THE INFLUENCE OF DIFFERENT STOCKING DENSITIES ON GROWTH PERFORMANCES OF ONCORHYNCHUS MYKISS (WALBAUM, 1792) IN A RECIRCULATING AQUACULTURE SYSTEM

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Abstract

The purpose of this experiment was to evaluate how stocking density influence the growth performance of juvenile rainbow trout (Oncorhynchus mykiss) reared in a recirculating system. The experiment took place over a period of 33 days. A number of 254 rainbow trout with an average weight of 29,51 ± 1,32 g were divided into four rearing units in order to create different stocking densities: in B1-2,64 kg/m³, with an average weight of 31,68 g/ex, B 2 – 5,16 kg/m³ with an average weight of 30,39 g/ex, B 3 – 7,12 kg/m³ with an average weight of 28,52 g/ex and B 4– 9,42 kg/m³ with an average weight of 27,46 g. At the end of the experiment specific growth rate (SGR) calculated in B1 variant was 2,84% (g/day), in B 2-3,36% (g/day) B 3-3,20% (g/day) and B 4-3,13% (g/day). The results of the experiment proved high technological plasticity in all the four variants. In particular, regarding the performance indicators, it has been observed that they are not influenced by the level of the stocking density. This fact concludes that the species approached in the study (Oncorhynchus mykiss) presents a high level of interest for the growth in recirculating systems of industrial aquaculture with a high level of intensity.

Key words: rainbow trout, recirculating aquaculture system, stocking density, technological performance indicators

INTRODUCTION

The rainbow trout (Oncorhynchus mykiss, Walbaum, 1792) compared to the other salmonid, is the species which is best suited for the intensive rearing, it has a rapid growth rate due to the high assimilation degree of the artificial food, as well as a high resistance to diseases.

The overwhelming importance of the stocking density for the technology of fish breeding was underlined by numerous authors who have studied this aspect on different species, as well as for different production systems (Victor C. 2004 [2]; Vasilean I., 2008 [10]).

The decrease of the growth rate is usually associated to the high level of the stocking density and was studied by numerous authors (Refstie 1977 and Holm 1986) [6]. Among the factors which influence the relationship between the density and the growth rate of a species there are the following: age, size, feeding intensity, water quality, type of production system [12], the morphology and the hydraulics of the rearing units.

It is well known the fact that high stocking densities can lead to stress (Wall,2000 [11]; Hastein,2004 [5]), can increase the vulnerability to disease (European Commission, 2004,[3]), can increase the frequency of physical injuries (Juell et al.,2003,[7]; North et al.,2006)[9], can lead to weakness of the immunity of the organism (Ellis et al.,2002)[4] can reduce the food consuming and the growth rate.

The objective of this experiment was to determine the optimum stocking density for the rainbow trout in a recirculating aquaculture system, in terms of maintaining a positive correlation between density and growth rate.
MATERIALS AND METHODS

The facilities used for this research was represented by the experimental recirculating system of the Aquaculture, Environmental Sciences and Cadastre Department, “Dunărea de Jos” University of Galati. This recirculating system is made up of the following: 4 glass tanks with a total volume of 0.320 m³ each (40×80×100 cm) and the conditioning unit of the water quality (Fig. 1).

The purpose of the conditioning unit is to control and maintain in an optimum domain the main physical-chemical parameters of the water: oxygen content, the concentration of ammonia nitrite, the concentration of solid particles, the pH and the carbon dioxide.

The biological material used for this experiment was represented by rainbow trout fingerlings (Oncorhynchus mykiss) with an average weight (±SD) of 29.51±1.32 g, fish from the fish farm Cislău. The initial individual weight values distribution for all variants showed a significant similarity with the normal distribution (p > 0.05 with the test KS).

The distribution on rearing unities (B) was made as it follows: the tank B₁ was assigned a number of 25 exemplars with an initial biomass of 792 g (2.64 kg/m³), the tank B₂ a number of 51 exemplars with an initial biomass of 1550 g (5.16 kg/m³), the tank B₃ was populated with 75 exemplars with an initial biomass of 2138,94 g (representing 7.12 kg/m³) and the tank B₄ was populated with 103 exemplars with an initial biomass of 2828.2 g (9.42 kg/m³).

