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Effects of Dietary Androstenedione Concentration on Growth of Tilapia Fry (Oreochromis aureus Linnaeus)

Funda Turan1 *, Mevlut Gurlek1 and Cemal Turan2
1Department of Aquaculture, Faculty of Fisheries and Aquaculture, University of Mustafa Kemal, 31040 Antakya, Hatay, Turkey
2Fisheries Genetics Laboratory, Faculty of Fisheries and Aquaculture, University of Mustafa Kemal, 31040 Antakya, Hatay, Turkey

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Key words: tilapia, Oreochromis aureus, androstenedione, steroid hormones, growth

Abstract
The effects of androstenedione on the growth, body composition, and survival of tilapia (Oreochromis aureus) fry were examined. Diets were supplemented by one of three androstenedione concentrations (50, 100, or 200 mg/kg) for twelve weeks. The growth rate significantly increased in fish fed 50 mg androstenedione per kg compared to the control (p<0.01) but decreased dramatically at concentrations beyond 50 mg/kg. The specific growth rate, protein efficiency ratio, and food conversion ratio were significantly better in the 50 mg/kg group than in the other groups. Crude protein and survival at all androstenedione levels did not significantly differ from those of the control but lipid content dropped with 100 mg/kg supplementation.

Introduction
Tilapia are the second most important group of food fishes in the world, after carps. The increasing world-wide importance of cultured tilapia has generated considerable research on improvement of the growth performance in this fish. Steroid hormones can be effective growth enhancers in fish culture. High growth response to exogenous sex steroids was reported in several fish species including tilapia, Oreochromis niloticus (Donaldson et al., 1979; Higgs et al., 1982; Pandian and Sheela, 1995; Alam and Monwar, 1998; Turan and Akyurt, 2003, 2005a). While this technique is widely used, it has practical limitations. In addition, increasing consumer resistance to the use of hormones in food production further limit its general applicability. Thus, researchers are seeking alternatives.

Androstenedione is an anabolic steroid precursor because it is converted to testosterone in the body. Androstenedione enhances sex drive and function and increases muscle mass, strength, and tolerance to stress (Phillips et al., 1994; Barrett-Connor et
Effects of dietary androstenedione on tilapia fry growth

Androstenedione, a natural substance in humans, animals, and pollen from many plants, is used as an alternative to hormone replacement therapy and can safely be used to restore healthy levels of testosterone in humans (Street et al., 1996). To our knowledge, there are no legal restrictions on the use of natural substances although there are international restrictions on the use of synthetic steroid hormones in comestible agricultural products (Ostrowski and Garling, 1988).

The positive effects of androstenedione in humans led us to examine possible benefits in fish culture. Turan and Akyurt (2005b) discovered that administration of dietary androstenedione significantly increased growth and improved protein and lipid levels in African catfish (*Clarias gariepinus*). The objective of the present work was to investigate the effects of dietary androstenedione on growth, body composition, and survival in the fry of another important cultured fish species, tilapia (*Oreochromis aureus*).

**Materials and Methods**

Tilapia (*Oreochromis aureus*) fry were obtained from the Mustafa Kemal University Aquaculture Research Unit and reared in a 200-l fiberglass tank. Fish were selected and randomly stocked into 50-l aquaria prior to the experiment. The stocking density was 15 fish (mean initial body weight 0.75±0.03 g) per aquarium. Aquaria were continuously aerated with an air stone and cleaned every afternoon by siphoning off accumulated waste materials. Approximately one-third of the water in each aquarium was replaced with aerated fresh water daily. Water temperature was maintained at 25-27°C and the photoperiod at 12 h light:12 h dark.

Three experimental diets containing three different dosages of androstenedione (50, 100, 200 mg/kg) were randomly assigned to triplicate groups of fish. To prepare the diets, androstenedione powder, supplied by Vitalabs Inc. (Georgia, USA), was mixed with a pulverized carp diet (28% protein, 12% lipid, 4% cellulose, 16% ash on wet basis; Akumak, Turkey) and water (450 ml/kg) and the mixture was extruded through a food grinder (Lee et al., 2004). The control diet was also mixed with 450 ml water. The extrusions were broken into small pieces and stored in a freezer until feeding. The fish were fed twice a day at 09:00 and 17:00 for twelve weeks. The daily feed allowance was 4% of the body weight per day, close to the maximum daily ration consumed by the juvenile tilapia during the acclimation stage.

Body weights were measured every two weeks after anesthetizing the fish with 300 mg/l lidocaine-HCL per 1000 mg/l NaHCO3 (Park et al., 1988). Weight gain, specific growth rate (SGR), food conversion ratio (FCR), protein efficiency ratio (PER), and survival rate were calculated as: wt gain = final wt - initial weight; SGR (in %) = 100[(lnWt2 - lnWt1)/(T2 - T1)]; where Wt1 and Wt2 are the mean body weights of the first and second samples (T1 and T2); FCR = dry feed intake/wet wt gain; PER = live body wt gain/protein intake; survival rate (in %) = 100[(initial no. of fish - no. of dead fish)/initial no. fish].

At the beginning of the experiment, five fish from each treatment (n = 20 fish) were killed by an overdose of lidocaine-HCL per 1000 mg/l NaHCO3 and stored at -20°C for determination of the proximate composition of the body. At the end of the feeding trial, five fish from each replicate (n = 15 fish per androstenedione level) were analyzed for whole body proximate composition according to standard methods (AOAC, 1990).

One-way analysis of variance was used to determine differences in growth and body composition among treatments. Duncan’s new multiple range test was used to evaluate differences between treatment means (Norusis, 1993).

