

Research Article

Effect of the water exchange rate in a recirculation aquaculture system on growth, glucose and cortisol levels in *Oreochromis niloticus*

Jesús Josafat De León-Ramírez¹ , Juan Fernando García-Trejo¹ , Leticia Felix-Cuencas¹ 
Samuel López-Tejeda¹ , Carlos Francisco Sosa-Ferreira²  & Alexa Ivanna González-Orozco³ 

¹Universidad Autónoma de Querétaro, Facultad de Ingeniería, Querétaro, México

²Universidad Autónoma de Querétaro, Facultad de Medicina, Querétaro, México

³Centro de Investigación en Ciencia Aplicada y Tecnología Avanzada, Querétaro, México

Corresponding author: Juan Fernando García-Trejo (fernando.garcia@uaq.mx)

ABSTRACT. In recirculating aquaculture systems (RAS), the water exchange rate influences the removal of waste compounds. However, the inappropriate exchange rate favors the presence of stress factors, causing alterations in the cultured organisms. Therefore, the aim was to determine the effect of the water exchange rate in the different productive stages (fingerling, juvenile, and adult) of Nile tilapia (*Oreochromis niloticus*). Three exchange rates were used: rates of 1.2 (T_1), 2.8 (T_2), and 5.3 (T_3) tank volume h^{-1} . The following were established as response variables: growth rate (GR), survival rate (SR), feed conversion rate, protein efficiency, and condition factor. Likewise, cortisol and glucose concentrations were established as explanatory variables. The results suggest that in the fingerling stage, the T_2 treatment contributes most to the productive performance (GR = 40.24 g and SR = 95%), keeping low levels of cortisol and glucose (6.76 ng mL^{-1} and 33.73 mg dL^{-1}). In the juvenile stage, T_3 treatment shows the best result both in productive performance (GR = 117.69 g and SR = 90%) and in cortisol and glucose concentrations (35.13 ng mL^{-1} and 70.67 mg dL^{-1}). Finally, all treatments present cortisol and glucose levels above the normal range in the adult stage, T_1 , where the highest productive performance is presented (GR = 90.06 g and SR = 95%). The information leads us to consider the variation in the exchange rate in a RAS through the different stages of the Nile tilapia to maintain the favorable conditions that lead to the highest performance.

Keywords: *Oreochromis niloticus*; blood chemistry; productive performance; stress; water quality; aquaculture

INTRODUCTION

In aquaculture, recirculating aquaculture systems (RAS) are a tool that contributes to optimizing the use of water and its management prior to the discharge of wastewater into the environment (Martins et al. 2010). These systems use mechanical and biological treatments that allow maintaining the required physicochemical conditions in the water for a greater time, reducing the amount of water used in each production cycle. Likewise, the amount of waste present in the water leaving the system is reduced (Verdegem et al. 2005, Schram et al. 2009).

The design of RAS is primarily based on the water quality needs for the different cultivated species and the intensity of the cultivation. In this regard, one of the

factors that intervene in the removal of compounds is the exchange rate or water flow (Schram et al. 2009), linked to the number of compounds that would be dissolved in the water (feed, feces, urine) (Timmons et al. 2002, Martins et al. 2009).

The above shows that the incorrect exchange rate favors the presence of different stress factors that lead to relevant alterations in the organisms (Barreto & Volpato 2006, Qiang et al. 2016). In RAS, the water exchange rate varies from one to six changes per hour (Summerfelt et al. 2004, D'Orbcastel et al. 2009, Buřič et al. 2016). It has been reported that low exchange rates favor the accumulation of toxic compounds in the water, which compromises the health of the fish (Deviller et al. 2005, Martins et al. 2009, Davidson et al. 2011). On the contrary, high exchange rates promote

greater water agitation, favoring the resuspension of solids and the release of toxic compounds (Schram et al. 2009, Cheng et al. 2017).

The alterations are grouped into three levels of response: primary, secondary, and tertiary. The primary response consists of the activation of the endocrine system, where the cortisol concentration is used as an indicator of the intensity of the response (Martínez-Porchas et al. 2009, Pankhurst 2011). The secondary response lies in physiological and metabolic changes, the glucose concentration being the indicator that allows determining the intensity of the response (Cnaani et al. 2004, Martínez-Porchas et al. 2009). Finally, the tertiary response has repercussions on the biological and productivity, limiting the growth of the organisms and thus the system's performance (Wu et al. 2015).

