

## **EFFECTS OF 17 $\alpha$ - METHYLTESTOSTERONE ON GROWTH PERFORMANCE AND SOME PHYSIOLOGICAL CHANGES OF NILE TILAPIA, *OREOCHROMIS NILOTICUS* L.) FINGERLINGS**

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### **ABSTRACT**

Different doses of 17  $\alpha$ -methyltestosterone hormone (MT) used as growth promoter was administrated to Nile tilapia; *Oreochromis niloticus* in fishmeal based pelleted diet for 90 days. The obtained results showed that low doses of 0.5, 1.0 and 2.5 mg MT/kg feed were not effective meanwhile the dose of 5.0 mg/kg was the optimum effective one in promoting significant final weight, weight gain and SGR of Nile tilapia. HSI was insignificantly changed at low MT doses, and slightly increased at high MT doses. Male and female GSI was significantly decreased at high MT doses (10, 20 and 40 mg/kg), while insignificant changes were observed at low doses. Feed intake and FCR were slightly changed at different doses of MT. The higher PER was obtained with moderate MT doses and the least ones were obtained with 20, 40 mg/kg or control. The changes in erythrocyte counts, haemoglobin content, haematecrit value and plasma glucose showed insignificant differences at all treatments. A significant reduction of plasma total protein was observed in fish fed 40 mg MT/kg, whereas it was insignificantly changed with other treatments. In contrast, the highest level in plasma total lipids was obtained at 40 mg MT/kg. In fish plasma, the activity of ALT was the highest with control fish and that fed low doses of 0.5 - 2.5 mg MT/kg, while the least one was obtained with 40 mg/kg. In contrast, AST activity was significantly increased with high MT doses of 20 and 40 mg/kg, while there were no significant changes among other treatments. The higher

activity of alkaline phosphatase (ALP) was obtained at low MT doses (0 to 2.5 mg/kg), after which ALP activities decreased to reach the lowest one at 40 mg/kg. Hepatic ALT and AST activities were increased with increasing MT doses where the maximum ALT and AST activities were obtained at 40 mg/kg, while the minimum ones were obtained at MT doses of 0 to 5 mg/kg. In fish muscle, the activity of ALT and AST showed significant reduction with increasing MT doses where the minimum one was obtained with 40 mg/kg. On the other hand, AST activity was insignificantly changed at all treatments.

## INTRODUCTION

Steroid hormones are usually used to induce sex inversion, and this technique is one of the most common techniques used to obtain all-male population (Mires, 1995; Gale *et al.*, 1999). Moreover, steroid hormones could be used as growth promoter in fish where they enhanced the weight gain of studied fish and enhanced the rate of muscle protein accretion (Higgs *et al.*, 1977; Donaldson *et al.*, 1979; Lone and Matty, 1980; Ostrowski and Garling, 1988). However, the value in using growth promoters in aquaculture depends upon the ability of these agents to enhance, or at least maintain normal rates of muscle deposition relative to the growth of other body components in treated fish. A decrease in percentage of muscle in the body with treatment would lower the actual efficiency of the anabolic effect. Conversely, an increase in the percentage of muscle would enhance the efficiency of treatment. Fagerlund and McBride (1975) found that a high treatment dose of methyltestosterone increased weight gains of coho salmon (*Oncorhynchus kisutch*), but the percentage of flesh weight of the fish decreased. In contrast, steroid treatments have increased the condition factor of some salmonids (Fagerlund and McBride, 1975 & 1977; Saunders *et al.*, 1977) suggesting the increase in the percentage of the muscle in fish body. An understanding of how anabolic steroids affect the efficiency of muscle deposition in fish would aid in evaluating the use of these agents in aquaculture.

Fish masculinization has increased somatic growth rate due to the avoidance of energy losses associated with gonads development and reproduction. Moreover, masculinized fish are desirable because they achieve a larger final size than females (MacIntosh and Little,

1995). Meyer (1991) studied the growth enhancement due to sex-reversal and the growth enhancement due to anabolic effects among 17  $\alpha$ -methyltestosterone (MT) treated and control *Tilapia hornorum*, *T. nilotica* and their hybrid. He observed the growth enhancement of these fishes which, representing the isolated anabolic effect of the MT.

The objective of this study was to investigate using of MT as a growth promoter on the growth performance, survival rate, feed utilization and proximate chemical analysis of Nile tilapia; *Oreochromis niloticus* L., and its impact on some biochemical parameters as well.