For this experiment, the feeding intensity was of 3% per body weight per day (for the first six days), respectively 4 % per body weight per day (for the following 27 days of the experiment), in two meals per day. The food used was Nutra PRO MP-T – extruded pellets with the diameter of 1.7 mm and with 50% protein content (Tab. 1). During the experiment there was only one mortality registered in variant B₄ with a weight of de 47.41 g.

Table 1. Biochemical composition for the Nutra PRO MP-T

<table>
<thead>
<tr>
<th>Composition</th>
<th>%</th>
</tr>
</thead>
<tbody>
<tr>
<td>Crude protein</td>
<td>50</td>
</tr>
<tr>
<td>Crude fats and oils</td>
<td>20</td>
</tr>
<tr>
<td>Crude fibre</td>
<td>0.7</td>
</tr>
<tr>
<td>Crude ash</td>
<td>8</td>
</tr>
<tr>
<td>Sodium</td>
<td>0.4</td>
</tr>
<tr>
<td>Calciu</td>
<td>1.0</td>
</tr>
<tr>
<td>Phosphorus</td>
<td>1.2</td>
</tr>
<tr>
<td>Vitamin A</td>
<td>6000UI/Kg</td>
</tr>
<tr>
<td>Vitamin D3</td>
<td>1200UI/Kg</td>
</tr>
</tbody>
</table>

During the experiment, the attention was directed towards the control of the quality water parameters in order to maintain them in optimum intervals for growth of the specie Oncorhynchus mykiss. Out of the water physical-chemical parameters which were monitored every day in the four rearing units, we mention the following: pH, temperature, dissolved oxygen concentration (DO). Other water chemical parameters were determined on a weekly basis (concentrations particles of N-NH₄, N-NO₂, N-NO₃, P-PO₄). We mention the fact that, once the residual gross particles were evacuated, the volume of the technological recirculating water was changed and this allowed, in a satisfactory proportion, the maintaining of the water physical-chemical parameters to an optimum level for the experimented species. The following equipment was used in order to determine the physical-chemical parameters of water: oxygen concentration and the percentage of saturation were measured with the help of Hanah HI 98186 oximeter, the pH was measured with the help of pH metre WTW model 340 NH₄, NO₂, NO₃, PO₄.
concentrations were measured with the help of the Spectroquant Nova 400 photometer.

At the end of the experiment, after all fish were weighed and measured, the following technological efficiency indicators were calculated: growth rate, food conversion ration, specific growth rate and the protein efficiency ratio using the following equations [8]:

- Weight gain (W) = Final weight (Wt) – Initial weight (W0) (g)
- Food conversion ratio (FCR) = Total feed (F) / Total weight gain (W) (g/g)
- Specific growth rate (SGR) = 100 x (ln Wt - ln W0) / t (% BW/zi)
- Protein efficiency ratio (PER) = Total weight gain (W) / amount of protein fed (P) (g)

The statistical analysis was performed with the help of the programme SPSS 15.0 for Windows. The normality of the distribution was verified with the help of the Kolmogorov-Smirnov Z test. The statistical differences between variables were tested with the help of the test Anova (α = 0,05).

RESULTS AND DISCUSSIONS:

The dynamics of the water quality parameters in the rearing units for a recirculating system depends on a number of factors, the most important being the following: the efficiency of the filtering units, stocking density, feeding frequency and intensity, biochemical composition of the fodder. (Cristea V. et al., 2002) [1].

During of the present experiment, the temperature registered similar values in all rearing units, the mean value being of 16,10±0,07°C. The dissolved oxygen (DO) varied according to the population density (mean DO was 6,20±0,58 mg/l), the lower values being registered in variant B4 (5,51±0,95 mg/l), respectively in variant B3 (5,93±1,01 mg/l).

For the species Oncorhynchus mykiss, the dissolved oxygen represents the main limitative factor and, for this reason, any deviation from the interval recommended can have serious consequences on the technological productivity. As it can be noticed in figure 2, the mean values of the dissolved oxygen concentration measured for B3 and B4 tanks are lower than those registered at the level of the tanks B1 and B2. The statistical comparison between the mean values of the dissolved oxygen recorded for the four sampling water points outlined significant differences (Anova, P<0,05).

