

## EFFECT OF TESTOSTERONE PROPIONATE IN ENHANCING GROWTH IN *MESONOEMACHEILUS TRIANGULARIS* (DAY, 1865)

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**Abstract:** Studies on indigenous ornamental fishes of Kerala were centered mainly on the survey of the various river systems, studies on the export potential of the available fishes and the development of the breeding technologies of certain fishes. No detailed study has been carried on the feeding and growth of these fishes under culture conditions. Hence the present study was carried out to study in detail the effect of anabolic steroid testosterone propionate (TP) in enhancing growth in *Mesonoemacheilus triangularis* under lab conditions. Individuals of *M. triangularis* of average size  $1.31 \pm 0.31$ g were selected for the experiment. *M. triangularis* were collected from River Kallar, Thiruvananthapuram, Kerala. The specimens were allowed to acclimatize prior to stocking in the experimental tanks of size 90 cm x 45 cm x 45 cm. Water quality parameters were examined periodically and was maintained under tolerable limits. The feeds required for the study were formulated by 'Square method' according to Hardy (1980) at 40% protein level. The biochemical composition of the ingredients were analysed prior to the formulation of feed. Testosterone propionate at 1 ppm, 2 ppm, 3 ppm and 4 ppm were prepared for the study. Both short term experiments and long term experiments were conducted. Each treatment had a triplicate. Feed was given at the rate of 5% body weight. Fishes were taken out from each experimental tank fortnightly and their length and weight were recorded. The gross body compositions of the experimental fishes were also analysed. The results of the study were subjected to one way analysis of variation (ANOVA) using statistical package SPSS version 10.1. In *M. triangularis* 2 ppm showed the best values in percentage gain in length, weight and SGR followed by 3 ppm, 1 ppm, 4 ppm and control. The feed utilization studies showed that the FCR value was highest in the control group in the fishes. The FCE values, FAE values, PER and protein digestibility values were highest in 2 ppm in *M. triangularis*. There was no significant variation in the proximate body composition of the various treatment groups. The present observations show that the administration of hormones through feed increases growth rate and thus reducing the period of growth and cost of production which may be highly advantageous from commercial aquarist's point of view.

**Key words:** Indigenous ornamental fish, Testosterone, Feed, Growth.

## INTRODUCTION

Studies on indigenous ornamental fishes are concentrated mainly on the breeding aspects and surveys on their availability in the water bodies. There is a limit for the maximum growth which can be obtained by giving optimum quantity of nutrients. If the fish has to be grown beyond physiological limits, it has to be either genetically manipulated or should be administered hormones or growth promoters (Keshavanath, 2005). The androgen 17 $\alpha$ -methyl testosterone (17 $\alpha$ -MT) has been extensively experimented to promote growth in several

species of fishes. James and Sampath (2006) recommended the optimum dosage of 10 mg/Kg of MT attaining higher Specific growth rate in *Xiphophorous helleri* and *Betta splendens*. The present study could help to arrive at the optimum dosage of testosterone propionate required to bring about maximum growth in *N Mesonoemacheilus triangularis*. This knowledge would be useful for aquarist as the attainment of maximum size of the fish at the minimum time is important for getting more market value for the ornamental fish.

## MATERIALS AND METHODS

Juveniles of *Mesonoemacheilus triangularis* of average size  $1.31 \pm 0.31$  g were collected from River Kallar, a hill stream near Ponmudi hills in Trivandrum, Kerala and were acclimatized for a week and then introduced into experimental tanks of size 90 cm x 45 cm x 45 cm supplied with aged tap water and provided with a biological filter. Both long term experiments and short term experiments were conducted and a triplicate was maintained for each treatment. For long term experiments fishes were stocked at the rate of 12 fishes per tank. The experiment was conducted for 75 days. Fishes were fed 5% body weight twice daily at 10.00 and 16.00 hrs. Half the water was exchanged weekly and water quality parameters were monitored and estimated throughout the experimental period. Fishes were taken out from each experimental tank fortnightly and their lengths and weights were recorded during the study period. The gross body or carcass composition of the control and experimental fish were analysed after 75 days of experimentation. Five fish from each tank were collected, killed and weighed. They were minced properly dried and stored in airtight vials. Total protein, lipid, moisture, ash and fibre of the samples were determined based on the methodology followed for the proximate composition of feed ingredients. The glycogen content was estimated by anthrone method as detailed by Oser (1965). In the case of short term experiment the quantity of feed was fixed at the rate of 10% dry weight per mean individual body weight. Each time the feed was left in the tank for 6 hours. The unconsumed feed was siphoned out and dried in the oven at 60°C and weighed. Faecal matter was siphoned out before the next feeding. The faecal matter thus collected was sieved through blotting silk and residue weighed after drying in the oven at 60°C. The dried faecal matter was then pooled together in containers and stored in the desiccator until further analysis. The quantity of feed was reassessed at an interval of 15 days according to the weight of the fish. After 30 days the final weight of all the fish was recorded. The calculations were done based on the formula of Kesavanath *et al.* (1991).

