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EFFECTS OF ANDROSTENEDIONE, A PHYTOANDROGEN, ON GROWTH AND BODY COMPOSITION IN THE AFRICAN CATFISH *CLARIAS GARIEPINUS*

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Abstract

The present study investigated the effects of a phytoandrogen, androstenedione, on growth, body composition, and survival in African catfish, *Clarias gariepinus*. Three concentrations (25, 50, and 75 mg/kg feed) were administered for 120 days. Survival of treated groups did not significantly differ from the control and ranged 94.44-97.78%. A significantly higher weight gain was obtained in the 50 and 75 mg/kg diets. The fish fed these diets also had significantly better food conversion and protein efficiency ratios. Apparent net protein utilization was better in treated groups than in the control. The protein and lipid contents of the carcass of treated fish were significantly higher than in the control, with the highest contents in the 50 mg/kg treatment. The highest ash content was in the 25 mg/kg treatment. In overall parameters, 50 mg androstenedione per kg diet was the optimal concentration.

Introduction

Fish growth can be stimulated by hormones. Thyroid hormones, androgens as well as growth hormones encourage growth (Donaldson et al., 1979; Higgs et al., 1982). Improving fish growth by hormone treatment has been studied in several teleost species (Pandian and Sheela, 1995; Piferrer, 2001). The amount of hormone consumed by each fish is miniscule and most is eliminated rapidly (Goudie et al., 1986; Cravedi et al., 1989). Although consumption of hormone treated fish creates no health problems (Tave, 1992), marketing difficulties prompt investigators to seek alternate applications.

Phytoandrogens are extracts from naturally occurring phytochemicals (diosgenin and

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sitosterol) that provide for the effective application of steroid hormone precursors. Phytoandrogens enhance sex drive and function and increase muscle mass, strength, and tolerance to stress (Pinker Nell and Jing, 1994; Barrett-Connor et al., 1999). The testosterone precursors – androstenedione and 4-androstenediol – enhance testosterone production because they are one small biochemical step away from testosterone (Street et al., 1996) and help elevate testosterone through anabolic steroid effects. Phytoandrogens are alternatives to hormone replacement therapy and can safely be used to restore healthy testosterone levels in men (Street et al., 1996). The effects of phytoandrogen range from increased muscle mass and strength and stronger bones to effects on the prostate in aging male (Tan and Culberson, 2003).

The positive effects of phytoandrogens on human growth led researchers to examine possible benefits in fish culture. The catfish *Clarias gariepinus* (Burchell, 1822), with an almost Pan-African distribution, also occurs in Asia Minor, Jordan, Israel, Syria, and Turkey (Yalcin, 2002). In Turkey, it is wide spread in southern and central Anatolian fresh waters such as the River Asi, where it is of commercial importance, and the Ceyhan, Seyhan, Goksu, Aksu and Sakarya Rivers (Yalcin, 2002). Androstenedione is a steroid hormone that occurs naturally in pollen from certain pine trees and European cactuses. The present study investigated the effects of androstenedione as a phytoandrogen on the growth, body composition, and survival of African catfish.

Materials and Methods

Fish were from the Mustafa Kemal University Fisheries Research Unit. Full-sibling fish with an average body weight of 41.70±1.14 g and total length of 18.2±1.02 cm were randomly stocked into 100-l aquaria at a density of 10 fish per aquarium and fed a commercial trout diet. Throughout the experiment, the aquaria were provided compressed air and continuously flowing water (2 l/min), controlled at 25±1°C. The photoperiod was maintained on a 12-h light:12-h dark schedule. Dissolved oxygen, measured once a week with a digital oxygen analyzer (YSI 5750), varied little and remained between 5.92 and 6.71 mg/l. Water pH was periodically monitored and stable with minor changes (7.8-8.0).

Fish were fed trout diets (Aquamaks, Turkey; 48% protein and 18% lipid on a wet basis). Androstenedione powder was supplied by Vitalabs, Inc. (Georgia, USA). Three androstenedione doses (25, 50, and 75 mg/kg diet) were tested. The powder was mixed into pulverized trout diet and distilled water (450 ml/kg). The control diet contained no androstenedione. The mixtures were extruded through a food grinder with a 2-mm die (Lee et al., 2004). The extrusions were broken into small pieces and stored in a freezer until a few hours before feeding. Experimental diets were analyzed for proximate composition according to methods of AOAC (1990).

The test diets were randomly assigned to triplicate groups of fish fed at approximately 3% of their body weight per day twice daily for 120 days. Fish were counted and weighed individually at monthly intervals after anesthetization with 300 mg/l lidocaine-HCL per 1000 mg/l $NaHCO_3$ (Park et al., 1988).

Weight gain and survival (Watanabe et al., 1990), food conversion and protein efficiency ratios (Steffens, 1989), specific growth rate (Clark et al., 1990), apparent net protein utilization (Bender and Miller, 1953), and hepatosomatic index (Pfeffer et al., 1991) were calculated (see footnotes to Table 1).

At the start of the experiment, five fish from each group (total 20) were treated with an overdose of lidocaine-HCL in a $NaHCO_3$ solution, and stored at -20°C for determination of proximate body composition. At the end of the feeding trial, five fish from each treated group (15 fish per treatment) were analyzed according to the methods used to analyze the experimental diets (AOAC, 1990).

One way ANOVA was used to analyze differences between groups, and Duncan test was used to analyze which groups caused the difference (Norusis, 1993).