![Fig. 2 The dynamics of the dissolved oxygen during the experiment](image)

The post-hoc analysis (Tukey B) framed the mean values of DO corresponding to the four rearing units in two distinctive subsets of values; mean DO values registered for B1 and B2 tanks were significant different comparing with B3 and B4 tanks. Concerning the dynamics of the metabolism and nitrification products, they maintained within the optimal range, the average values of nitrate, nitrite, ammonia and phosphates being centralized in figure 2.

Table 2 Synthetic table with the average values (± SD) of the nitrogen and phosphate compounds

<table>
<thead>
<tr>
<th>Parameters</th>
<th>B1</th>
<th>B2</th>
<th>B3</th>
<th>B4</th>
<th>AFB</th>
<th>EFB</th>
</tr>
</thead>
<tbody>
<tr>
<td>NO₃-N</td>
<td>17.84±0.03</td>
<td>16.96±0.05</td>
<td>17.66±0.07</td>
<td>36.48±0.02</td>
<td>17.58±0.11</td>
<td>18.16±0.08</td>
</tr>
<tr>
<td>NO₂-N</td>
<td>0.152±0.01</td>
<td>0.158±0.06</td>
<td>0.152±0.05</td>
<td>0.162±0.06</td>
<td>0.162±0.08</td>
<td>0.154±0.10</td>
</tr>
<tr>
<td>NH₄-N</td>
<td>0.226±0.02</td>
<td>0.284±0.01</td>
<td>0.118±0.02</td>
<td>0.09±0.01</td>
<td>0.094±0.03</td>
<td>0.078±0.02</td>
</tr>
<tr>
<td>PO₄-P</td>
<td>0.52±0.04</td>
<td>0.426±0.06</td>
<td>0.364±0.01</td>
<td>0.386±0.05</td>
<td>0.33±0.03</td>
<td>0.35±0.04</td>
</tr>
</tbody>
</table>

Where: B1, B2, B3, B4 – system’s rearing unities
SBF – inlet biological filter
EFB – outlet biological filter
For this experiment, the water quality parameters maintained in the optimum domain, corresponding to the species *Oncorhynchus mykiss*; the equipment for the conditioning of the quality of water have succeeded in treating and reusing the technological water, taking into consideration the fact that the water daily loss did not exceeded 10 % out of the total volume of the system.

At the end of the experiment, the stocking density reached 26.42 kg/m³ in tank B₄. At the end, the final average weight of the exemplars was between 80.88 g/ex in tank B₁ and 92.22 g/ex in tank B₂ (Fig. 3).

Comparison of mean weight of the four independent variables represented by fish colonies kept in different stocking densities, there were found important differences from a statistic point of view. (Anova, p<0.05). The weight homogeneity of the tested groups was verified and confirmed done with the help of the test Levene (P>0.05). The post-hoc test outlined two subsets of values: the average weight of fish in tank B₃ being significantly different of the average weight recorded in the other tanks.

Analysis of the technological indicators (Table 3), calculated for all variants emphasized the fact that stocking density has no relevant effect on the nutrients retention and feed efficiency, this aspect being underlined by the FCR and PER values. Thus, we can notice almost identical values of FCR for the experimental variants B₂, B₃ and B₄ (0.62, 0.65 and 0.68) where the stocking density was of 2.28, 3.04 and, respectively 3.93 times higher than the control group. The protein efficiency ratio (PER) also recorded important values of 2.40 g for the tank B₁ and 3.24 g for the tank B₂.

