EFFECT OF DIFFERENT RATIO OF ADDITION OF NaCl TO THE DIETS ON SOME GROWTH TRAITS IN GRASS CARP *CTENOPHARYNGODON IDELLA*

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Abstract
Grass carp at average weight of 21.5 + 1.5 g were fed for seventy days on four diets with 30% protein content containing different ratios of table salt (NaCl): 1, 3, 5 and 7% to study the effect of salty diet on the rate and efficiency of feed conversion and the amount of feed and protein intake. Results showed an improvement in the rate and efficiency of feed conversion and the amount of protein intake in the second treatments (3% NaCl) and the third (5% NaCl) compared to the first treatment (control 1% NaCl) and the fourth (7% NaCl). The feed conversion rates were 4.54, 2.68, 2.74 and 4.71, and the average of feed conversion efficiency were 22.02, 37.31, 36.50, 21.23%, and the protein intake rates were 19.32, 24.13, 23.78 and 18.84 g in salty diets 1, 3 and 5 and 7% NaCl for the first to fourth treatments respectively. In another experiment to study the effect of salty diets on the feed intake, fish at average weight of 35.12 + 1.66 g were fed on the same diets used in the growth experiment. Results indicated an increase in feed intake rates in the second and third treatments (3% and 5% salt), they were 4.45 and 4.18 g / 100 g fish respectively, and the decrease in the first and fourth treatments (1% and 7% salt) they reached 3.75 and 3.42 g feed / 100 g fish, respectively.

Key words: feed conversion rate and efficiency, feed intake, protein intake, grass carp.

Introduction
Fish were divided according to their salty tolerance to three types, the Euryhaline fish, the species that migrate between the freshwater and saltwater during their lifetime, which have the ability to tolerate a wide range of saline changes in the surrounding environment such as in some Salmonids, Mesohaline fish are species that tolerate medium range of salinity of the environment such as brackish water fish, while the Stenohaline fish include species that tolerate a narrow range of saline changes in their water ambient, and are trapped in their environment and unable to move to another environment that differs in its salt concentration from its original environment (Al-Khshali, 2015). Iono-osmoregulation is one of the most important methods of success of fish to overcome the saline stress caused by the fluctuation of saline concentrations and thus control the amount of water and salts concentration in the body to maintain the internal stability of homeostasis, and some organs are responsible for the regulation of osmotic pressure of body fluids in freshwater fish, thereby fish can cope with stresses and maintain their life in their environments. Most of freshwater fish are stenohaline fish because they do not tolerate high saline concentrations and suffer significant losses when transported to salt water due to the occurrence of the so-called Osmotic shock. However, recent studies have shown that this problem can be overcome and the salt tolerance of fish through the use of salty feeding technique, which enables the fish to adapt to live in an environment whose salinity is outside the range in which the particular species lives, although these fish (fresh or marine) have evolved to live and survive in specific conditions over the long yermin their original habitat (McCormick and Bradshaw, 2011). The effect of salinity on growth be attributed to its role in reducing fish appetite, feed conversion rates, and low feed consumption due to increased energy expenditure rates for iono-osmoregulation and decrease the energy assigned for growth (Al-Khshali,2017). The effect of salinity on fish growth varies depending on the age, species, feeding behavior, appetite, and other environmental factors, so it is difficult to determine an ideal saline concentration for the growth of a particular species of fish (Boeuf and Payan, 2014). The values of feed intake are determined according to the fish species and size, feeding habits, dietary composition and physiological status (Alsaadi and Al-Khshali, 2018). The aim of this study is to investigate the effect of introducing different proportions of table salt (NaCl) in diets on some growth and feeding characteristics of grass carp.
Materials and Methods

Preparation of Salty Diets

Four salty diets with 30% protein content were prepared in the laboratory, feed materials (Table 1) were imported from the local market. They were sampled and mixed with each other after adding NaCl by 1, 3, 5 and 7%, 400 ml of water was added to make a homogeneous paste and insert in a 3 mm slot machine to get wet and soft filaments dried on an airtight wooden table, then cut to pellets at 5 mm size.

Table 1: The percentage of feed materials used in the formation of four salty diets

<table>
<thead>
<tr>
<th>Feed Constituent</th>
<th>Diet 1 (control) 1% salt</th>
<th>Diet 2 3% salt</th>
<th>Diet 3 5% salt</th>
<th>Diet 4 7% salt</th>
</tr>
</thead>
<tbody>
<tr>
<td>Fish meal</td>
<td>20</td>
<td>20</td>
<td>20</td>
<td>20</td>
</tr>
<tr>
<td>Yeast</td>
<td>10</td>
<td>10</td>
<td>10</td>
<td>10</td>
</tr>
<tr>
<td>Soybean</td>
<td>35</td>
<td>35</td>
<td>35</td>
<td>35</td>
</tr>
<tr>
<td>Rough rice</td>
<td>10</td>
<td>10</td>
<td>10</td>
<td>10</td>
</tr>
<tr>
<td>Wheat bran</td>
<td>12</td>
<td>10</td>
<td>8</td>
<td>6</td>
</tr>
<tr>
<td>Yellow corn</td>
<td>9</td>
<td>9</td>
<td>9</td>
<td>9</td>
</tr>
<tr>
<td>min. &amp;vit.</td>
<td>1</td>
<td>1</td>
<td>1</td>
<td>1</td>
</tr>
<tr>
<td>Corn oil</td>
<td>2</td>
<td>2</td>
<td>2</td>
<td>2</td>
</tr>
<tr>
<td>Salt (NaCl)</td>
<td>7</td>
<td>5</td>
<td>3</td>
<td>1</td>
</tr>
</tbody>
</table>

