Dietary effects of extruded feed on biochemical and hematological indices of Rainbow trout 
(*Oncorhynchus mykiss*)

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Abstract
The effect of 0, 70, 80, and 100% levels of extruded feed on biochemical and hematological indices of rainbow trout (*Oncorhynchus mykiss*) was studied at 480 fries with an average weight of 90±5 g in the form of a completely randomized design. 16 fish were randomly collected from each treatment at the middle and end of the 90-s experiment. According to the findings, the highest amount of protein in the carcasses was related to fish fed with 85% extruded feed. Also, the protein content of carcass was significantly different from that of extruded food before consumption (p <0.05). The highest amount of carcass Ca and Mn was related to 85% extruded food. There was also a significant difference between Mn and Ca levels in carcasses before and after extruded feed (p <0.05). There was no significant difference in albumin levels during the treatments. In the mid-period measurement, a significant difference was observed between total blood protein of 100% pellet and 70% extruded treatment (p <0.05). The highest was observed in 5.2 g/dl of fed with 70% extruded feed and the lowest was 4.24g/dl in the group of fed with 70% pellet. In general, the results showed that the use of high levels of extruded could be replaced with pellet supplements in the diet of rainbow trout (*Oncorhynchus mykiss*) without any negative effect on blood indices.

Keywords: Albumin, Rainbow trout, Total protein, Extruded

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Introduction
Aquaculture is one of the ways of economic growth of developing countries, because it creates employment, production, currency and ultimately improves the living standards of the people of these countries (Pulatsu et.al., 2004). Despite the importance of nutrition in aquaculture, but most aquaculture breeders in Iran are not fully familiar with nutrition issues and thus providing quality food and reasonable prices will be one of the important challenges of aquaculture in the near future (Nafisi Bahabadi, 2001). A short breeding period, very good quality meat, very good production per unit area and a much higher price than other fish are other advantages of choosing rainbow trout (Oncorhynchus mykiss) for breeding (Dore, 1991).

Chaharmahal and Bakhtiari province has suitable water potentials and has 10% of Iran's water resources. The amount of trout production in Chaharmahal and Bakhtiari province has increased from 5337 kg in 2005 to 17154 tons in 2015. Also, the amount of rainbow trout production in the whole country has increased from 34760 to more than 140632 tons in the same time, which is a very significant figure. (Statistics of Jihad Agriculture Organization Chaharmahal and Bakhtiari Province, 2015).

One of the methods used when making fish feed is pelleting the feed. Commercial pellets can be prepared in three ways: a) Compressed and sinking pellets using pellet binders, b) Extruder or immersed pellets with porosity (sponge state), c) Expander with use steam and high pressure.

Feed production using extruders has a long history in the world. The production of large quantities of feed through extruders began in the late 1950s and early 1960s. Sources of raw materials and reduction of environmental degradation due to lack of feed in water sources and prevention of environmental pollution. The aim of this study was to evaluate the levels of some serum parameters such as total protein and albumin in trout fed with pellet feed and different extruded levels.

Materials and methods
In this experiment, 480 fry rainbow trout with an average initial weight of 90±5 g were used for 90 days in one of the rainbow trout farms in Chaharmahal and Bakhtiari province. Temperature, dissolved oxygen, pH, ammonia and water nitrite were arefully monitored before the experiment.

In order to evaluate the performance of serum indices of rainbow trout using pellet feed and different extruded levels, an experiment was conducted in a completely randomized design with 4 treatments and each treatment in 4 replications (each treatment of fish 70, 85 and 100% of their nutritional needs were extruded and one treatment received 100% of pellet feed).

Apparent digestibility
To evaluate the use of feed protein at the end of the period, 16 samples from...
each treatment and 4 fecal per replicate were taken and in the laboratory, the amount of protein remaining in the samples was determined by appropriate method (Nakano et al., 1991).

Measurement of total protein in serum
The method of experiment was performed by photometric method according to Biuret method according to the instructions of Pars Azmun kit (Thomas, 1995).

Measurement of albumin in the serum
The method of experiment was performed by Bромocреsol GREEN photometric method according to the instructions of Pars Azmun Company. In this experiment, serum albumin or Bромocреsol GREEN forms a complex at acidic pH (Thomas 1995; Johnson et al., 1999).