Results

There were no significant differences in survival between groups (Table 1). Analysis of variance showed that the weight gain and specific growth rate were significantly higher in fish fed the 50 and 75 mg/kg diets than in the control and 25 mg/kg groups. The food conversion ratio, protein efficiency ratio, and apparent net protein utilization were also significantly better in the 50 and 75 mg/kg groups than in the control.

The chemical composition of the fish is given in Table 2. The moisture content was the same in all the groups. The protein, lipid, and ash contents of the treated fish were significantly higher than in the control. There were no significant differences in hepatosomatic index.

Discussion

Androgens are effective growth promoters in channel catfish (*Ictalurus punctatus*; Gannam and Lovell, 1991), bagrid catfish (*Pseudobagrus fulvidraco*; Park et al., 2003), and common carp (*Cyprinus carpio*; Pandian and Sheela, 1995). Our work examined a natural alternative to synthetic growth hormones in fish culture. The study revealed that androstenedione-based diets improved growth and increased the level of protein and fat in African catfish. Beneficial effects of phytoandrogens in humans were described by Pinker Nell and Jing (1994), Barrett-Connor et al. (1999), and Tan and Culberson (2003). This is the first report to our knowledge regarding the potential of andro-stenedione as a growth-promoting agent in fish culture.

The survival rate of treated groups did not differ from the survival rate of the control. Treatments involving synthetic steroids result in higher mortality in most species (Pandian and Sheela, 1995). Nagy et al. (1981), Mair et al. (1987), and Kim et al. (1997) showed that augmenting the dose and timing of 17 α-MT lowered the survival rate. The present study suggests that androstenedione may be used in growth studies of African catfish.

Table 1. Effects of different concentrations of dietary androstenedione on survival, growth performance, and feed utilization efficiencies of African catfish *Clarias gariepinus* after 120 days (means±SE of triplicate groups).

Dose (mg\kg)	*Wt gain (%)*	*SGR (%/day)*	*FCR*	*PER*	*ANPU (%)*	*Survival (%)*
0	258.24±5.18^{a}	1.06±0.01^{a}	2.48±0.01^{b}	0.84±0.006^{a}	21.96±0.01^{a}	95.56±1.11^{a}
25	260.57±7.09^{a}	1.07±0.01^{a}	2.45±0.02^{b}	0.85±0.008^{a}	24.71±0.01^{b}	94.44±1.11^{a}
50	293.30±1.80^{b}	1.14±0.03^{b}	2.25±0.08^{a}	0.92±0.003^{b}	25.90±0.03^{c}	97.78±2.11^{a}
75	296.24±3.90^{b}	1.14±0.03^{b}	2.24±0.02^{a}	0.93±0.008^{b}	24.72±0.05^{b}	94.44±1.14^{a}

Values in a column with different superscripts significantly differ ($p<0.05$).
Wt gain = [(final wt - initial wt)/initial wt] x 100
SGR = Specific growth rate = $[(\ln_{W2} - \ln_{W1})\backslash(T2 - T1)] \times 100$, where W1 and W2 are mean body weights of the first and second samples and T1 and T2 indicate the days on which the samples were taken
FCR = food conversion ratio = dry feed intake/wet wt gain
PER = protein efficiency ratio = wt gain/protein intake
ANPU = apparent net protein utilization = (protein retained/protein intake) x 100

Table 2. The effect of different concentrations of dietary androstenedione on the chemical composition (wet basis) and hepatosomatic index (HSI) of African catfish *Clarias gariepinus* (means±SEM of triplicate groups).

Dose (mg\kg)	*Chemical composition (%)*				
	Moisture	*Crude protein*	*Crude lipid*	*Ash*	*HSI (%)*
Initial	75.45±0.25	17.83±0.14	2.50±0.02	1.1±0.03	-
0	72.84±0.19[a]	18.18±0.09[a]	2.65±0.05[a]	1.23±0.01[a]	1.22±0.03[a]
25	72.65±0.22[a]	18.62±0.06[b]	3.79±0.03[b]	1.38±0.01[d]	1.21±0.05[a]
50	72.14±0.19[a]	21.04±0.04[c]	5.17±0.01[d]	1.28±0.01[b]	1.22±0.06[a]
75	72.88±0.17[a]	20.88±0.04[c]	4.78±0.08[c]	1.32±0.01[c]	1.27±0.03[a]

Means in a column with different superscripts differ significantly ($p<0.05$).
HSI = (liver wet wt/total body wet wt) x 100

Analysis of variance showed that the growth rate at the end of the experiment was significantly higher in treated fish. The best growth rate, food conversion ratio, and protein efficiency ratio were obtained in the 50 and 75 mg/kg groups. Androstenedione increased the protein and lipid levels in the fish, although Gannam and Lovell (1991) reported that androgens reduce fat deposition in channel catfish.

Some synthetic steroids are effective growth-promoting agents when administered at low doses. On the other hand, increasing mortality and decreasing growth are characteristic of treatment with increasing doses of estrogen and androgen, although growth promotion after such treatments has occasionally been reported (Pandian and Sheela, 1995; Piferrer, 2001). In our study, androstenedione had positive effects on African catfish. Due to these reasons, the natural hormone androstenedione may be used as an alternate growth-promoting agent for African catfish. The 50 mg/kg diet proved optimal for the tested parameters. Further investigations can be carried out to study the effects of androstenedione on other culturable fish species.

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