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# Effects of Intermittent Exercise during Initial Rainbow Trout *Oncorhynchus mykiss* Rearing in Tanks Containing Vertically-Suspended Environmental Enrichment

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## Abstract

This study evaluated the effect of an exercise routine on the hatchery rearing performance of juvenile rainbow trout (*Oncorhynchus mykiss*) reared in circular tanks containing vertically-suspended environmental enrichment. This experiment occurred in two sequential trials, with fish remaining in the same treatment group in each trial. The first trial began upon initial feeding and lasted for 47 days with velocities in the exercised tanks alternated bi-weekly between 5 cm s<sup>-1</sup> and 8 cm s<sup>-1</sup>. The second trial began the day after the first trial ended and lasted for 98 days, with velocities in the exercised tanks alternating between 5 cm s<sup>-1</sup> and 11 cm s<sup>-1</sup>. Velocities in the unexercised tanks stayed constant at 5 cm s<sup>-1</sup> throughout the study. No significant differences in final tank weight, gain, percent gain, individual weight, individual length, feed conversion ratio, specific growth rate, or condition factor were found between the two treatments in either trial. These results indicate that an intermittent exercise regime does not improve the hatchery rearing performance of juvenile rainbow trout grown in circular tanks with vertically-suspended enrichment.

Keywords: Exercise, Structure, Enrichment, Rainbow trout, Oncorhynchus mykiss

# Introduction

Modifications to fish rearing units to encourage natural behaviors or mimic natural habitats have been studied with many different fish species [1-4]. Different forms of environmental enrichment have been shown to increase growth and post-stocking survival of fish raised in hatcheries [5]. Environmental enrichment research has particularly focused on salmonids during hatchery rearing [6-10].

Occupational enrichment is a category of environmental enrichment including exercise [5]. Fish are typically forced to exercise by increasing water velocities [9-11], which has generally been associated with increased fish growth. However, when fish are over-exercised, fatigue and reduced rearing performance can occur [12,13]. Most exercise studies start with relatively larger and older salmonids [2,3,9,14-16]. Beginning an exercise routine at initial feeding has not occurred.

Physical structure has been added to fish rearing tanks as a form of environmental enrichment [17-24]. Vertically-suspended structures were developed to add enrichment but still maintain circular tank hydraulic self-cleaning. Kientz and Barnes [25] and Kientz et al. [26] reported an increase in rainbow trout (*Oncorhynchus mykiss*) growth using suspended aluminum rods and suspended strings of plastic spheres. Positive results using a variety of vertically-suspended structures with a variety of salmonid species have been reported [27-33].

Voorhees et al. [16] evaluated the combination of verticallysuspended structure and an exercise routine during juvenile rainbow trout rearing and found no significant interaction between structure and exercise. Given the lack of paucity of information on exercise in conjunction with vertically-suspended structure, and exercise in general with small salmonids, the objective of this study was to evaluate the effects of an exercise routine, beginning at initial feeding, on the rearing performance of rainbow trout reared in circular tanks containing vertically-suspended environmental enrichment.

#### Methods

This experiment was conducted at McNenny State Fish Hatchery, rural Spearfish, South Dakota, USA, using degassed and aerated well-water (constant temperature 11°C; total hardness as  $CaCO_3$ , 360 mg L<sup>-1</sup>; alkalinity as  $CaCO_3$ , 210 mg L<sup>-1</sup>; pH, 7.6; total dissolved solids, 390 mg L<sup>-1</sup>) using 2,000-L circular tanks (1.8 m diameter x 0.6 m deep; 0.4 m water depth). This experiment used Arlee strain rainbow trout in two sequential trials.

All tanks contained vertically-suspended environmental enrichment, which consisted of an array of four suspended aluminum angles (2.5-cm wide on each angle side x 57.15-cm long) [27] suspended from a corrugated plastic cover [34]. The angles were placed so the peak of the angle faced into the water current.

Fish were fed every 15 minutes during daylight hours using automatic feeders. Feeding rates were determined by the hatchery constant method [35], with an expected feed conversion ratio of 1.1. Total feed fed was 116.7 kg per tank throughout the entire experiment. The experimental design for each trial was similar, with treatments being either unexercised (control) or exercise. Velocities in the control tanks were maintained at 5 cm s<sup>-1</sup>, the minimum velocity required for hydraulic self-cleaning. Exercise occurred by changing the angle of the incoming water (spray bar) to increase in-tank water velocities; incoming water flows were not changed. The exercise regime was alternating every 84 hours (3.5 days) between velocities of 5 cm s<sup>-1</sup> and higher velocities of 8 to 11 cm s<sup>-1</sup> depending on the trial. The exercise regime started after one week of acclimation to the lower velocity of 5 cm s<sup>-1</sup>. Velocities were measured using a flowmeter (JDC Electronics Flowatch Flowmeter, JDC, Yverdon-les-Bains, Switzerland), with all readings taken directly across from the incoming water spray bar approximately 20 cm deep (halfway from water surface).