Bloodletting
To draw blood from a heparin-free syringe, it was inserted obliquely from behind the posterior edge of the anterior fin and 2 mL of blood was collected. Then a centrifuge with 3000 rpm was used for 5 minutes to separate serum from blood (Dati, 1996).

Statistical analyzes
The normality of the data was assessed using the Kolmogorov-Smirnov test. Statistical analysis of data was performed by one-way analysis of variance (One-way ANOVA) and data were compared by Duncan test at 5% level. All statistical analyzes were performed using SPSS statistical software version 18.

Results and discussion
Investigation of chemical properties of fish carcasses
Mean and standard error of some chemical properties of rainbow trout carcasses with different levels of extrusion compared to pellet feed are given in Table 1.

Table 1: Mean and standard error of some chemical properties of rainbow trout carcasses with different levels of extrusion compared to pellet feed.

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Observation No.</th>
<th>Protein (%)</th>
<th>Ca (mg/100 g)</th>
<th>P (mg/100 g)</th>
<th>Mn (mg/100 g)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Before of test</td>
<td>4</td>
<td>15.36&lt;sup&gt;b&lt;/sup&gt;</td>
<td>59.4&lt;sup&gt;b&lt;/sup&gt;</td>
<td>151.2&lt;sup&gt;a&lt;/sup&gt;</td>
<td>19.96&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
<tr>
<td>0%</td>
<td>4</td>
<td>18.5&lt;sup&gt;a&lt;/sup&gt;</td>
<td>66.76&lt;sup&gt;a&lt;/sup&gt;</td>
<td>5.134&lt;sup&gt;a&lt;/sup&gt;</td>
<td>22.1&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>85%</td>
<td>4</td>
<td>18.37&lt;sup&gt;a&lt;/sup&gt;</td>
<td>66.3&lt;sup&gt;a&lt;/sup&gt;</td>
<td>16.174&lt;sup&gt;a&lt;/sup&gt;</td>
<td>21.74&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>70%</td>
<td>4</td>
<td>18.8&lt;sup&gt;a&lt;/sup&gt;</td>
<td>67.87&lt;sup&gt;a&lt;/sup&gt;</td>
<td>44.173&lt;sup&gt;a&lt;/sup&gt;</td>
<td>22.89&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>Mean±SE</td>
<td>-</td>
<td>0.3</td>
<td>1.1</td>
<td>9.6</td>
<td>0.47</td>
</tr>
</tbody>
</table>

Different letters in each column indicate a significant difference in level.

Carcass protein
As Table 1 shows, the lowest carcass protein content in fish before the experiment was 15.36%, which with the carcass protein content after extruded feed at different levels (100, 85, 70%) and pellet feed consumption the difference was significant (p<0.05). But
the highest protein content in fish carcasses was 19.22%, which was related to fish fed 85% extruded, and the lowest was 18.5% related to pellets, but the difference between them was There was no statistically significant (P> 0.05) (Fig. 1).

Figure 1: Carcass protein (%) in different treatments of the experimental period.

Carcass calcium
The amount of calcium remaining in fish carcasses before the experiment was 59.4 mg/100 g. The amount of calcium in fish carcasses after the experiment in treatments with different levels of extrusion (100, 85, 70%) and pellet feed consumption was significantly different (p<0.05). The highest amount of calcium in fish carcasses in the amount of 67.87 mg% was related to fed fish or 85% extruded feed. There was no significant difference between calcium in fish carcasses in control treatment and different levels of extruded (100, 85, 70%) at the end of the period (Fig. 2).

Figure 2: Carcass calcium (mg/100 g) in different treatments of the experimental period.

Carcass magnesium
There was no significant difference between the amount of magnesium in carcasses of fish before the experiment with carcasses fed with 100% extruded feed and also fish fed with pellet feed and different levels of extruded (Fig. 3). The amount of magnesium before the start of the experiment was 19.96 mg%, which was significantly different from the amount of magnesium in the treatments fed with pellet feed and the levels of 85% and 70% of extrusion (p<0.05).

Figure 3: Carcass magnesium (mg/100 g) in different treatments of the experimental period.
Carcass phosphorus

No significant difference was observed between phosphorus content of fish carcasses before the start of the experiment and after the end of the period in the treatments fed with pellet feed and different extrusion levels (Fig.4).

Investigation of blood serum factors

serum albumin

As shown in Table 2, except at the beginning of the period, blood albumin levels during the experimental stages were not the same in different groups and no statistically significant difference was observed between them.