## Experimental Diets

Feed ingredients: Dried anchovy (fish meal), groundnut oil cake (GOC), tapioca powder and rice bran (RB), were used for the preparation of the feed. All the ingredients were dried, powdered and sieved thoroughly into fine powder. Vitamins and mineral premix were included as common to the feed. The protein content of the experimental feed was fixed around 40% protein level and formulated according to the method of Hardy (1980). Testosterone propionate (TP) under the trade name Testosterone of Infar India Limited at 1 ppm, 2 ppm, 3 ppm and 4 ppm were prepared for the study. Appropriate quantity of basal diet ingredients and vitamin-mineral were added. A control feed was also prepared out of tapioca flour, rice bran, groundnut oil cake and fish meal along with vitamin mineral mix. The ingredients were hand kneaded with sufficient quantity of water and made into soft dough. The dough was then steam cooked and cooled before adding vitamin minerals. The final product was pelletised with a pelletiser. The pellets thus obtained were dried in an oven below 40°C until it became crisp. The dried pellets were broken into small pieces and stored separately in air tight containers.

## Analytical methods

Total nitrogen was estimated by microkjeldahl method and the protein was computed by multiplying the nitrogen value with the factor 6.25 based on Belcher and Godbert (1954). For carbohydrate estimation, phenol-sulphuric acid method by Dubois *et al.* (1956) was followed. Total lipid estimation was done by the method of Bligh and Dyer (1959). Gross energy (KJ g<sup>-1</sup>) was calculated using the energy factors of 23.65 KJ g<sup>-1</sup>, 17.17 KJ g<sup>-1</sup> for protein, carbohydrate and lipid respectively as per Jia *et al.* (1991). The moisture content was determined by keeping the samples in hot air oven at 95°C to 100°C for 30 minutes and then dried at 60°C till a constant weight was obtained. The crude fibre content in the samples was determined by acid base digestion and ash content by burning the sample at  $550 \pm 20^\circ\text{C}$  for 6 hrs in a muffle furnace based on A.O.A.C (2000).

## Statistical analysis

Data from each replicate with glass tank as a unit were analyzed for comparison among different dietary treatments. The results of the study were subjected to one way analysis of variation (ANOVA) using statistical package SPSS version 10.1, SPSS. Inc. Chicago, U.S.A, 1999. The results were compared using Duncans multiple range test at  $p < 0.05$  significance level.

## RESULTS AND DISCUSSION

Mean weight of *Mesonoemacheilus triangularis* fed on control and experimental diets during different time intervals of experimental duration showed that the percentage gain in weight for the different treatments were found to be varying with the highest percentage gain in 2 ppm (16.074%) and the lowest percentage gain in control (5.436%). The second, third and fourth highest values were attained by 3 ppm (12.975%), 1 ppm (10.000%) and 4 ppm (8.333%) groups respectively. The results of the same are represented in Table 1. There was significant ( $p < 0.01$ ) difference in the percentage increase in weight among the different groups. No significant difference in percentage gain in weight was observed between control and 4 ppm. The SGR (%) values were found to be varying between 0.031% (control) to 0.086% (2 ppm). There was significant ( $p < 0.01$ ) difference in the SGR (%)

between the different groups. No significant difference in SGR (%) was observed between control and 4 ppm. The proximate composition of the fish muscle showed that there was no significant variation in any of the parameters examined between the different groups.

Feed utilization studies of *M. triangularis* fed on control and experimental diets for 30 days are given in Table 2. There was a statistically significant ( $p < 0.01$ ) difference in the production, consumption, FCR, FCE, assimilation and SGR values between the different treatments. There was significant ( $p < 0.05$ ) difference in faecal remains, PER and protein digestibility between the different groups. There was no significant difference in the FAE between the different groups.

In the experiments in *M. triangularis* with testosterone there was observed an increase, peak and then a decline in length, weight and SGR with increasing levels of hormone. The experiment in *M. triangularis* showed maximum growth for 2 ppm followed by 3 ppm, 1 ppm and 4 ppm levels. Growth reduction in *Xiphophorus helleri* and *Betta splendens* at higher doses of 17 -MT due to the catabolic action of hormones was observed by James and Sampath, 2006. The FCE values, FAE values, PER and protein digestibility values were highest in 2 ppm group in *M. triangularis*. Felix *et al.* (1990), in their experiments with black molly found significant