In general, the water exchange rate within the RAS remains constant throughout the culture; however, little has been addressed on the possibility of exchange rate variation in the different stages of the productive cycle. Therefore, the objective of this work is to determine the effect of the water exchange rate in the different productive stages (fingerling, juvenile, and adult) based on growth, cortisol, and glucose levels in the Nile tilapia (*Oreochromis niloticus*) specimens.

MATERIALS AND METHODS

Recirculation system

Three RAS were used. Each of the RAS was made up of three geomembrane tanks, each with a capacity of 1000 L, a sedimentation tank of 250 L, a canister-type filter equipped with a UV lamp; likewise, each RAS had an accumulation tank for filtered water prior to its reincorporation into the culture tanks (Fig. 1).

Biological material

A total of 2250 Nile tilapia (*Oreochromis niloticus*) fingerlings were used for 180 days. The fingerlings were 25 days old with an initial mean weight and a standard deviation of 0.85 ± 0.04 g. The fingerlings were distributed in nine tanks (three from each RAS) at a density of 250 individuals on 1000 L (Félix-Cuencas et al. 2021). During the experimental period, the tilapia specimens were fed four times a day with a balanced feed of the Malta-Cleyton® brand (Table 1). The feed not consumed in each of the schedules was removed after 30 min of being supplied. The amount of feed for each tank was adjusted weekly following biometry.

Experimental design

A randomized block experimental design was used, with three treatments (each of the RAS) and three

repetitions (each tank inside the RAS), where the experimental unit was 250 individuals of Nile tilapia. The treatments consisted of three water exchange rates per hour: T₁ (rate of 1.2 with an exchange volume of 3600 L h⁻¹), T₂ (rate of 2.8 with an exchange volume of 8400 L h⁻¹), and T₃ (rate of 5.3 with an exchange volume of 15,900 L h⁻¹).

Water quality

The water in the tanks was monitored daily for temperature and dissolved oxygen variables using a Hach HQ40d®. Likewise, the concentrations of phosphate, nitrates, nitrites, and non-ionized ammonia were monitored and determined by the Hach DR6000® spectrophotometer under the methods 8180, 8039, 8057, and 8038, respectively.

Productive performance

The response variables were the growth rate (GR), the survival rate (SR), the feed conversion rate (FCR), the protein efficiency (PE), and the condition factor (CF) using the following formulas:

$$GR (g) = \text{final weight (g)} - \text{initial weight (g)} \quad (1)$$

$$SR (\%) = \frac{\text{initial number of animals}}{\text{final number of animals}} \times 100 \quad (2)$$

$$FCR = \frac{\text{grams of feed consumed}}{\text{grams of increase in weight}} \quad (3)$$

$$PE = \frac{\text{increase in weight}}{\text{grams of protein ingested}} \quad (4)$$

$$CF = \frac{\text{weight (g)}}{\text{length (cm}^3\text{)}} \times 100 \quad (5)$$

Extraction and quantification of cortisol and glucose

Blood extraction and quantification of the mentioned compounds were carried out to evaluate the effect of the water exchange rate on cortisol and glucose levels. Before sampling, the fish have fasted for 24 h (Abdel-Tawwab et al. 2005). The sampling was carried out on days 60, 120, and 180 of the experimental periods, taking from each treatment: 30 (fingerlings), 20 (juveniles), and 10 (adults) fish, respectively. The fish were anesthetized using a eugenol solution (100 mg L⁻¹) with an exposure time of 3 min (Santiago-Rucínque et al. 2017).

The samples were taken from the caudal vein and were deposited in a tube with a coagulant for centrifugation at 3500 rpm for 15 min, taking the blood plasma for subsequent analysis. Cortisol concentration was determined using an ELISA kit from the Neogen® brand, while the glucose was determined using an analytical test kit from the Biodiagnostic® brand (Inoue et al. 2008, Haraz et al. 2018).