Table 2: Mean and standard error of rainbow trout blood albumin (g/dl) in different assays with different levels of extrusion compared to pellet feed.

<table>
<thead>
<tr>
<th>Treatment</th>
<th>No.</th>
<th>Beginning</th>
<th>Middle</th>
<th>Final</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>8</td>
<td>3.58a</td>
<td>3.24a</td>
<td>3.03a</td>
</tr>
<tr>
<td>100%</td>
<td>8</td>
<td>3.25ab</td>
<td>3.18a</td>
<td>3.05a</td>
</tr>
<tr>
<td>85%</td>
<td>8</td>
<td>3.17ab</td>
<td>3.14a</td>
<td>3.06a</td>
</tr>
<tr>
<td>70%</td>
<td>8</td>
<td>2.9b</td>
<td>3.27a</td>
<td>2.9a</td>
</tr>
<tr>
<td>Mean±SE</td>
<td>-</td>
<td>0.19</td>
<td>0.06</td>
<td>0.11</td>
</tr>
</tbody>
</table>

Different letters in each column indicate a significant difference in level.

Total blood protein

As shown in Table 3, the amount of total protein at the beginning and end of the experimental period was the same and there was no significant difference (p<0.05). The highest was observed in the amount of 5.2 g/dl in fish fed with 70% extruded feed and the lowest was observed in the amount of 4.24 in the group of fish fed with 70% of pellet feed.

Table 3: Mean and standard error of total blood protein (g/dl) of rainbow trout in different measurement periods using different levels of extrusion compared to pellet feed.

<table>
<thead>
<tr>
<th>Treatment</th>
<th>No.</th>
<th>Beginning</th>
<th>Middle</th>
<th>Final</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>8</td>
<td>4.2a</td>
<td>4.24ab</td>
<td>5.12a</td>
</tr>
<tr>
<td>100%</td>
<td>8</td>
<td>4.1a</td>
<td>4.48ab</td>
<td>4.84a</td>
</tr>
<tr>
<td>85%</td>
<td>8</td>
<td>3.70a</td>
<td>4.9ab</td>
<td>4.82a</td>
</tr>
<tr>
<td>70%</td>
<td>8</td>
<td>3.50a</td>
<td>5.2a</td>
<td>4.98a</td>
</tr>
<tr>
<td>Mean±SE</td>
<td>-</td>
<td>0.29</td>
<td>0.23</td>
<td>0.32</td>
</tr>
</tbody>
</table>

Different letters in each column indicate a significant difference in level.
Apparent digestibility
As can be seen in Table 4, no significant difference was observed between pellet-fed fish and 100% extruded. Also, 85% and 70% of the extruded group had no significant difference. A significant difference was observed between pellet and 100% extruded groups with the other two groups (p<0.05).

Table 4: Mean and standard error of Apparent digestibility protein remaining in rainbow trout feces with different extrusion levels compared to pellets.

<table>
<thead>
<tr>
<th>Treatment</th>
<th>No.</th>
<th>Protein (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>4</td>
<td>13.6</td>
</tr>
<tr>
<td>100%</td>
<td>4</td>
<td>14.9</td>
</tr>
<tr>
<td>85%</td>
<td>4</td>
<td>12.3</td>
</tr>
<tr>
<td>70%</td>
<td>4</td>
<td>11.5</td>
</tr>
<tr>
<td>Mean±SE</td>
<td>-</td>
<td>0.49</td>
</tr>
</tbody>
</table>

Different letters in each column indicate a significant difference in level.

Nutrition is one of the main pillars of breeding organisms, including aquatic organisms. If the quantitative and qualitative conditions of fish feed are not taken into account, fish metabolism will be problematic and in addition to reduced growth, fish will not be able to cope with adverse environmental conditions as well as disease. In addition, any problems in the fish feeding system will cause great economic losses. On the other hand, about 70% of the costs of fish farming are related to nutrition, so any savings in this area will increase the productivity of fish farming. The purpose of this study was to compare different levels of extruded feed with pellet feed.

Serum indices measured (albumin and total protein) in all measured treatments were in the normal range and pellet feed did not lead to a decrease in total protein and albumin levels. In the study, after chemical analysis of fish carcases in terms of protein, calcium, phosphorus and magnesium, it was observed that the amount of protein, calcium and phosphorus in carcases fed with different levels of extrusion and pellets was not significantly different. There was a significant difference (p<0.05).

References


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