# Effect of administering different doses of 17α-methyltestosterone in Heteropneustes fossilis

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Abstract. Heteropneustes fossilis were injected 0, 2.5, 5, 10, 20, 40, 60 or 80 mg 17amethyltestosterone/kg fish. They were fed ad libitum on chopped Lepidocephalichthys thermalis and reared for 21 days. The steroid acts as an appetite stimulant and consequently both feeding and growth rates increased 2 to 3 times, when given the optimum dosage of 40 mg/kg. It also doubles the growth efficiency. 17α-methyltestosterone is recognized as an appetite stimulant and an anabolic steroid.

Keywords. Catfish; anabolic steroid; appetite stimulant; growth efficiency; Heteropneustes fossilis.

## Introduction

In recent years, the anabolic steroids, which are known to augment growth rate and reduce feed-cost in animal husbandry, have attracted the attention of pisciculturists. 17α-methyltestosterone, an anabolic steroid, is very much recommended in livestock industry. Clemens et al (1966) administered 20–30 mg 17α-methyltestosterone/kg feed to female guppy (Lebistes reticulatus) and recorded negative growth. Conversely, Fagerlund and McBride (1979) treated Oncorhynchus kisutch with 10 mg/kg of the steroid and reported that the treatment enhanced the growth efficiency. These contradictory results suggest that perhaps the anabolic property of 17α-methyltestosterone is dose-dependent. Also it is not clear whether the optimum dose is a species-specific character. Pandian (1982) recognised that while some steroids enhanced growth rate through increased food consumption, the others accomplished it through increased growth efficiency. He named the former as appetite stimulants and the latter as ones with anabolic property. The present investigation was undertaken to identify the optimum dosage of  $17\alpha$ -methyltestosterone to the catfish H. fossilis and to know whether the steroid displays anabolic property alone or acts also as an appetite stimulant.

## Material and methods

Juveniles of H. fossilis  $(9 \pm 0.2 \text{ g})$  were acclimated to the laboratory conditions and feeding schedule. Eight groups, each comprising three individuals were used. The experiments were performed in cylindrical aquaria (40 cm diameter and 141 capacity) for 21 days in a laboratory (29  $\pm$  1°C), where except for feeding and observation there was no disturbance.

The fish were fed ad libitum on chopped and weighed Lepidocephalichthys thermalis

for 2 hr every day between 0900 and 1100 hr. Unfed remains were collected carefully and suitable corrections made for fluid loss  $(5 \pm 0.27 \%)$  by fish pieces during the feeding period. Faeces was collected by filtering the aquarium once in three days.

The 'sacrifice method' (Maynard and Loosli 1962) was used to determine the water content and energy values of the test individuals before the commencement of the experiment. Before taking the initial and final weights, the test fish were subjected to starvation for 24 hr to ensure complete evacuation of the gut. The test fish were weighed in a single pan analytical balance sensitive to 0·1 mg. Caloric contents of fish, food and faeces were determined using a semi-microbomb calorimeter (Parr Instrument Co., Moline, USA) following the procedure described in the instruction manual for Bomb calorimetry.

The following scheme of energy balance was followed (Petrusewicz and MacFadyen 1970):

$$C = F + U + M + P,$$

where C is the food consumed, F the faeces, U the urine, M the energy lost as heat due to metabolism and P the growth (= conversion). Food energy absorbed (A) was calculated by subtracting F from C and P by subtracting the energy content of the fish at the commencement of the experiment from that at termination. Rates of feeding, absorption and conversion were calculated by dividing the respective quantum of energy by the products of fish weight (g) and the duration of the experiment (21 days) and expressed in terms of J/g live weight/day. Efficiency (%) of absorption was calculated by relating A to C, and the conversion efficiency (%) by relating P to A.

The required doses of the tested hormone were prepared using sesame oil as the carrier solution. Individuals in the control group received sesame oil alone. Volume of the hormone solution injected intramuscularly into the test individuals was maintained constant at  $60 \mu l$  for all the tested doses, using  $100 \mu l$  sterilized tuberculin syringe (Scientific Glass Engineering, Australia).