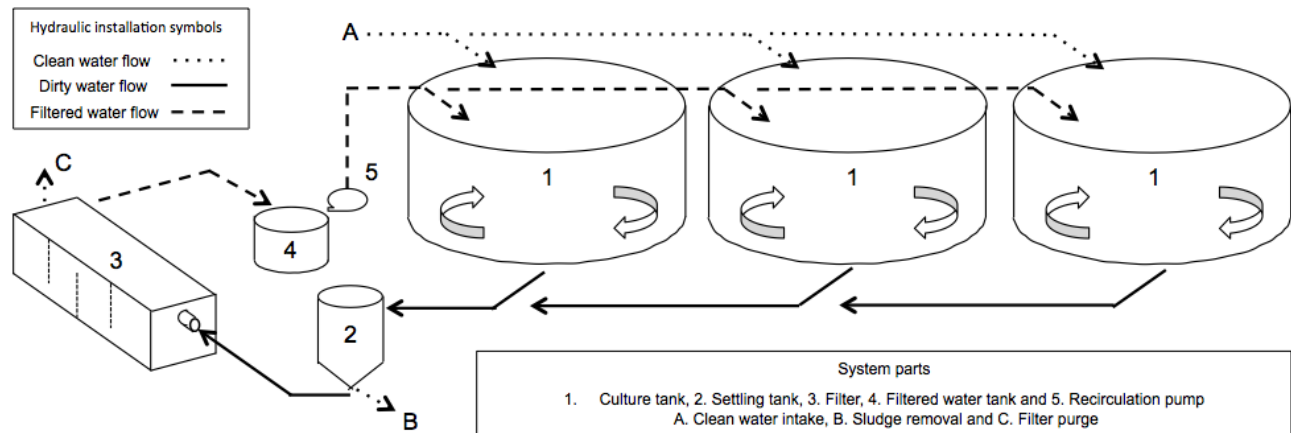


Figure 1. General diagram of recirculating aquaculture systems used.

Table 1. Description of the meal plan used during the experiment.

| Fish stage | Fish weight (g) | Percentage of feed supplied (%) | Feeding times (h) and ration percentages | Characteristics of the feed | | |
|-------------|-----------------|---------------------------------|--|-----------------------------|--------------------|-------------------|
| | | | | Pellet diameter (mm) | Protein amount (%) | Lipids amount (%) |
| Fingerlings | 0.1-5 | 10 | | 0.8 | 45 | 16 |
| Fingerlings | 5-10 | 10 | 08:00 h (15%) | 1.5 | 45 | 16 |
| Fingerlings | 10-40 | 8 | 11:00 h (30%) | 2.5 | 45 | 16 |
| Juveniles | 40-60 | 6 | 14:00 h (35%) | 2.5 | 45 | 16 |
| Juveniles | 60-160 | 4 | 17:00 h (20%) | 3.5 | 40 | 3.5 |
| Adults | 160-250 | 2 | | 4.5 | 35 | 5.5 |

Statistics analysis

Data analysis was performed using JMP® software (9.0.1). The data collected for each variable were subjected to an analysis of variance (ANOVA); the results were expressed as mean \pm standard deviation. Likewise, Tukey's test was performed to determine the significant differences between the means of the treatments, using a significance level of $P < 0.05$.

RESULTS

Water quality

During the 180 days of experimentation (considering the three development stages: fingerling, juvenile, and adult), the values obtained in the water quality monitoring were within the tolerance ranges for the culture of *Oreochromis niloticus* (Table 2). From the above, it can be inferred that the water purifying elements were efficient in maintaining quality, considering the density of organisms used, the water exchange rates, and the accumulation of compounds throughout the experiment. However, this does not rule out the relationship between the concentration of different compounds present in the water and each stage's productive performance.

Productive performance

The results of productive performance in the fingerling stage presented significant differences (Table 3). The GR values showed that T₂ (exchange rate of 2.8) had the highest weight gain (40.24 ± 1.83 g), while diet T₁ and T₃ (exchange rate of 1.2 and 5.3 respectively) did not show differences ($P < 0.05$) among them. SR showed significant differences between treatments, with T₂ being the best-valued treatment ($95 \pm 1\%$). Likewise, the T₂ treatment was located with the outstanding values for the rest of the variables (FCR = 1.61 ± 0.02 , PE = 1.37 ± 0.03 , and CF = 1.06 ± 0.03).