## MATERIALS AND METHODS

### Feed Preparation

The chemical used was 17  $\alpha$ -methyltestosterone hormone (MT) produced by Sigma (St. Louis, MO) as sources for MT. A semi-purified fish diet containing 32% crude protein, 10.0% crude lipids, 7.5% ash, 5.6% fibers and moisture 8.36% was prepared. The fabricating of MT-feed was done as described in Teichert-Coddington *et al.* (2000). Different levels of MT were dissolved in 95% ethyl alcohol at a rate of 75ml/kg feed and sprayed to fish diet during rapid blending in a small ribbon mixer. Alcohol was evaporated from the feed by forced air as it was slowly conveyed from the mixer to the keeping plastic bags, which stored in a refrigerator (2 °C). The tested diets were approximately similar in all the nutrient contents but containing different levels of MT. The different MT concentrations were 0.5, 1.0, 2.5, 5.0, 10, 20 and 40 mg/kg feed, and control diet did not contain MT.

### Fish Culture Technique

Healthy fish of Nile tilapia (*Oreochromis niloticus* L.) weighing 10-15 g/fish were collected from the nursery ponds of Central Laboratory for Aquaculture Research at Abbassa, Abo-Hammad, Sharqia, Egypt. Fish were acclimated in indoor tank for 2 weeks to laboratory conditions. Ten fish were frozen at -20 °C for chemical analysis. The fish with mixed sex were distributed randomly in glass aquaria of 130-liter capacity at a rate of 20 fish/aquarium that containing aerated tap water. Each aquarium was supplied with

compressed air via air-stones from air pumps.

One of the tested diets was fed to fish frequently at a rate of 3% of live body weight twice daily for 90 days. Each MF level was randomly assigned by three replicates. Siphoning a portion of water from each aquarium was done every day for excreta removing and an equal volume of well-aerated tap water was provided from a storage fiberglass tank. The temperature was adjusted at  $27 \pm 1$  °C by using thermostats. Fish in each aquarium was biweekly weighed and subsequently the amount of given feed was calculated. Dead fish were removed and recorded daily.

### Proximate Analysis of Diet and Fish

The basal diet and pooled samples of 12 fish from each treatment were analyzed using standard methods of the Association of Official Analytical Chemists (AOAC 1990) for moisture, protein, fats and ash.

### Growth Parameter

Growth performance was determined and feed utilization was calculated as follows:

$$\text{Weight gain} = W_2 - W_1$$

$$\text{Specific growth rate (SGR)} = 100 (\ln W_2 - \ln W_1) / T$$

Where  $W_1$  and  $W_2$  are the initial and final w, respectively, and T is the number of days of the feeding period.

$$\text{Feed conversion ratio (FCR)} = FI / (B_2 - B_1)$$

Where FI,  $B_1$  and  $B_2$  are the feed intake, the biomass at the start and end, respectively.

$$\text{Protein efficiency ratio (PER)} = (B_2 - B_1) / PI$$

Where  $B_1$  and  $B_2$  are the biomass at the start and the end of the experiment, and PI is the protein intake.

$$\text{Hepatosomatic index} = \text{weight of liver} / \text{weight of fish} \times 100$$

$$\text{Gonadosomatic index} = \text{weight of gonads} / \text{weight of fish} \times 100$$

### Biochemical Analyses

At the end of experiment, the blood samples were taken from caudal vein of an anaesthetized fish by sterile syringe using EDTA solution as anticoagulant. These blood samples were used for determining erythrocyte count, haemoglobin content and haematocrit

value (Britton, 1963). Plasma was obtained by centrifugation at 3000 rpm for 15 min and nonhaemolyzed plasma was stored in deep freezer for further biochemical analyses. After decapitation of fish, samples of liver and muscle were taken and frozen for further biochemical analysis. Activities of aspartate aminotransferase (AST) and alanine aminotransferase (ALT) were determined colorimetrically using AST and ALT kits reagent supplied by Egyptian American Co. for Laboratory Services, Egypt, according to Reitman and Frankel (1957). Alkaline phosphatase (ALP) was measured by using kits reagent supplied by Diamond Diagnostic Co. according to Rec (1972). Glucose was determined using glucose kits supplied by Boehring Mannheim kit according to Trinder (1969). Total protein content in plasma, muscle and liver was determined colorimetrically using protein kits reagent supplied by Egyptian American Co. for Laboratory Services, Egypt, according to Henry (1964). Total lipids contents in plasma, muscle and liver was determined colorimetrically using lipid kits reagent supplied by Egyptian American Co. for Laboratory Services, Egypt, according to Joseph *et al.* (1972).