Acclimation of Experimental Fish

150 of grass carp weighing ranged between 18-45 g were brought from fish farm, south of Baghdad to the lab in cork containers. Fish were distributed on 20 glass tanks filled with 60-liter of water chlorine-free and supplied with ventilation to provide oxygen. After 24 hours, fish were fed on the control diet (1% NaCl - Table 1) twice a day at 3% of body weight.

Growth experiment

Fish at average weight of 21.5 ± 1.5 g were distributed on 12 glass tanks (40 x 60 x 60 cm) with 6 fish /tank, and were fed on four diets contained different ratios of salt are 1, 3, 5 and 7%. Each diet represented independent treatment with three replicates. Fish were fed on diets at 3% of body weight for 10 weeks, and were weighed in each treatment every two weeks, the amount of feed provided to the fish was adjusted according to the new weight. Levels of dissolved oxygen, pH and water temperature were measured throughout the experiment. The following criteria were calculated according to Gerking (1971):

a. Feed Conversion Rate (FCR) = Weight of feed intake (g) / Wet weight increase of fish (g)

Below are the formulas for B and C:

B. Feed conversion efficiency (FCE) = \{\text{weight increase of fish (g)} / \text{weight of feed intake (g)}\} \times 100

C. Protein intake (PI)

Protein intake (g) = \{\text{feed intake} \times \text{ratio of protein in the diet}\} / 100

Determining the Feed Intake

Grass carp at average weight of 35.12 + 1.66 g were fed on four diets containing different ratios of NaCl (1, 3, 5 and 7%), all of which had 30% protein content. Fish were fed ad libitum (5% of body weight) during 14 days, experiment fish were distributed on eight glass tanks with two replicates as 6 fish / tank. Non-consumable feed was withdrawn to be collected, dried, weighed and subtracted from the amount of feed provided to fish to estimate the amount of food consumed per unit of feed/100 g fish. The concentration of dissolved oxygen, pH and water temperature were measured throughout the experiment.

Statistical Analysis

Completely Random Design (CRD) was used according to SAS (2004, SAS) for data analysis, and the significant differences between the means of treatments were compared according to Duncan test on probability level of 0.05.

Results and Discussion

Environmental Factors

Water temperature averages during the experiment ranged between 21-23°C, dissolved oxygen ranged between 5.3-5.7 mg / l, and the pH ranged from 7.4 to 7.6. These ranges are all within the safe levels of grass carp survival (Hattingh et al., 1975). Table 2 showed that the medium proportions of NaCl (3 and 5%) used in salty diets (second and third) had a significant effect (p <0.05) in raising the rate and efficiency of feed conversion, compared to the first treatment (control 1% salt) and the fourth (7% salt). Results of the statistical analysis showed that there was no significant differences between the second and third treatments, and between the first and fourth treatments. The second treatment exceeded the rate and the efficiency of the feed conversion followed by the third treatment.

Table 2: The rate and efficiency of feed conversion and protein intake in grass carp fed on different salty diets.