In the juvenile stage (Table 4), the GR and SR variables did not show differences between T₁ and T₂ however, the values of both treatments were lower than T₃ (GR = 131.43 ± 6.64 g and SR = $95 \pm 3\%$). In the case of FCR and PE, T₃ was the best-valued treatment (1.74 ± 0.04 and 1.4 ± 0.04 , respectively). However, CF did not show significant differences concerning T₁ and T₂.

Finally, in the adult stage (Table 5), T₃ was the treatment with the lowest growth rate (71.05 ± 10.12 g), showing differences concerning T₁ and T₂ (93.97 ± 9.15 and 90.06 ± 8.76 g, respectively). Regarding SR

Table 2. Water quality in the different treatments: T₁ (exchange rate of 1.2), T₂ (exchange rate of 2.8), and T₃ (exchange rate of 5.3). Values are presented as means \pm standard deviation of samples collected during the experimental period. Values with different superscripts present significant differences ($P < 0.05$).

| Variable | Values of culture | T ₁ | T ₂ | T ₃ |
|---|-------------------|-------------------------------|-------------------------------|--------------------------------|
| | | Fingerling stage | | |
| Temperature (°C) | 20 - 32 | 23.7 \pm 1.8 ^a | 24.1 \pm 1.2 ^a | 24.6 \pm 0.9 ^a |
| Dissolved oxygen (mg L ⁻¹) | 4 - 9 | 7.36 \pm 0.35 ^a | 7.45 \pm 0.32 ^a | 7.94 \pm 0.43 ^a |
| Nitrates (mg L ⁻¹) | <300 | 15.66 \pm 3.93 ^b | 29.18 \pm 4.25 ^a | 24.05 \pm 3.12 ^{ab} |
| Nitrites (mg L ⁻¹) | <5 | 0.36 \pm 0.11 ^b | 0.95 \pm 0.14 ^a | 0.74 \pm 0.15 ^a |
| Non-ionized ammonia (mg L ⁻¹) | <2 | 0.79 \pm 0.12 ^a | 0.32 \pm 0.19 ^{ab} | 0.51 \pm 0.16 ^b |
| Phosphate (mg L ⁻¹) | <1.4 | 0.30 \pm 0.05 ^b | 0.43 \pm 0.03 ^a | 0.39 \pm 0.04 ^{ab} |
| Juvenile stage | | | | |
| Temperature (°C) | 20 - 32 | 21.6 \pm 1.1 ^a | 22.1 \pm 0.9 ^a | 21.9 \pm 1.0 ^a |
| Dissolved oxygen (mg L ⁻¹) | 4 - 9 | 6.92 \pm 0.42 ^a | 6.73 \pm 0.36 ^a | 6.84 \pm 0.37 ^a |
| Nitrates (mg L ⁻¹) | <300 | 23.12 \pm 3.53 ^b | 35.86 \pm 7.43 ^a | 34.03 \pm 6.86 ^{ab} |
| Nitrites (mg L ⁻¹) | <5 | 0.60 \pm 0.13 ^b | 1.28 \pm 0.19 ^a | 1.01 \pm 0.12 ^a |
| Non-ionized ammonia (mg L ⁻¹) | <2 | 1.09 \pm 0.18 ^a | 0.68 \pm 0.17 ^b | 0.79 \pm 0.12 ^{ab} |
| Phosphate (mg L ⁻¹) | <1.4 | 0.42 \pm 0.07 ^a | 0.54 \pm 0.08 ^a | 0.52 \pm 0.09 ^a |
| Adult stage | | | | |
| Temperature (°C) | 20 - 32 | 25.8 \pm 1.3 ^a | 26.2 \pm 1.3 ^a | 25.6 \pm 1.2 ^a |
| Dissolved oxygen (mg L ⁻¹) | 4 - 9 | 5.93 \pm 0.38 ^a | 6.28 \pm 0.41 ^a | 5.74 \pm 0.23 ^a |
| Nitrates (mg L ⁻¹) | <300 | 33.38 \pm 6.73 ^b | 53.38 \pm 5.09 ^a | 46.98 \pm 6.09 ^a |
| Nitrites (mg L ⁻¹) | <5 | 0.98 \pm 0.25 ^b | 1.56 \pm 0.09 ^a | 1.34 \pm 0.21 ^{ab} |
| Non-ionized ammonia (mg L ⁻¹) | <2 | 1.4 \pm 0.13 ^a | 0.99 \pm 0.14 ^b | 1.02 \pm 0.11 ^b |
| Phosphate (mg L ⁻¹) | <1.4 | 0.55 \pm 0.05 ^b | 0.68 \pm 0.08 ^a | 0.68 \pm 0.07 ^a |