### Statistical Analysis

The obtained data were subjected to one-way analysis of variance according to Snedecor and Cochran (1982). Differences between means were at the 5% probability level using Duncan's new multiple range test (Duncan, 1955).

## RESULTS

### Growth Performance Parameters

Results in Table 1 showed that the final weight of Nile tilapia was significantly increased ( $P < 0.05$ ) with increasing MT doses up to 5 mg MT/kg. No significant changes were observed among doses of 5 to 40 mg MT/kg and ranged from 31.1 to 31.8 g/fish. There is no significant difference between control and low MT doses and final weight was ranged from 27.9 to 28.8 g/fish;  $P > 0.05$ ). Similarly, weight gain and SGR were increased significantly with moderate MT doses (5 and 10 mg/kg) to the same value (20.5 g/fish) and insignificantly decreased with high MT doses (20 and 40 mg/kg; 19.8 and 19.6 g/fish, respectively).

The effect of different MT doses on hepatosomatic (HSI) and

gonadosomatic (GSI) indices was also studied. Fig 1 showed that HSI was insignificantly changed at low MT doses, and slightly increased at high MT doses. However, the maximum HSI was obtained at 5 mg MT/kg (2.519), while the lowest one was obtained at control (1.767). On the other hand, female and male GSI was significantly decreased at high MT doses (10, 20 and 40 mg/kg), while insignificant changes were observed at low doses (Fig 1). The maximum GSI of female and male fish was obtained at control (3.78 and 2.798, respectively), while the lowest ones were obtained at 40 mg MT/kg (1.86 and 1.45, respectively).

Results in Fig 2 show that feed conversion rate (FCR) was slightly changed and ranged from 1.54 to 1.88. Also, Fig 2 indicated that protein efficiency ratio (PER) increased with increasing MT doses up to 5 mg/kg (2.22) and slightly declined up to 40 mg/kg (2.1). The least PER was obtained with control fish (1.81).

Results in Table 2 show that moisture content in fish body was significantly higher in fish fed 10 and 5 mg MT/kg (77.03 and 76.2%, respectively;  $P < 0.05$ ), while the moisture content at 20 and 40 mg/kg was similar to that of low MT doses and control and ranged from 74.13 to 74.76% without significant difference. Protein contents in fish body were the lowest in control fish (55.3%) followed by that of 20 and 40 mg MT/kg (56.64 and 56.34%, respectively;  $P > 0.05$ ). Contrarily, total lipids contents in fish body were the highest in control fish (24.27%) and doses of 10-40 mg/kg, while the less ones were obtained at doses from 0.5 to 50 mg/kg (18.31%-20.12%;  $P > 0.05$ ). Ash content was higher with doses from 0.5 to 5 mg/kg (23.57%-21.78%;  $P > 0.05$ ), whereas the least ones were obtained with control and 40 mg/kg.

### Physiological Parameters

Results in Table 3 show that the changes in erythrocyte counts were insignificant, and ranged from 1.486 to 1.834  $10^6/\text{mm}^3$ . Also, haemoglobin content and haematecrit value showed insignificant differences and ranged from 4.55 to 5.66 g/100 ml for haemoglobin content and from 15.0 to 18.8% for haematecrit value.

Concerning glucose level in fish plasma, data in Table 4 show that there were no significant changes in all treatments and ranged from 123.3 to 141.9 mg/L. A significant reduction of plasma total protein was observed in fish fed 40.0 mg MT/kg (1.51 g/100ml), whereas the plasma total protein was insignificantly changed with

other treatments and ranged from 1.744 to 2.602 g/100ml (Table 4). In contrast, the highest level in plasma total lipids was obtained at 40.0 mg MT/kg (31.5 g/L), while the plasma total protein was insignificantly changed with other treatments and ranged from 19.1 to 22.0 g/100 ml.