![Fig. 3 The variation of the average individual weight – median, minimum, maximum values and quartiles registered at the end of the experiment for all experimental variants](image-url)

**Table 3** The synthetic table regarding the technological performance indicators for the rainbow trout in a recirculating aquaculture system, under different levels of intensity

<table>
<thead>
<tr>
<th>Growth performance</th>
<th>B₁</th>
<th>B₂</th>
<th>B₃</th>
<th>B₄</th>
</tr>
</thead>
<tbody>
<tr>
<td>Number of fish</td>
<td>25</td>
<td>51</td>
<td>75</td>
<td>103</td>
</tr>
<tr>
<td>Initial biomass (g)</td>
<td>792</td>
<td>1550</td>
<td>2138.94</td>
<td>2828.20</td>
</tr>
<tr>
<td>Mean individual weight (g/fish)</td>
<td>31.68</td>
<td>30.39</td>
<td>28.52</td>
<td>27.46</td>
</tr>
<tr>
<td>Initial stocking density (kg/m³)</td>
<td>2.64</td>
<td>5.16</td>
<td>7.12</td>
<td>9.42</td>
</tr>
<tr>
<td>Final numbers of fish</td>
<td>25</td>
<td>50</td>
<td>75</td>
<td>103</td>
</tr>
<tr>
<td>Final biomass (g)</td>
<td>2022</td>
<td>4611</td>
<td>6143</td>
<td>7935</td>
</tr>
<tr>
<td>Mean final fish weight (g/fish)</td>
<td>80.88</td>
<td>92.22</td>
<td>81.91</td>
<td>77.04</td>
</tr>
<tr>
<td>Final stocking density (kg/m³)</td>
<td>6.73</td>
<td>15.35</td>
<td>20.46</td>
<td>26.42</td>
</tr>
<tr>
<td>Individual weight gain (g)</td>
<td>49.20</td>
<td>61.83</td>
<td>53.39</td>
<td>49.58</td>
</tr>
<tr>
<td>Total weight gain (g)</td>
<td>1230.00</td>
<td>3061.00</td>
<td>4004.06</td>
<td>5106.80</td>
</tr>
<tr>
<td>Specific growth rate (SGR) (%/day)</td>
<td>2.84</td>
<td>3.36</td>
<td>3.20</td>
<td>3.13</td>
</tr>
<tr>
<td>Daily growth rate - (g/kg/day)</td>
<td>1.49</td>
<td>1.87</td>
<td>1.62</td>
<td>1.5</td>
</tr>
<tr>
<td>Feed conversion ratio FCR (g/g)</td>
<td>0.83</td>
<td>0.62</td>
<td>0.65</td>
<td>0.68</td>
</tr>
<tr>
<td>Protein efficiency ratio PER (g/g)</td>
<td>2.40</td>
<td>3.24</td>
<td>3.06</td>
<td>2.95</td>
</tr>
</tbody>
</table>

In this experiment, it was noticed a slight intensification of the heterogeneity regarding all tested groups, especially in the tanks B₁ and B₃ where the variation coefficient has higher values than in tanks B₂ and B₄ (29% and 24% as compared to 21% and
Nevertheless, we cannot tell that the higher stocking density led to an increase of the variability, this being caused by other factors among which the most important are: the dynamics of the physico-chemical parameters of the quality of water, genetic variability, and hydrodynamics at the level of the tanks, etc. Also, by the regression power of the individual weight and length of the tested groups, we can notice slight differences regarding the allometric factor, respectively the condition of the exemplars from the tested groups (Fig. 4).

Thus, we can say that the condition of the fish in the case of variant B₂, reflected by the high value of the allometric factor correlated with the technological indicators whose value represents the best growth performance.

![Figure 4](image)

**Figure 4** Regression length growth of rainbow trout groups maintained in different conditions of intensity

**CONCLUSIONS**

The purpose of this paper was to evaluate the productive potential of recirculating aquaculture systems in the case of rainbow trout under different conditions of intensity. The increase of the stocking density in the case of the rainbow trout, for the present experiment, leads to a slight decrease of the growth rate, insignificant from a statistic point of view ($p>0.05$). In the case of fish maintained under technological stress conditions induced by density, the increase of the heterogeneity of the group was not outlined. In this case, the efficiency of feeding and retaining of nutrients was not significantly affected by the stocking density.

Although some authors recommends, in the case of the rainbow trout, stocking densities up to 172 kg/m³ (Pickering and Pottiuger, 1987) we consider that densities of more than 25 kg/m³ cannot be practiced without physiological consequences.

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