Table 3. Productive performance that presents the means \pm standard deviation of growth rate (GR), survival rate (SR), feed conversion rate (FCR), protein efficiency rate (PE), and condition factor (CF) at the end from the fingerling stage (60 days of experimentation). Values with different superscripts present significant differences ($P < 0.05$).

| Variable | Fingerling stage | | |
|--------------------|-------------------------------|-------------------------------|-------------------------------|
| | T ₁ | T ₂ | T ₃ |
| Initial weight (g) | 0.88 \pm 0.03 | 0.85 \pm 0.03 | 0.83 \pm 0.03 |
| Final weight (g) | 37.92 \pm 1.38 | 41.09 \pm 2.09 | 38.14 \pm 1.75 |
| GR (g) | 37.04 \pm 1.27 ^b | 40.24 \pm 1.83 ^a | 36.31 \pm 1.39 ^b |
| SR (%) | 83 \pm 3 ^b | 95 \pm 1 ^a | 87 \pm 3 ^b |
| FCR | 1.82 \pm 0.05 ^b | 1.61 \pm 0.02 ^c | 1.94 \pm 0.06 ^a |
| PE | 1.25 \pm 0.03 ^b | 1.37 \pm 0.03 ^a | 1.14 \pm 0 |
| CF | 0.86 \pm 0.02 ^b | 1.06 \pm 0.03 ^a | 0.77 \pm 0.02 ^c |

and CF, the treatments did not show significant differences. For FCR and PE, T₁ was the best-valued treatment (1.83 \pm 0.03 and 1.51 \pm 0.02, respectively).

Cortisol and glucose concentration

Cortisol levels showed significant differences between treatments (Fig. 2). At the end of the fingerling stage (day 60), T₂ was the treatment with the lowest concentration (6.76 \pm 0.59 ng mL⁻¹). At the end of the juvenile stage (day 120), T₃ was the treatment with the lowest concentration (35.13 \pm 4.35 ng mL⁻¹). Finally,

on day 180 of the T₁ experiment the treatment was the lowest concentration (110.12 \pm 8.64 ng mL⁻¹).

On the other hand, glucose levels also showed significant differences between treatments (Fig. 3). At the end of the fingerling stage (day 60), T₂ was the treatment with the lowest concentration (33.73 \pm 0.98 mg dL⁻¹). At the culmination of the juvenile stage (day 120), T₁ was the treatment with the highest concentration (70.67 \pm 3.53 mg dL⁻¹). Finally, at the end of the experimentation (day 180), T₁ was the treatment with the highest concentration (98.63 \pm 4.92 mg dL⁻¹).

Table 4. Productive performance that presents the means \pm standard deviation of growth rate (GR), survival rate (SR), feed conversion rate (FCR), protein efficiency rate (PE), and condition factor (CF) at the end from the fingerling stage (120 days of experimentation). Values with different superscripts present significant differences ($P < 0.05$).

| Variable | Juvenile stage | | |
|--------------------|--------------------------------|--------------------------------|--------------------------------|
| | T ₁ | T ₂ | T ₃ |
| Initial weight (g) | 37.92 \pm 1.38 | 41.09 \pm 2.09 | 38.14 \pm 1.75 |
| Final weight (g) | 143.61 \pm 6.68 | 158.78 \pm 7.79 | 169.57 \pm 5.05 |
| GR (g) | 105.69 \pm 5.75 ^b | 117.69 \pm 6.14 ^b | 131.43 \pm 6.64 ^a |
| SR (%) | 89 \pm 2 ^b | 90 \pm 1 ^b | 95 \pm 3 ^a |
| FCR | 2.18 \pm 0.02 ^a | 2.01 \pm 0.02 ^b | 1.74 \pm 0.04 ^c |
| PE | 1.11 \pm 0.03 ^b | 1.21 \pm 0.05 ^b | 1.40 \pm 0.04 ^a |
| CF | 0.92 \pm 0.03 ^a | 1.05 \pm 0.05 ^a | 1.02 \pm 0.05 ^a |