**Results**

The effects of different concentrations of dietary androstenedione on growth and survival are given in Table 1. Growth, SGR, and PER were significantly better in fish fed 50 mg/kg than in the control (p<0.01). However, growth dramatically decreased when androstenedione was supplemented at more than 50 mg/kg. The initial and final chemical compositions of the fish are given in Table 2.
Moisture, protein, and ash contents did not vary among treatments ($p>0.05$). The lipid contents were also similar except in the 100 mg/kg treatment where lipid was dramatically lower.

**Discussion**

This study examined androstenedione-based diets as a natural alternative to synthetic growth hormones and resulted in improved growth in *O. aureus* fry. Turan and Akyurt (2005b) also obtained significantly higher growth in African catfish (*Clarias gariepinus*) fed androstenedione-based diets. In the present study, the growth rate dramatically decreased after 50 mg/kg supplementation, indicating that high concentrations (100-200 mg/kg) of androstenedione have a negative effect on growth in tilapia fry. Likewise, Pandian and Sheela (1995) and Piferrer (2001) reported the negative effect of high doses of synthetic steroids on fish growth. There is no clear explanation why such a phenomenon occurs. Therefore, further studies are needed to investigate the reason.

Table 1. The effects of different concentrations of dietary androstenedione on growth performance, feed utilization efficiency, and survival of tilapia, *Oreochromis aureus*, fry (means±SE of triplicate groups).

<table>
<thead>
<tr>
<th>Androstenedione (mg/kg feed)</th>
<th>0</th>
<th>50</th>
<th>100</th>
<th>200</th>
</tr>
</thead>
<tbody>
<tr>
<td>Initial body wt (g)</td>
<td>0.75±0.03&lt;sup&gt;a&lt;/sup&gt;</td>
<td>0.75±0.03&lt;sup&gt;a&lt;/sup&gt;</td>
<td>0.75±0.03&lt;sup&gt;a&lt;/sup&gt;</td>
<td>0.75±0.03&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>Final body wt (g)</td>
<td>12.02±0.89&lt;sup&gt;b&lt;/sup&gt;</td>
<td>15.00±0.47&lt;sup&gt;c&lt;/sup&gt;</td>
<td>8.53±0.50&lt;sup&gt;a&lt;/sup&gt;</td>
<td>8.43±0.89&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>Wt gain (g)</td>
<td>11.26±0.86&lt;sup&gt;b&lt;/sup&gt;</td>
<td>14.24±0.44&lt;sup&gt;c&lt;/sup&gt;</td>
<td>7.78±0.52&lt;sup&gt;a&lt;/sup&gt;</td>
<td>7.68±0.86&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>SGR</td>
<td>3.07±0.03&lt;sup&gt;b&lt;/sup&gt;</td>
<td>3.32±0.02&lt;sup&gt;c&lt;/sup&gt;</td>
<td>2.69±0.10&lt;sup&gt;a&lt;/sup&gt;</td>
<td>2.68±0.11&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>FCR</td>
<td>1.70±0.12&lt;sup&gt;b&lt;/sup&gt;</td>
<td>1.35±0.04&lt;sup&gt;b&lt;/sup&gt;</td>
<td>2.46±0.15&lt;sup&gt;a&lt;/sup&gt;</td>
<td>2.53±0.28&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>PER</td>
<td>2.12±0.16&lt;sup&gt;b&lt;/sup&gt;</td>
<td>2.68±0.08&lt;sup&gt;c&lt;/sup&gt;</td>
<td>1.46±0.09&lt;sup&gt;a&lt;/sup&gt;</td>
<td>1.45±0.16&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>Survival (%)</td>
<td>95.56±4.44&lt;sup&gt;a&lt;/sup&gt;</td>
<td>97.78±2.22&lt;sup&gt;a&lt;/sup&gt;</td>
<td>93.33±3.84&lt;sup&gt;a&lt;/sup&gt;</td>
<td>95.55±2.22&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
</tbody>
</table>

Values in a row with different superscripts significantly differ ($p<0.001$) except for final body weight where differences are significant if $p<0.01$.

Table 2. The effects of different concentrations of dietary androstenedione on the chemical composition of tilapia *Oreochromis aureus* fry (means±SE of triplicate groups).

<table>
<thead>
<tr>
<th>Composition ( % wet basis)</th>
<th>Initial</th>
<th>Androstenedione (mg/kg feed)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>0</td>
</tr>
<tr>
<td>Moisture</td>
<td>75.30±0.30</td>
<td>74.40±0.70&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>Crude protein</td>
<td>18.25±0.65</td>
<td>19.16±0.76&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>Crude lipid</td>
<td>7.28±0.42</td>
<td>8.94±0.30&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>Ash</td>
<td>0.98±0.01</td>
<td>0.99±0.01&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
</tbody>
</table>

Values in a row with different superscripts significantly differ ($p<0.001$).
The crude protein and ash contents were not significantly affected by androstenedione dosage but the lipid content in treated tilapia fry dramatically decreased after the 100 mg/kg supplementation level. This negative effect of androstenedione on lipid content is in accordance with the results of Gannam and Lovell (1991) who reported that androgens reduce fat deposition in channel catfish.

In conclusion, under our experimental conditions, inclusion of 50 mg androstenedione per kg feed improved feed utilization in tilapia fry, resulting in a higher growth rate. Therefore, androstenedione can be used as a natural alternative to synthetic hormones. Further, the effects of androstenedione administration on plasma metabolite and hormone concentrations in fish should be studied.

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