As shown in Table 5, in fish plasma, the activity of ALT was the highest with control fish and that fed low doses of 0.5 to 2.5 mg MT/kg (20.6-17.5 IU/L), while the least one was obtained with 40.0 mg/kg (11.1 IU/L). In contrast, AST activity in fish plasma was significantly increased with high MT doses of 20 and 40 mg/kg (143.9 and 165.1 IU/L, respectively), while there was no significant changes among other treatments (90.9-99.0 IU/L;  $P < 0.05$ ). Concerning alkaline phosphatase (ALP) activity in fish plasma, the higher activities were obtained at low MT doses (0 to 2.5 mg/kg) after which ALP activities decreased till the lowest one at 40 mg/kg (24.7 IU/L;  $P < 0.05$ ).

Hepatic ALT and AST activities were increased with increasing MT doses where the maximum ALT and AST activities were obtained at 40 mg/kg (19.8 and 140.6 IU/g fresh wt, respectively), while the minimum ones were obtained at MT doses of 0 to 5 mg/kg (Table 5).

Concerning ALT and AST activities in fish muscle, data in Table 6 show that ALT activity decreased with increasing MT doses where the minimum one was obtained with 40.0 mg/kg (14.1 IU/g fresh wt). On the other hand, AST activity in fish muscle was insignificantly changed at all treatments and ranged from 259.6 to 279.7 IU/g fresh wt.

## **DISCUSSION**

Anabolic steroids are potentially useful compounds in aquaculture due to their ability to increase weight gains and muscle deposition of treated fish. Steroid treatment may produce fish of more robust size containing more muscle per unit length than untreated fish without changing the anabolic efficiency or proportion of muscle to whole body weight (Ostrowski and Garling, 1988). The obtained results in the present study revealed that administration of 17  $\alpha$ -methyltestosterone (MT) induced significant increase in fish growth of treated Nile tilapia. These results are in accordance with those of

Lone and Matty (1980), Lewis and Sower (1992), Woo *et al.* (1993), Satpathy *et al.* (1995) and Sambhu and Jayaprakas (1997).

The increase in fish growth may be because of that MT induced the feed digestion and absorption rate causing increase in body weight (Yamazaki, 1976). Furthermore, Lone and Matty (1981) mentioned that MT administration increased the proteolytic activity of the gut in mirror carp (*Cyprinus carpio*) leading to increase the growth rate. Whether this increased enzyme activity might have a direct effect of the steroids on *de novo* synthesis of protease enzyme in the gut. Furthermore, MT treatment may stimulate thyroid and internal functions as well as insulin secretion from the pancreatic B cells of fish (Van Overbeeke and McBride 1971; Higgs *et al.*, 1976; Higgs *et al.*, 1977). Also, androgenic steroids may promote the release of growth hormone from the pituitary somatotrops fish (Higgs *et al.*, 1976).

The effect of 17  $\alpha$ -methyltestosterone on the gonads appears to be complex. In our study, MT administration excess of 2.5 mg/kg induced the degenerative changes in the ovaries and testes similar to those of Yamazaki (1972) in pink and chum salmon and by Hirose and Hibiya (1968 a, b) in goldfish rainbow trout. Furthermore, Higgs *et al.* (1977) found clear signs of gonads degeneration in coho salmon affected by MT causing fish sterility, which might be considered advantageous in fish culture because less food energy would be channeled into gonads development and therefore more would be available for body growth.

The water content in fish body was significantly higher in fish fed 5 and 10 mg MT/kg, while at 20 and 40 mg/kg it was similar to that of low MT doses and control. Protein contents in fish body were the lowest in control, 20 and 40 mg MT/kg. Contrarily, total lipid contents were the highest in control fish and doses of 10-40 mg/kg, while the less ones were obtained at doses from 0.5 to 5 mg/kg. Ash content was higher with doses from 0.5 to 5 mg/kg, whereas the least ones were obtained with control and 40 mg/kg. These changes in proximate fish body composition may be related to hormone doses. In this regard, Fagerlund and McBride (1975) and Higgs *et al.* (1977) found that water content and lipid content were higher, while protein content decreased in MT-treated coho salmon than control. These results are acceptable because the high doses of anabolic steroids have often produced side effects in treated fish, including changes in



proximate composition of muscle (Fagerlund and McBride, 1975, 1977; Lone and Matty, 1980).