Table 5. Productive performance that presents the means \pm standard deviation of growth rate (GR), survival rate (SR), feed conversion rate (FCR), protein efficiency rate (PE), and condition factor (CF) at the end from the fingerling stage (180 days of experimentation). Values with different superscripts present significant differences ($P < 0.05$).

| Variable | Adult stage | | |
|--------------------|-------------------------------|-------------------------------|--------------------------------|
| | T ₁ | T ₂ | T ₃ |
| Initial weight (g) | 143.61 \pm 6.68 | 158.78 \pm 7.79 | 169.57 \pm 5.05 |
| Final weight (g) | 237.58 \pm 10.37 | 248.84 \pm 9.83 | 240.62 \pm 11.56 |
| GR (g) | 93.97 \pm 9.15 ^a | 90.06 \pm 8.76 ^a | 71.05 \pm 10.12 ^b |
| SR (%) | 95 \pm 1 ^a | 95 \pm 3 ^a | 92 \pm 3 ^a |
| FCR | 1.83 \pm 0.03 ^b | 1.91 \pm 0.06 ^b | 2.12 \pm 0.04 ^a |
| PE | 1.51 \pm 0.02 ^a | 1.45 \pm 0.03 ^b | 1.31 \pm 0.02 ^c |
| CF | 1.02 \pm 0.05 ^a | 1.05 \pm 0.05 ^a | 1.06 \pm 0.04 ^a |

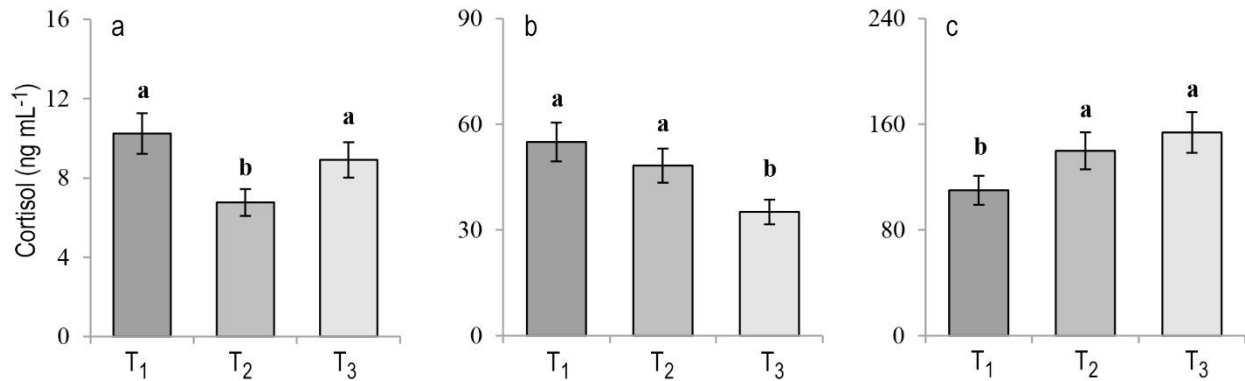


Figure 2. Cortisol concentrations for T₁ (exchange rate of 1.2), T₂ (exchange rate of 2.8), and T₃ (exchange rate of 5.3) after a) 60, b) 120, and c) 180 days. Values are presented as means \pm standard deviation; significant differences ($P < 0.05$) are indicated with different superscripts.

DISCUSSION

The temperature variable oscillated below the optimal value of 28°C (Azaza et al. 2008), without presenting significant differences between the treatments during the three stages of development (juvenile, juvenile, and adult). The dissolved oxygen concentration had a decrease throughout the development stages, being that higher total biomass in the tanks leads to higher oxygen

consumption (Valbuena-Villareal et al. 2006, Tomalá et al. 2014); however, within each development stage (juvenile, juvenile, and adult), the treatments did not show significant differences. So, it can be inferred that the dissolved oxygen did not limit the development of the fish. Therefore, their interference with the different treatments was ruled out.

The values obtained were below the toxic levels for tilapia cultivation regarding nitrogenous compounds.

