosterone 0.075 mg per 25 g of feed.**

Week	Diet III (Egg yolk+hormone)	Control (Broiler starter)
0	0.02±0.01	0.02±0.02
1	0.05±0.02	0.04±0.03
2	0.10±0.02	0.08±0.04
3	4.50±0.30	4.00±0.25
4	10.50±0.50	9.80±0.43
5	17.50±0.45	15.00±0.55

The tilapia fish meal used in this study was initially prepared for feeding shrimp. This meal consisted of 23.5% shrimp head meal, 35% tilapia fish meal and 41.5% other ingredients. The lower growth rate obtained with Diet II may be due to the presence of growth inhibiting compounds in the

prepared tilapia fish meal, resulting low digestibility. The protein availability in egg yolk meal is high when compared to the other two meals. This may have contributed to the observed higher growth rate of fish when fed with egg yolk (Table 3 and 4).

Eventhough 17-α Methyltestosterone had been used in other studies to obtain monosex culture, a detailed preparation of the basal diet has not been mentioned in any of those studies. The present study clearly indicates that the basal diet containing egg yolk or fish meal did not exert any influence on the sex changing effect of 17-α Methyltestosterone when it is incorporated at the level of 0.125 mg or 0.075 mg per 25 g feed.

CONCLUSIONS

It could be concluded that the basal diet containing egg yolk or fish meal did not have any influence on the production and/or growth rate of monosex tilapia when 17-α Methyltestosterone was used. For this methodology to be extended to commercial scale, suitable refined steps have to be developed to obtain a 100% male population.

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