The metabolic pathways of fish could be distinguished throughout assessing some physiological parameters. The present study showed insignificant changes in glucose level, erythrocyte count, haemoglobin level and haematocrit value, however, these results reflect the healthy status of the cultured fish at all treatments. Concerning the plasma total protein, it was significantly decreased at high MT doses. This result may be due to the fact that androgens regulate protein synthesis by binding to cytosolic or nuclear receptors for steroids that then modulates transcription (Chan and O'Malley, 1976; O'Malley and Tsai, 1992). Moreover, the decrease in plasma total lipids in fish fed low levels of MT may be due to the increase of energy demand, which led to more consumption of protein and lipids (Dange and Masurekar, 1984).

Transamination represents one of the principal pathways for synthesis and deamination of amino acids, thereby allowing an interplay between carbohydrates and protein metabolism during the fluctuating energy demands of the organism in various adaptive situations. They also are considered to be important in the assessment of the state of the liver as well as some of the organs (Verma *et al.*, 1981). Therefore, attention has been focused on the changes in ALT and AST activities, which promote gluconeogenesis from amino acids, as well as the effects of changes in aminotransferase activities on the liver condition (Hilmy *et al.*, 1981; Rashatwar and Ilyas, 1983). Furthermore, the changes of ALT, AST and ALP activities might be altered by a variety of chemical, biological and physiological factors. The alterations in enzymes activities may be due to the disturbance in the Krebs's cycle. Decreased activity of Krebs's cycle causes a decrease in Krebs's cycle intermediates, thereby, ALT and AST compensate through providing  $\alpha$ -ketoglutarate (Salah El-Deen and Rogers, 1993).

Detailed studies on the environmental fate of androgens are not available but under certain conditions may produce secondary effects. MT is susceptible to breakdown when exposed to light or high temperature (AHFS Drug information, 1997). Both fungi and bacteria can metabolize exogenous steroids. Many different steroid metabolic reactions, including metabolism of MT, are possible in bacteria (Schubert *et al.*, 1972; Jankov, 1977) as well as metabolism

of steroids to CO<sub>2</sub> and H<sub>2</sub>O (Sandor and Mehdi, 1979). In an outdoor ponds, the combination of light, temperature and microbial degradation should result in a rapid break down of MT. Phelps *et al.* (2000) studied the fate of MT when given to tilapia fry in outdoor hapa. They found MT levels in water from within the treatment hapa to be in general similar to pretreatment levels of pond water. Likewise MT levels in soils were similar pre and post treatment.

On the other hand, digested MT is rapidly metabolized and excreted. Curtis *et al.* (1991) fed tilapia fry for 30 days a feed containing radioactive labeled MT. Ten days after 21-d treatment only a trace of MT could be found. In a study by Goudie *et al.* (1986), the head and viscera were found to contained >90% of the radio-labeled MT, and after 21 days post-treatment <1% remained. Johnstone *et al.* (1983) found >95% of the radio-labeled MT in the viscera and no radioactivity could be found 50 h post-treatment. This rapid metabolism and excretion of MT by a fish treated early in its life history, combined with the extended period needed to produce a marketable size fish results in a safe consumer product (Phelps, 2001).

Finally, from the obtained results, it could be recommended that the optimum dose of 17  $\alpha$ -methyltestosterone hormone that could be used as growth promoter for Nile tilapia was 5 mg/kg feed. Also, the application of hormone should ceased before 30 days of human consumption through which the applied hormone would breakdown and fish would be safe for human consumption.

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**EFFECTS OF 17  $\alpha$ - METHYLTESTOSTERONE ON SOME  
PHYSIOLOGICAL CHANGES OF NILE TILAPIA  
FINGERLINGS**

**Table 1.** Growth performance parameters of Nile tilapia (*O. niloticus*) fed diet containing different levels of 17  $\alpha$ -methyl testosterone hormone (mg/kg feed) used as growth promoter.