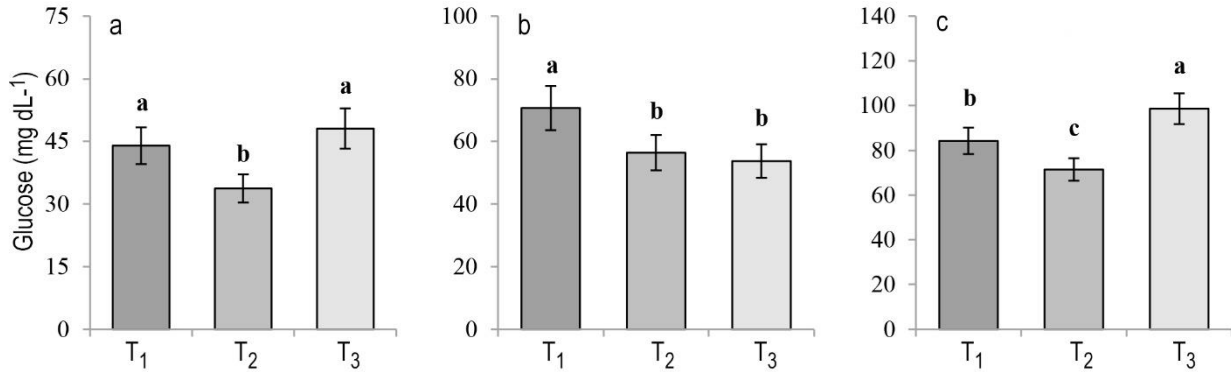


Figure 3. Glucose concentrations for T₁ (exchange rate of 1.2), T₂ (exchange rate of 2.8), and T₃ (exchange rate of 5.3) after a) 60, b) 120, and c) 180 days. Values are presented as means \pm standard deviation; different superscripts indicate significant differences ($P < 0.05$).

During the three stages of development (fingerling, juvenile, and adult), the highest concentrations of non-ionized ammonia (0.79 ± 0.12 , 1.09 ± 0.18 and 1.4 ± 0.13 mg dL⁻¹, respectively) present in T₁ could be attributed to the accumulation of waste substances due to the low water rotation (Sun et al. 2011). On the other hand, the concentrations in T₃ (0.51 ± 0.16 , 0.79 ± 0.12 and 1.02 ± 0.11 mg dL⁻¹, respectively) could be because the flow velocity did not allow the water to have the necessary interaction and residence time in the elements purification system (sedimentation tank and filter) for the proper removal of non-ionized ammonia (Cheng et al. 2017).

The fingerling stage's growth and survival data suggest that a mean water exchange rate (T₂) favors these variables. The above can be linked to adequate removal of waste substances and a nitrification process (Michaud et al. 2006). The FCR, PE, and CF coincide for T₂ with outstanding values over the rest of the treatments. The CF for T₁ and T₃ presented values lower than 1, suggesting that the fish immersed in these treatments are under a certain degree of stress (Migiro et al. 2014). In the T₁ treatment, the non-ionized ammonia concentration could be indicated as the stress factor for the fish of said treatment (Cavero et al. 2004). Regarding T₃, the water disturbance due to the higher exchange rate could be indicated as the stress factor.

In the juvenile stage, the growth and survival values were favored by the higher exchange rate (T₃ = 5.3); this agrees with that reported by Obirikorang et al. (2019), who concluded that an exchange rate of six times the volume of the tank per hour promotes greater weight gain. Despite not showing significant differences with the other treatments, the CF for T₁ presented values lower than 1, suggesting that the fish are immersed in a certain degree of stress under this

treatment. The above could be associated with the non-ionized ammonia concentration, being that in this treatment (T₁), the highest value (1.09 ± 0.18 mg L⁻¹) was obtained for the juvenile stage.

At this stage, all treatments present values higher than 1 for CF, suggesting that there are no stress factors that intervene in the growth of adults. The values corresponding to the GR in the adult stage suggest that a low and medium water exchange rate (T₁ and T₂) favor this variable. On the other hand, the three evaluated treatments effectively contribute to SR with values greater than 92%.