The first trial began on January 14, 2020 and lasted until March 2, 2020, for a total of 47 days. Each of eight tanks received 1.2 kg (approximately 6,600 fish) of trout (individual mean  $\pm$  SE, weight =  $0.2 \pm 0.0$  g, total length  $26.8 \pm 0.4$  mm, n = 30). Feeding rates were projected at 0.08 cm day<sup>-1</sup>, a rate at or slightly above satiation. Fish were fed #1 granules (Fry, Skretting USA, Tooele, Utah, USA) until February 4, when feed was switched to #2 granules (Fry, Skretting USA, Tooele, Utah, USA). Four tanks received the control velocity and four tanks received the exercise regime (n = 4). The higher velocity used in the exercise regime was 8 cm s<sup>-1</sup>. At the end of the trial, total tank weight was obtained by weighing all fish in each tank to the nearest 0.1 kg. In addition, ten individual fish from each tank were weighed to the nearest 0.1 g and total length measured to the nearest 1.0 mm.

The second trial used the fish from the first trial and began on March 3, 2020, immediately after cessation of the first trial. The second trial lasted for 98 days until June 9, 2020, with the trout from the first trial subjected to the same treatment (control or exercised). Fish in the control treatment for the first trial were pooled (combined into one tank) and split into seven tanks, with each tank receiving 9.1 kg (approximately 3,600 fish). Initial mean ± SE weights and total lengths were 2.5  $\pm$  0.1 g and 61  $\pm$  1 mm (n = 40) respectively. The exercised fish from the first trial were also pooled and split into seven tanks, with each tank receiving 9.3 kg (mean  $\pm$  SE, weight = 2.7  $\pm$  0.1 g, total length 61  $\pm$  1 mm, n = 40). Feeding rates were based on a projected growth of 0.075 cm day-1. Fish were fed #2 granules (Fry, Skretting USA, Tooele, Utah, USA) until March 26, when the fish received 1.5 mm extruded floating pellets (Protec Trout, Skretting USA, Tooele, Utah, USA). The higher velocity used in the exercise regime was 11 cm s<sup>-1</sup>. At the end of the trial, total tank and individual fish data was recorded as in the first trial.

The following formulas were used:

Gain = end weight - start weight $Gain (%) = 100 * \frac{gain}{start weight}$  $Feed conversion ratio (FCR) = \frac{food fed}{gain}$  $Specific growth rate (SGR) = 100 * \frac{ln (end weight) - ln(start weight)}{number of days}$  $Condition factor (K) = 10<sup>5</sup> * \frac{fish weight}{fish length <sup>2</sup>}$  Data were analyzed using SPSS (24.0) statistical program (IBM Corporation, Armonk, New York, USA). T-tests were used with significance was predetermined at p < 0.05. This experiment was carried out within the American Fisheries Society "Guidelines for the Use of Fishes in Research" [36] and within the guidelines of the Aquatics Section Research Ethics Committee of the South Dakota Department of Game, Fish and Parks, USA.

#### Results

In both trials, no significant differences were found in final tank weight, gain, percent gain, feed conversion ratio, or percent mortality (Table 1). In the first trial, mean feed conversion ratios were relatively low at 0.75 in the control and 0.77 in those tanks of fish that were exercised. Feed conversion ratios were higher in the second trial and nearly identical between the groups at 1.12 in the control and 1.11 in the exercised treatment. Overall feed conversion for both trials combined was identical between the control and exercised groups. Mortality was relatively low and did not exceed 1.2% in either trial.

Individual total length, weight, specific growth rate, and condition factor were also not significantly different between the two groups in either trial (Table 2). Specific growth rate decreased from 5.59 and 5.73 in the control and exercised groups in the first trial respectively, to 2.81 and 2.70 in the second trial.

|         |                     | Unexercised     | Exercised       |
|---------|---------------------|-----------------|-----------------|
| Trial 1 | n                   | 4               | 4               |
|         | Initial weight (kg) | 1.2             | 1.2             |
|         | Final weight (kg)   | $16.0 \pm 0.8$  | 16.3 ± 0.3      |
|         | Gain (kg)           | $14.7 \pm 0.8$  | $15.0 \pm 0.3$  |
|         | Gain (%)            | $1,207 \pm 64$  | $1,232 \pm 22$  |
|         | FCR                 | $0.75 \pm 0.04$ | $0.77 \pm 0.01$ |
|         | Mortality (%)       | $1.2 \pm 0.3$   | $1.0 \pm 0.1$   |
|         | n                   | 7               | 7               |
|         | Initial weight (kg) | 9.1             | 9.3             |
|         | Final weight (kg)   | 118.3 ± 2.6     | $116.7 \pm 1.9$ |
| Trial 2 | Gain (kg)           | 109.2 ± 2.6     | 107.6 ± 1.9     |
|         | Gain (%)            | 1,200 ± 28      | $1,183 \pm 21$  |
|         | FCR                 | $1.12 \pm 0.03$ | $1.11 \pm 0.02$ |
|         | Mortality (%)       | $0.8 \pm 0.1$   | 0.6 ± 0.1       |
| Overall | FCR                 | 0.94            | 0.94            |

**Table 1:** Mean ( $\pm$  SE) final tank weight, gain, percent gain, feed conversion ratio (FCR<sup>a</sup>), and percent mortality of rainbow trout reared with or without exercise.