**Table 1.** Growth parameters of *N. triangularis* fed with different levels of TP for 75 days

Parameters	F ratio	Treatments				
		Control	1 ppm	2 ppm	3 ppm	4 ppm
Initial length (cm)		3.271 ± 0.214	3.571 ± 0.269	4.100 ± 0.597	4.240 ± 0.310	5.090 ± 0.240
Final length (cm)		3.329 ± 0.180	3.900 ± 0.289	4.600 ± 0.589	4.640 ± 0.320	5.490 ± 0.230
Percentage gain in length (%)	** 323.634	a 0.122 ± 0.006	c 9.213 ± 0.461	d 12.195 ± 0.610	c 9.434 ± 0.472	b 7.859 ± 0.393
Initial weight (g)		1.486 ± 0.227	1.247 ± 0.113	1.686 ± 0.267	1.657 ± 0.251	1.200 ± 0.350
Final weight (g)		1.571 ± 0.243	1.372 ± 0.125	1.957 ± 0.315	1.872 ± 0.229	1.300 ± 0.330
Percentage gain in weight (%)	** 110.684	a 5.436 ± 0.387	b 10.000 ± 0.500	d 16.074 ± 0.804	c 12.975 ± 0.649	a 8.333 ± 0.417
SGR (%)	** 89.604	a 0.031 ± 0.002	b 0.055 ± 0.003	d 0.086 ± 0.004	c 0.071 ± 0.004	a 0.046 ± 0.002

a, b, c, d- Means with the same superscript do not differ significantly (Duncan's multiple range test) \*\*  $p < 0.01$

**Table 2.** Feed utilization studies of *N. triangularis* fed with different levels of TP

Parameters	F ratio	Control	1 ppm	2 ppm	3 ppm	4 ppm
Initial weight (g)		0.680 ± 0.135	0.720 ± 0.057	0.690 ± 0.130	0.710 ± 0.082	0.700 ± 0.274
Final weight (g)		0.868 ± 0.040	0.946 ± 0.050	0.976 ± 0.050	0.948 ± 0.050	0.906 ± 0.050
Production (g)	** 31.139 **	a 0.188 ± 0.010	bc 0.226 ± 0.010	d 0.286 ± 0.010	c 0.238 ± 0.010	ab 0.206 ± 0.010
Consumption (g)	6.774 *	a 4.480 ± 0.220	a 4.920 ± 0.250	b 5.390 ± 0.270	a 4.810 ± 0.240	a 4.540 ± 0.230
Faecal output (g)	2.729 **	a 0.530 ± 0.030	ab 0.570 ± 0.030	b 0.600 ± 0.030	ab 0.580 ± 0.030	ab 0.550 ± 0.030
FCR	9.408 **	a 23.830 ± 1.190	ab 21.770 ± 1.090	c 18.846 ± 0.940	bc 20.210 ± 1.010	ab 22.039 ± 1.100
FCE (%)	9.643 **	a 4.196 ± 0.210	d 4.593 ± 0.230	e 5.306 ± 0.270	c 4.948 ± 0.250	b 4.530 ± 0.230
Assimilation (g)	754.058	3.950 ± 0.020	4.350 ± 0.020	4.790 ± 0.020	4.230 ± 0.020	3.990 ± 0.020
FAE (%)	NS 0.025 **	88.170 ± 4.410	88.415 ± 4.420	88.868 ± 4.440	87.942 ± 4.400	87.885 ± 4.390
SGR (%)	21.424 **	a 0.353 ± 0.020	b 0.395 ± 0.020	d 0.502 ± 0.030	b 0.418 ± 0.020	c 0.457 ± 0.020
PER	9.931 *	a 0.105 ± 0.010	ab 0.115 ± 0.010	b 0.133 ± 0.010	bc 0.124 ± 0.010	ab 0.113 ± 0.010
Protein digestibility (%)	2.329	a 81.300 ± 2.350	ab 85.610 ± 2.510	b 90.530 ± 2.810	ab 87.110 ± 2.360	a 82.150 ± 1.130

a, b, c, d– Means with the same superscript do not differ significantly (Duncan's multiple range test) \*\*p<0.01; \*p<0.05, NS– Non-significant

influence of 17 -MT on the feeding rate and food conversion efficiency. They observed a maximum growth at 20 ppm followed by 25 ppm and at 50 ppm the growth performance was the poorest. The appetizing and anabolic effect of hormone incorporated feed as recognised by several authors is proved in the present experiment. The present experiment shows that the oral administration of TP is effective in enhancing growth of *M. triangularis*. The higher growth of fish can be correlated with better assimilation of diet and high conversion efficiency associated with hormone incorporation. Level of hormone is the only variable in the feeds used in the present study. The present observations show that the administration of hormones through feed increases growth rate and thus reducing the period of growth and cost of production which may be highly advantageous from commercial aquarist's point of view.

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