The cortisol concentration had an increase through the developmental stages. In the fingerling stage, despite significant differences between the treatments, all values were within the normal range reported (5-60 ng mL⁻¹) for *Oreochromis niloticus* (Barreto & Volpato 2006). The descending order concentrations for cortisol (T₁ = 10.24 ± 0.85 , T₃ = 8.91 ± 0.93 and T₂ = 6.76 ± 0.59 ng mL⁻¹) coincide with the same order for the concentration of non-ionized ammonia (T₁ = 0.79 ± 0.12 , T₃ = 0.51 ± 0.16 and T₂ = 0.32 ± 0.19 mg L⁻¹). Likewise, the cortisol values were below the upper limit reported (60 ng mL⁻¹). The highest concentration of cortisol in this stage (54.98 ± 5.09 ng mL⁻¹) was observed in T₁ again coinciding with the highest value of non-ionized ammonia (1.09 ± 0.18 mg L⁻¹); therefore, it can be inferred that in the fingerling and juvenile stage the non-ionized ammonia concentration led to an increase in cortisol.

In the adult stage, the cortisol values for the three treatments (T₁ = 110.12 ± 8.64 , T₂ = 139.89 ± 5.81 and T₃ = 153.78 ± 4.34 mg L⁻¹) exceeded the normal range (5-60 ng mL⁻¹), which can be attributed in the first instance to stress related to biomass in culture tanks; being that the density of each treatment was greater

than 40 kg m⁻³ at the time of the treatments measurement ($T_1 = 41.9$, $T_2 = 50.3$ and $T_3 = 46.1$ kg m⁻³). Likewise, a combined effect with the non-ionized ammonia concentration can be considered to explain the variation between treatments.

The glucose concentration increased through the developmental stages, being linked to the increase in cortisol, being that the hormone contributes to the biosynthesis of glucose (Abreu et al. 2009). In the fingerling stage, the highest concentrations were present in T_3 and T_1 (48.09 ± 2.67 and 43.98 ± 1.28 mg dL⁻¹, respectively), treatments that likewise had higher cortisol values. The values of the three treatments were below the reported normal limit (<70 mg dL⁻¹) for *O. niloticus* (Kajimura et al. 2003). Likewise, the values were below that reported by Santiago-Rucínque et al. (2017) and Zeitoun et al. (2017) (67.88 and 52.6 mg dL⁻¹, respectively).

In the juvenile stage, the highest concentration of glucose was present in T_1 (70.67 ± 3.53 mg dL⁻¹), a treatment that in this stage obtained the highest concentration of cortisol. The T_1 value was slightly above the normal limit (<70 mg dL⁻¹), while T_2 and T_3 (56.38 ± 4.05 and 53.72 ± 4.11 mg dL⁻¹, respectively) were below said limit. Likewise, T_2 and T_3 presented values like those reported by Obirikorang et al. (2019) (55.26 and 61.74 mg dL⁻¹, respectively) for a similar water exchange rate; while in the same work by Obirikorang with an exchange rate similar to T_1 , a lower value was reported (53.46 mg dL⁻¹), however, in this experiment, less biomass was managed in the culture tanks.

Finally, in the adult stage, the glucose concentrations for the three treatments were above the normal limit (<70 mg dL⁻¹), directly related to the high cortisol levels for this stage. Likewise, glucose values in adults were above that reported by Martins et al. (2009) (49.5 mg dL⁻¹), where the biomass (57.5 kg m⁻³) and the exchange rate (4.3) were like those of this research. However, the culture period was shorter (57 days), which could explain the lower accumulation of glucose in the blood, considering the concentration of cholesterol and glucose in the different stages of the culture (fingerlings, juveniles, and adults). It is contemplated that the physiological stage of the fish together with the change in population density (9, 39, 60 kg m⁻³ approximately for fingerlings, juveniles, and adults, respectively) and the competition for feed caused the stimulation of the axis hypothalamic-pituitary-adrenal, as well as the expression of genes related to glucose transport, which would explain the rising release of cortisol and glucose (Zeitoun et al. 2017). In this way, in both cases, the metabolic

variation on the different days of culture could be explained.

CONCLUSION

In this research, the effect of the water exchange rate on the different stages of the Nile tilapia (fingerling, juvenile, and adult) was determined. The study provides information that allows considering the exchange rate modification throughout a productive cycle. It is concluded that the exchange rate in a RAS influences the productive performance of each of the stages. In the same way, it affects the levels of cortisol and glucose that indicate the presence of stress factors in the different exchange rates. This situation could be corroborated by measuring antioxidant enzymes (catalase, superoxide dismutase, glutathione peroxidase, and glutathione reductase). The preceding, to enhance growth and reduce as much as possible the presence of stress factors that could reduce the productive performance.

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