<sup>a</sup>FCR = Food fed/gain.

**Table 2:** Individual fish mean ( $\pm$ SE) total length, weight, specific growth rate (SGR<sup>a</sup>), and condition factor (K<sup>b</sup>) of rainbow trout reared with or without exercise.

|         |             | Unexercised     | Exercised       |
|---------|-------------|-----------------|-----------------|
| Trial 1 | Length (mm) | 60 ± 2          | 61 ± 1          |
|         | Weight (g)  | $2.5 \pm 0.2$   | $2.7 \pm 0.2$   |
|         | SGR         | $5.59 \pm 0.19$ | $5.73 \pm 0.16$ |
|         | К           | $1.14 \pm 0.05$ | $1.16 \pm 0.01$ |
| Trial 2 | Length (mm) | 146 ± 2         | $143 \pm 2$     |
|         | Weight (g)  | 38.5 ± 1.6      | 37.2 ± 2.1      |
|         | SGR         | $2.81 \pm 0.04$ | 2.70 ± 0.05     |
|         | К           | $1.24 \pm 0.01$ | $1.26 \pm 0.02$ |

 ${}^{a}SGR = 100 * (ln (end weight) - ln (start weight))/(number of days).$  ${}^{b}K = 10^{5} * [(fish weight)/(fish length)^{3}].$ 

# Discussion

The results of this study indicate that exercise beginning with initial feeding does not improve the growth of rainbow trout during hatchery rearing. This study is unique because it used small rainbow trout, which were only 27 mm long at the start of the experiment. Most research using exercise in juvenile salmonids begins when the fish are larger. For example, Parker and Barnes [37] found positive effects of exercise in a trial using 72 mm long rainbow trout. Voorhees et al. [16] conducted an exercise study with 69 mm rainbow trout and Reiser et al. [11] used rainbow trout that were 409 mm long. Exercise studies using other salmonid species have also used larger fish, such as the 130 mm Arctic charr (*Salvelinus alpinus*) used by Christiansen and Jobling [12].

The lack of difference in growth between the control and exercise regime tanks in this study support the observations of Voorhees et al. [16] that a combination of both physical and occupational enrichment was not necessary to improve the hatchery rearing performance of rainbow trout. Voorhees et al. [16] reported that either exercise or vertically-suspended structures increased trout growth, but that no further improvements were realized when both forms of environmental enrichment were combined. The lack of any improvement in growth observed using exercise in this study was likely due to the presence of vertically-suspended structures in both the exercise and non-exercise tanks.

The water velocities used for exercise in this experiment may have affected the results. At the start of the first trial, the exercise velocities were 5 cm s<sup>-1</sup>, which based on fish size was approximately 3.1 body lengths per second. By the end of the second trial, relative velocities in the exercised fish had decreased to approximately 0.9 body lengths per second. The preferred relative velocity for optimal exercise in salmonids has been reported to be between 1.5 and 2.0 body lengths per second [6,37,38]. The relatively high velocities used for exercise at the start of the trial may have been particularly deleterious. Exercising Chinook salmon (*Oncorhynchus tshawytscha*) at 3.0 body lengths per second resulted in poorer feed conversion ratios compared to those exercised at 1.5 body lengths per second [37].

The feed conversion ratios obtained in this study are consistent with the observations of Huysman et al. [29] and Voorhees et al. [16]. They are also similar to studies using similarly sized rainbow trout in environmental enrichment studies [15,25,27]. The decreased feed conversion ratio in the first trial compared to the second trial was likely due to the smaller size of the fish [39].

It is possible that the high rearing densities encountered at the end of the second trial may have influenced trout growth [40-42]. It is possible, as suggested by Huysman et al. [29], that the trout in the exercised tanks initially grew more rapidly, with growth slowing as they achieved relatively high densities faster than the unexercised tanks. In other words, the fish in the control tanks may have grown more slowly throughout the course of the second trial and only achieved the high final densities because of the long duration of the experiment. Interestingly, the final tank densities observed in this study were higher than those reported by Huysman et al. [29]. The specific growth rates of this study are consistent with those of similarly-sized rainbow trout reared using vertically-suspended enrichment [15,16,29]. However, Gregory and Wood [7] reported a much lower specific growth rate for juvenile rainbow trout of similar size who were exercised intermittently than was observed in this experiment. The difference may be because of the inclusion of vertically-suspended enrichment, which has been shown to improve specific growth rate [25,27]. It may also be due to differences in fish size, water temperature, water chemistry, diet, or the genetic strain used, though the results are similar to other non-enriched salmonid exercise studies [7,15,43].

In conclusion, the results of this study indicate no significant improvement in the rearing performance of rainbow trout when exercised in the presence of vertically-suspended environmental enrichment. Further research should examine the interaction of exercise routines and vertically-suspended enrichment, including evaluating exercise routines designed to minimize the risk of exercise fatigue.

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