## 3. Results

## 3.1 Consumption

H. fossilis treated with different doses of  $17\alpha$ -methyltestosterone consumed significantly more food than the sham-treated ones. With increasing dose, feeding rate progressively increased from 182 J/g live fish/day in the control to 303 J/g live fish/day in that treated with 40 mg/kg (table 1). However, higher doses of 60 or 80 mg/kg resulted in the depression of feeding rate. On statistically analysing the data, it became apparent that the differences between those receiving 40-60 mg/kg on one hand, and the remaining groups on the other were highly significant (P < 0.01). Therefore,  $17\alpha$ -methyltestosterone acts as an appetite stimulant in H. fossilis where maximum appetite is stimulated, when the fish is treated with 40-60 mg/kg (figure 1)

## 3.2 Absorption

Absorption efficiency ranged from 96.6-96.9%. Analysis of variance of the data in absorption efficiency revealed that neither the hormone nor the doses have any

**Table 1.** Rates of consumption, absorption, production (J/g live fish/day) and efficiencies (%) of absorption and conversion as a function of dosage of  $17\alpha$ -methyltestosterone in H. fossilis  $(9 \pm 0.2 \text{ g})$ .

Dosage (mg/kg)	Consumption rate (Cr)	Absorption rate (Ar)	Production rate (Pr)	Absorption efficiency	Net conversion efficiency
0	182 ± 11·0	177 ± 10·6	38 ± 2·7	96.9 + 0.0	$21.7 \pm 2.6$
2.5	$195 \pm 3.2$	$190 \pm 3.0$	$41 \pm 5.9$	$96.9 \pm 0.3$	$21.4 \pm 2.9$
5	$196 \pm 1.6$	190 ± 1.9	$49 \pm 3.9$	$96.8 \pm 0.2$	$25.9 \pm 2.3$
10	235 ± 17·6	$230 \pm 19.3$	64 ± 2·4	$96.6 \pm 0.2$	$28.5 \pm 1.9$
20	$241 \pm 11.2$	$234 \pm 10.0$	75 ± 4·6	$96.7 \pm 0.2$	$32.3 \pm 1.4$
40	$303 \pm 16.2$	$293 \pm 15.9$	$115 \pm 12.1$	$96.7 \pm 0.1$	$39.2 \pm 2.3$
60	$278 \pm 3.5$	$269 \pm 3.7$	$52 \pm 5.8$	$96.9 \pm 0.1$	$19.1 \pm 2.0$
80	$236 \pm 4.0$	$228 \pm 4.0$	41 ± 2·4	$96.8 \pm 0.2$	$17.8 \pm 0.8$

Each value represents the average performance of a minimum of 3 individuals.

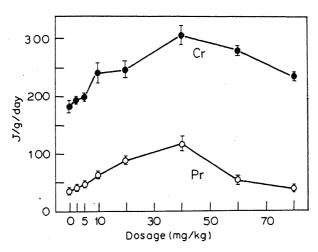


Figure 1. Consumption rate and production rate of *H. fossilis* treated with different doses of  $17\alpha$ -methyltestosterone (vertical line represents standard deviation).

significant influence on absorption efficiency (Nirmala and Pandian 1983). Absorption rate of the individuals receiving different doses of the tested hormone was dependent on feeding rate. Individuals displaying high feeding rate absorbed the food at a faster rate. Highest absorption rate was observed in the 40 mg/kg group (293 J/g/day).

#### 3.3 Conversion

Individuals receiving 40 mg/kg dose converted the food at the rate of  $115 \,\mathrm{J/g/day}$ , compared to  $38 \,\mathrm{J/g/day}$  in the control group ( $P < 0.05 \,\mathrm{significant}$ ). Thus the hormone (40 mg/kg) treated group displayed a three-fold increase in growth over control. In groups treated with 60 or  $80 \,\mathrm{mg/kg}$ , the rate decreased to  $52 \,\mathrm{and} \,41 \,\mathrm{J/g/day}$ , respectively. Briefly, the steroid enhanced growth up to an optimum dose ( $40 \,\mathrm{mg/kg}$ ), after which higher doses showed decline in conversion rate. Figure 1 shows that as the feeding rate increases, the production rate also increases. Besides, there is a positive

parallel trend between the dose on the one hand, and feeding and conversion rates on the other.