Items	Treatments							
	Control	0.5	1.0	2.5	5.0	10	20	40
Initial weight (g/fish)	11.5 $\pm$ 0.3	11.4 $\pm$ 0.2	11.3 $\pm$ 0.1	11.1 $\pm$ 0.3	10.8 $\pm$ 0.2	11.3 $\pm$ 0.2	11.4 $\pm$ 0.4	11.5 $\pm$ 0.1
Final weight (g/fish)	27.9 b $\pm$ 0.55	28.5 b $\pm$ 0.03	28.4 b $\pm$ 0.43	28.8 b $\pm$ 0.98	31.3 a $\pm$ 0.15	31.8 a $\pm$ 0.09	31.2 a $\pm$ 0.09	31.1 a $\pm$ 1.44
Weight gain (g/fish)	16.4 c $\pm$ 0.72	17.1 c $\pm$ 0.06	17.1 c $\pm$ 0.35	17.7 bc $\pm$ 1.01	20.5 a $\pm$ 0.30	20.5 a $\pm$ 0.15	19.8 ab $\pm$ 0.43	19.6 ab $\pm$ 1.30
S G R (%/d)	0.985 d $\pm$ 0.042	1.017 cd $\pm$ 0.003	1.023 cd $\pm$ 0.009	1.059 bcd $\pm$ 0.04	1.182 a $\pm$ 0.023	1.149 ab $\pm$ 0.018	1.118 ab $\pm$ 0.038	1.105 abc $\pm$ 0.04
Survival (%)	100.0 a $\pm$ 0.0	100.0 a $\pm$ 0.0	96.7 a $\pm$ 2.04	96.7 a $\pm$ 2.04	100.0 a $\pm$ 0.0	96.7 a $\pm$ 2.04	96.7 a $\pm$ 2.04	96.7 a $\pm$ 2.04

The same letter in the same row is not significantly different at  $P < 0.05$ .

**Table 2.** Proximate chemical analysis (on dry matter basis) of whole body of Nile tilapia (*O. niloticus*) fed diet containing different levels of 17  $\alpha$ -methyl testosterone hormone (mg/kg feed) used as growth promoter.

Items %	Treatments							
	Control	0.5	1.0	2.5	5.0	10	20	40
Moisture	74.13 d ± 0.12	74.26 d ± 0.15	74.56 d ± 0.26	75.43 c ± 0.33	76.20 b ± 0.32	77.03 a ± 0.18	74.76 cd ± 0.13	74.47 d ± 0.27
Crude Protein	55.3 c ± 0.25	58.10 a ± 0.44	57.50 a ± 0.40	58.17 a ± 0.20	58.13 a ± 0.06	57.41 a ± 0.25	56.64 b ± 0.35	56.34 b ± 0.20
Total lipids	24.27 a ± 0.58	18.31 c ± 1.25	19.32 c ± 0.99	20.12 bc ± 0.49	20.12 bc ± 0.57	21.96 ab ± 0.67	21.53 ab ± 0.09	22.13 ab ± 0.26
Ash	20.30 c ± 0.56	23.57 a ± 0.47	23.21 ab ± 0.56	21.63 abc ± 0.84	21.78 abc ± 0.55	20.43 c ± 0.60	21.70 abc ± 0.61	21.43 bc ± 0.49

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Table 3. Changes in erythrocyte count, haemoglobin content and haematocrit value in the blood of Nile tilapia (*O. niloticus*) fed diet containing different levels of 17  $\alpha$ -methyl testosterone hormone (mg/kg feed) used as growth promoter.

Items	Treatments							
	Control	0.5	1.0	2.5	5.0	10	20	40
Erythrocyte count ( $10^6/\text{mm}^3$ )	1.518 a $\pm 0.054$	1.486 a $\pm 0.148$	1.650 a $\pm 0.089$	1.610 a $\pm 0.145$	1.704 a $\pm 0.094$	1.694 a $\pm 0.087$	1.554 a $\pm 0.113$	1.834 a $\pm 0.073$
Haemoglobin content (g/100 ml)	4.55 a $\pm 0.43$	4.85 a $\pm 0.29$	4.70 a $\pm 0.51$	4.99 a $\pm 0.29$	5.35 a $\pm 0.26$	5.66 a $\pm 0.65$	5.22 a $\pm 0.42$	4.87 a $\pm 0.61$
Haematocrite value (%)	17.2 a $\pm 1.1$	18.4 a $\pm 1.2$	18.4 a $\pm 1.7$	16.0 a $\pm 0.4$	15.0 a $\pm 1.2$	15.6 a $\pm 0.5$	18.6 a $\pm 1.4$	18.8 a $\pm 1.2$

The same letter in the same row is not significantly different at  $P < 0.05$ .

**Table 4.** Changes in glucose, protein and lipids in the plasma of Nile tilapia (*O. niloticus*) fed diet containing different levels of 17  $\alpha$ -methyl testosterone hormone (mg/kg feed) used as growth promoter.