<table>
<thead>
<tr>
<th>Ratio of salt in diet %</th>
<th>Feed conversion rate</th>
<th>Feed conversion efficiency (%)</th>
<th>Protein intake(g)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>4.54 a</td>
<td>22.02 b</td>
<td>19.32 b</td>
</tr>
<tr>
<td>3</td>
<td>2.68 b</td>
<td>37.31 a</td>
<td>24.13 a</td>
</tr>
<tr>
<td>5</td>
<td>2.74 b</td>
<td>36.50 a</td>
<td>23.78 a</td>
</tr>
<tr>
<td>7</td>
<td>4.71 a</td>
<td>21.23 b</td>
<td>18.84 b</td>
</tr>
</tbody>
</table>
Results showed that the effect of salty feeding was positive in the protein intake for all the treatments throughout the experiment. The highest levels of protein intake were in the second treatment followed by the third, then the first and the fourth. Results of the statistical analysis showed significant differences (p <0.05) between the second and third treatments on the one hand, and between the first and fourth treatments on the other hand. The medium percentages of NaCl (3 and 5%) used in the formation of diets had a positive effect on improving the rate and efficiency of feed conversion and protein intake. This can be attributed to the internal stability of fish, and the ionic and water balance occurred between internal environment (fish body) and external environment (water ambient), as well as regularity of metabolic rates in fish, where the energy taken from feed does not spent much of it for the osmoregulation, where of increasing the amount of energy available for growth (Al-Khashali, 2015). Bolton (2016) noted the beneficial effects of low and medium levels of table salt NaCl in stimulating the mechanism of ionic and osmoregulation, increasing fish appetite, thus raising feed conversion rates, while low rates and efficiency of feed conversion were observed in Atlantic salmon Salmo salar fed on high levels of salt due to increased food passage through the gastrointestinal tract and a lack of full utilization of food due to salt effect in accelerating food evacuation rates (Salman and Eddy, 1990). Shortage in fish growth and low feed conversion rates in hypertonic environments have been reported in several studies, with a decline in the conversion rate of goldfish Carassius auratus (AlKhashali, 2015) and Common carp Cyprinus carpio (Alhilali and Al-Khashali, 2016) with increased salinity, while Gilthead sea bream Sparus aurata showed good feed conversion rate at salinity of 12 g/l. compared with 38 g/l. (Laiz-Carrion et al., 2015). DeBoeck et al. (2010) indicates that the transfer of common carp Cyprinus carpio to salinity of 10 g /l has a negative effect on the feed conversion rate and protein intake. A decrease in the growth rate of goldfish Carassius auratus exposed to salinity of 8 and 10 g /l was observed, compared to salinity of 2 g /l and fresh water (Luz et al., 2008).It was found that a decrease in feed conversion rate to16.59, 24.49 and -20.11, when the salinity increased to 5, 10 and 15 g/L respectively, compared to the control treatment (13.32) in common carp (Al-Khashali, 2017). The growth rate in grass carp Ctenopharyngodon idella and common carp was decreased with exposure to 5 and 10 g /l, compared with those exposed on tap water (0.1 g/l) (Jaafar, 2012). A significant decrease in feed conversion rate was observed with increased salinity in goldfish exposed to the salt concentrations of 0, 1, 2, 3, 4 and 5 g/l. (Lawson and Alake, 2014).

### Feed Intake

Water temperature ranged from 20 to 21°C, dissolved oxygen 5.5-5.9 mg/l. and pH 7.0-7.4, these environmental factors were appropriate for the living and growth of grass carp (Hattingh et al., 1975). Grass carp feeding on diets with different proportions of NaCl resulted in a difference in their dietary responses. The percentage of feed intake in low- and high-salty diets (1 and 7% salt) were decreased to 3.75 and 3.42 respectively, while at medium proportions of NaCl (3 and 5% salt) were 4.75% and 4.18%, respectively (Table 3).

#### Table 3 : Rate of feed intake in grass carp fed on different saline diets

<table>
<thead>
<tr>
<th>Ratio of salt in diet %</th>
<th>Feed intake (g/100 g fish)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>3.75 b</td>
</tr>
<tr>
<td>3</td>
<td>4.45 a</td>
</tr>
<tr>
<td>5</td>
<td>4.18 a</td>
</tr>
<tr>
<td>7</td>
<td>3.42 b</td>
</tr>
</tbody>
</table>

High rate of feed intake at the 3 and 5% NaCl levels may be due to the fish ability to adapt to the new conditions, where NaCl in the second and third diets have stimulated the mechanism of discharge or removal of excessive salts from the body, reducing the effect of osmotic shock, activating chloride cells which is responsible for the ion transport in gills, and increase the activity of the ATPase enzyme (adenosine triphosphatase), which is the main key to the movement of salts in chloride cells, as well as the feeding of fish on salty diets reduces the concentrations of plasma ions and the possibility of returning to the normal levels after short period, whereof reduces the amount of energy spent for the ionic regulation, thus increasing the energy allocated for growth, and reaching the internal stability (homeostasis). The low feed intake in the first treatment (control 1% salt) can be due to the low proportion of NaCl in the diet, which were not enough to stimulate the mechanism of ion exchange, and this little proportion of salt almost does not cover the need of body cells for the important elements and minerals, because only the surplus of ions is used in the ionic regulation and balance between the fish and its ambient. While the lower feed intake in the fourth treatment (7% salt) may be attributed to the saline stress that fish exposed resulting from the high salt load and the high concentrations of ions in the blood, which requires additional energy allocated to the ion regulation in an attempt of the fish to get rid of excess salts, re-ion concentration to their normal levels, which in turn leads to decreased appetite, reduce the amount of feed consumed as a percentage of body weight, as well as
weak metabolism (Al-Khshali and Alhilali, 2017). Results of this study agree with several studies, where the feed intake in gilthead seabream *Sparus aurata*, transferred to saline concentrations 8, 18, 28 g /l. and natural sea water, decreased with increasing saline concentrations (Conides, 2014). It was noted that the exposure of common carp *Cyprinus carpio* to sodium chloride solution (10 g / L) has reduced the feed intake by as much as 70% (DeBoeck *et al*., 2010). Arjona *et al* (2014) recorded a decline in the feed intake and growth rate of *Solea senegalensis* (32 g /L) compared with lower salts.

**Conclusion**

This study had confirmed that the improvement in the rate and efficiency of feed conversion and the amount of protein intake occurred in the second treatments (3% NaCl) and the third (5% NaCl), compared to the first treatment (control 1% NaCl), and the fourth (7% NaCl), because of the positive role of medium ratios of NaCl in the body homeostasis of grass carp, and then reducing the energy expenditure devoted for the osmoregulation process.

**References**