## 3.4 Conversion efficiency

 $17\alpha$ -methyltestosterone is not only an appetite stimulant but also boosts growth efficiency. Administration of 40 mg  $17\alpha$ -methyltestosterone/kg significantly (P < 0.05) enhanced the efficiency to 39%. But the efficiency decreased in the group treated with 60 or 80 mg/kg and was even lower (19%) than that of the control (21%, figure 1) showing that above an optimal dosage (> 40 mg/kg)  $17\alpha$ -methyltestosterone functions as an anti-anabolic agent. Similar result was reported for *Channa striatus* which was treated with a range (0 to 30 mg/kg) of  $17\alpha$ -methyltestosterone (Nirmala and Pandian 1983).

#### 4. Discussion

The objectives of applying anabolic steroids in fish culture practices are (i) to reduce feed-cost through increased growth efficiency and (ii) to reduce the production time by expediting feeding and growth rates. Application of  $17\alpha$ -methyltestosterone induced appetite and enhanced food consumption in H. fossilis; that  $17\alpha$ -methyltestosterone acts as appetite stimulant in fishes, has been demonstrated in Carassius auratus (Yamazaki 1976), Oncorhynchus kisutch (Fagerlund and McBride 1975a,b; McBride and Fagerlund 1973, 1976), Epinephelus salmoides (Chua and Teng 1980) and in Channa striatus (Nirmala and Pandian 1983). In O. kisutch only a single dose was tested by supplementing it with diet; C. striatus was injected with different doses and it was reported that the group receiving 20 mg/kg displayed maximum appetite. Therefore it may be concluded that  $17\alpha$ -methyltestosterone is an appetite stimulant and the magnitude of its stimulating ability is realized at different doses in fish. Growth rate of H. fossilis is dependent on feeding rate. Hence application of  $17\alpha$ -methyltestosterone considerably reduces the time cost of production by expediting feeding and growth rates.

Application of  $17\alpha$ -methyltestosterone not only increases the growth rate but also enhances the growth efficiency in H. fossilis (figure 2). The increase in the efficiency was from 21% in the control to 28 or 39% in the 10 or 40 mg/kg receiving groups. Fagerlund and McBride (1979) reported a 5% increase in efficiency of O. kisutch which received 10 mg of hormone/kg fish. It appears that had they tried a higher dose, O. kisutch could have doubled its efficiency. Nirmala and Pandian (1983) observed that C. striatus displayed the highest efficiency, when treated with 20 mg/kg (figure 1). Chua and Teng (1980) supplemented the feed with different doses of  $17\alpha$ -methyltestosterone (1 to 12 mg/kg food) to Epinephelus salmoides, reared in floating cages in a tropical estuary and found that the fish showed maximum efficiency, when the hormone was supplemented at the rate of 9 mg/kg feed (= 194 mg/kg fish). Hence  $17\alpha$ -methyltestosterone may be identified as an anabolic steroid for fishes. The negative growth efficiency observed in Lebistes reticulatus by Clemens et al (1966) can be traced to sexual dimorphism exhibited by the fish; in this species, females grow larger than males and when  $17\alpha$ -methyltestosterone, an androgenic hormone, was injected into a

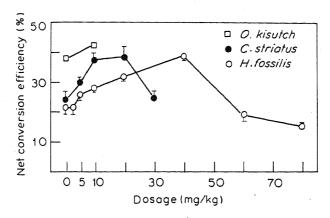


Figure 2. Conversion efficiency of H. fossilis treated with different doses of  $17\alpha$ -methyltestosterone (vertical line represents standard deviation. For comparison, data reported by Nirmala and Pandian (1983) for C. striatus and Fagerlund et al (1979) for O. kisutch are presented).

female, the female exhibited negative growth or 'masculinizing effect'. Donaldson et al (1979) indicated that at higher doses, some steroids exert deleterious effects on various organs and cumulatively cancel the growth promoting effects. This may be the reason for the decline in almost all parameters of H. fossilis receiving higher doses (> 40 mg/kg). In general, sexual dimorphism is not very apparent in most commercial fishes. Hence in the young ones,  $17\alpha$ -methyltestosterone may serve as an appetite stimulant and also as an anabolic steroid.

## Acknowledgement

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