Items	Treatments							
	Control	0.5	1.0	2.5	5.0	10	20	40
Glucose (mg/L)	125.5 a ± 4.9	123.8 a ± 7.1	132.6 a ± 7.6	123.3 a ± 6.6	123.9 a ± 8.8	120.0 a ± 9.5	141.9 a ± 10.0	129.4 a ± 7.6
Protein (g/100 ml)	2.116 ab ± 0.344	1.998 ab ± 0.304	1.994 ab ± 0.313	2.116 ab ± 0.376	1.998 ab ± 0.321	2.602 a ± 0.325	1.744 ab ± 0.199	1.510 b ± 0.171
Lipids (g/L)	19.1 b ± 1.9	21.9 b ± 1.5	19.1 b ± 1.8	21.3 b ± 1.1	21.9 b ± 1.4	22.0 b ± 2.5	19.9 b ± 1.2	31.5 a ± 3.1

The same letter in the same row is not significantly different at  $P < 0.05$ .

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**Table 5.** Changes in activities of alanine aminotransferase (ALT), aspartate aminotransferase (AST) and alkaline phosphatase (ACP) in plasma, muscle and liver of Nile tilapia (*O. niloticus*) fed diet containing different levels of 17  $\alpha$ -methyl testosterone hormone (mg/kg feed) used as growth promoter.

Items	Treatments								
	Control	0.5	1.0	2.5	5.0	10	20	40	
Plasma	ALT (TU/L)	20.6 a ± 1.1	17.5 ab ± 1.2	17.6 ab ± 0.9	17.5 ab ± 1.4	16.0 b ± 1.06	16.2 b ± 0.6	16.7 b ± 0.7	11.1 c ± 1.1
	AST (TU/L)	92.5 b ± 9.9	92.4 b ± 8.0	90.9 b ± 14.2	99.0 b ± 5.9	91.5 b ± 8.1	91.4 b ± 31.4	143.9 a ± 9.0	165.1 a ± 12.5
	ACP (TU/L)	57.4 ab ± 3.1	57.2 ab ± 5.0	63.4 a ± 3.5	59.3 ab ± 3.2	51.4 bc ± 3.9	41.6 cd ± 3.8	38.9 d ± 2.0	24.7 e ± 3.2
Liver	ALT (TU/g)	9.8 d ± 0.7	8.4 d ± 0.8	11.3 cd ± 1.4	10.0 cd ± 1.3	10.3 cd ± 1.8	14.8 bc ± 2.6	17.9 ab ± 1.2	19.8 a ± 1.6
	AST (TU/g)	76.9 d ± 6.1	84.1 d ± 3.8	84.1 d ± 4.9	83.9 d ± 9.3	87.8 cd ± 4.3	107.2 bc ± 6.9	118.5 b ± 8.0	140.6 a ± 8.1
Muscle	ALT (IU/g)	34.1 a ± 1.7	37.8 a ± 2.0	38.5 a ± 2.7	34.7 a ± 2.8	26.3 b ± 1.9	20.6 b ± 1.5	21.5 b ± 1.8	14.1 c ± 1.1
	AST (IU/g)	273.7 a ± 9.4	275.4 a ± 14.3	268.8 a ± 14.5	271.5 a ± 11.2	261.1 a ± 16.5	275.4 a ± 14.6	259.6 a ± 15.3	279.7 a ± 16.2

The same letter in the same row is not significantly different at  $P < 0.05$ .

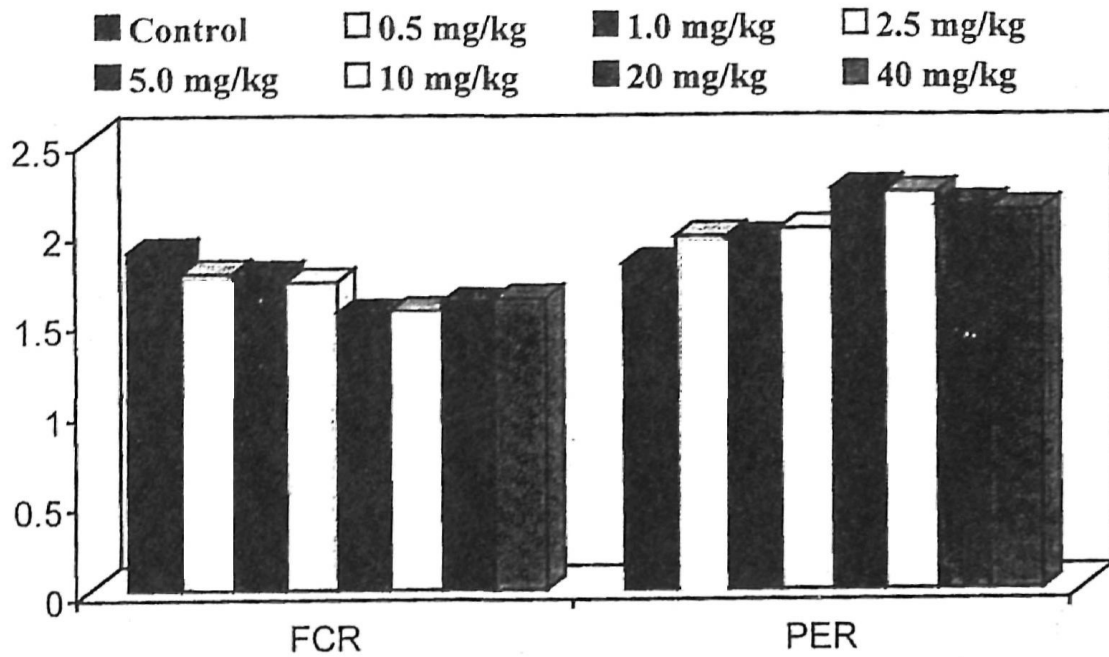


Fig. 2. Changes in feed conversion ratio (FCR) and protein efficiency ratio (PER) of Nile tilapia fed different doses of MT hormone (mg/kg) used as growth promoter for 90 days.

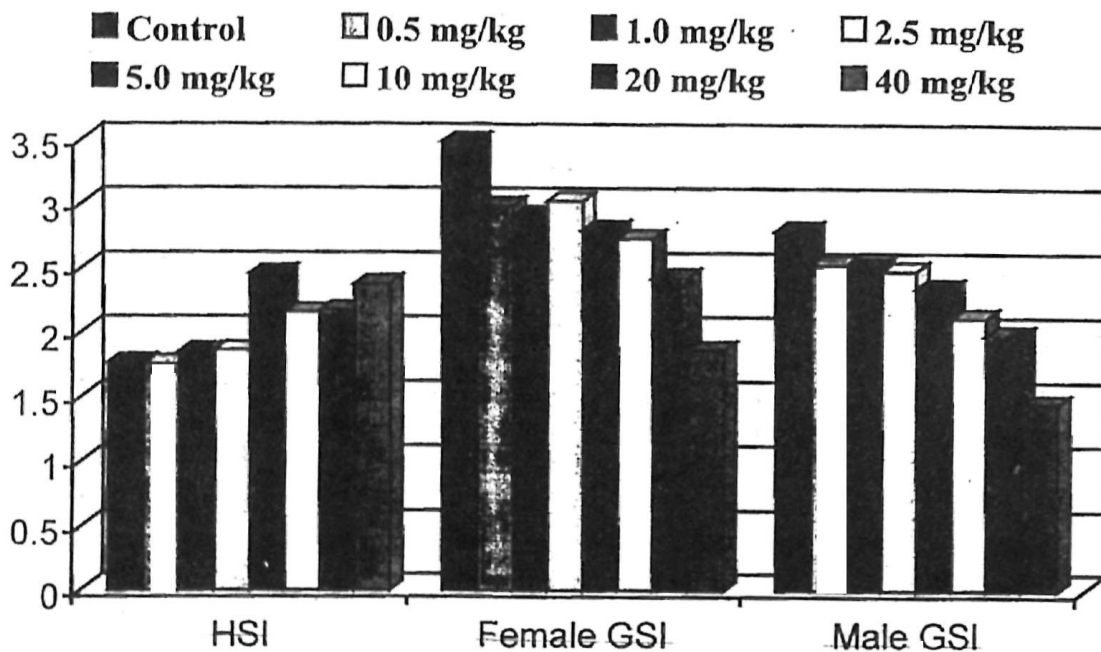


Fig. 1. Changes in hepatosomatic (HSI), and female and male gonadosomatic (GS) indices of Nile tilapia (*O. niloticus*) fed diet containing different doses of M hormone (mg/kg feed) used as growth promoter for 90 days.