




## Influence of saline environment and depuration time on quality and proximate composition of Nile tilapia fillet (*Oreochromis niloticus*)

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### Abstract

This study investigated the depuration time of tilapia in saline and freshwater environments and its influence on animal performance and meat quality. Depuration until the seventh day resulted in significant reductions in average weight, condition factor, viscerosomatic index, hepatosomatic index and fillet yield in both environments. Regarding proximate composition, the saline environment promoted the maintenance of crude protein, moisture and ash percentages. Depuration in the freshwater environment resulted in reduced percentages of crude protein, ash and energy and increased humidity. Total lipids were reduced in both environments, but with a significantly lower in saline. Maximum rigor index occurred in up to three and nine hours for fish depurated in freshwater and saline environments, respectively. During the post-rigor period, the rigor index experienced a sharp drop in the freshwater environment, while in the saline environment it remained similar to that of the pre-depuration group. Depuration in a saline environment for up to seven days was efficient at maintaining of proximate composition and *rigor mortis*, however, depuration until the fifth day is indicated to maintain performance indices.

**Keywords:** starvation; *rigor mortis*; pre-slaughter; salinity; proximate composition.

**Practical Application:** The saline environment can be practical and efficient in the depuration of tilapia.

### 1 Introduction

Fish production and consumption have increased significantly over the years, driven by population growth and increased *per capita* consumption, capitalization and technological development (Food and Agriculture Organization, 2020). Due to their nutritional qualities, fish play an important role in human nutrition (Claret et al., 2014; Sary et al., 2022). Thus, aquaculture has become an alternative for food production and, among the species produced, tilapia stands out with favorable zootechnical characteristics and good acceptance in the world market (Associação Brasileira da Piscicultura, 2020; Albergaria et al., 2022).

Currently, there is increased scientific interest in developing and implementing specific requirements for animal welfare in aquaculture with the aim of improving meat quality (Daskalova, 2019; Segner et al., 2019). The usual strategy for quality control is to transfer affected fish by undesirable compounds into clean water for depuration before marketing (Dionigi et al., 1998; Howgate, 2004). The depuration process was efficient for several species such as in tilapia (*Oreochromis niloticus*) (Guzmán-Guillén et al., 2014), European whitefish (*Coregonus lavaretus*) (Lindholm-Lehto et al., 2019), grass carp (*Ctenopharyngodon idella*) (Lv et al., 2018), Atlantic salmon (*Salmo salar*) (Burr et al., 2012), channel catfish (*Ictalurus punctatus*) (Dionigi et al., 1998).

However, the depuration can take from a few days to weeks depending on several factors and animals are kept with little or no food during depuration (Lv et al., 2018; Moretto et al., 2022). Food deprivation can degrade endogenous energy sources (lipids, glycogen and proteins) in the maintenance of fish physiological homeostasis, leading to weight loss and alteration of the proximate composition (Poli et al., 2005; Li et al., 2018; Sakyi et al., 2020; Barai et al., 2022).

The stress pre-slaughter may influence time of rigor such as rapid depletion of energy source and accelerates *rigor mortis* (Huss, 1995; Matos et al., 2010; Castro et al., 2017). Adequate handling conditions are essential to achieve an acceptable product in terms of the welfare of the fish and the meat quality (Gatica et al., 2008; Mendes et al., 2017). Studies have described reduced stress in fish after handling under isosmotic conditions, as it reduces the need for physiological energy for osmoregulation and the maintenance of homeostasis (Wurts, 1995; Kombat et al., 2021). The use of salt during management routines is a common practice in fish farms, however, few studies have evaluated animals during depuration in a saline environment. Therefore, the present study evaluated the effects of pre-slaughter depuration in saline and freshwater environments on tilapia (*Oreochromis niloticus*) meat quality.

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## 2 Material and methods

### 2.1 Animals and experimental condition

The experiment was performed using adult fish of *O. niloticus* with an average weight of  $552.16 \pm 21.11$  g and an average total length of  $32.1 \pm 2.3$  cm. The animals were reared at the Unidade de Cordeiro of the Fundação Instituto de Pesca do Estado do Rio de Janeiro (FIPERJ), in a  $10 \text{ m}^3$  cage at a density of 50 kg of fish  $\text{m}^{-3}$ . Fish were fed a commercial ration of 4 mm (Pira Evolution TR/Guabi Aqua®), provided twice a day until apparent satiety, for the 30 days prior to the experiment. Water quality parameters of the cultivation water were measured daily during the 30-day period before the experimental tests and were as follows: water temperature  $23.8 \pm 1.2$  °C, pH  $7.27 \pm 0.25$  and dissolved oxygen  $5.73 \pm 0.48$  mg  $\text{L}^{-1}$ . The experimental trials were approved by the Ethics Committee on the Use of Animals CEUA/FIPERJ, Rio de Janeiro, Brazil, protocol 001/2021.

### 2.2 Experiment

Fish feeding was suspended 24 h before the removal of fish from the net cage to empty the gastrointestinal tracts of the animals. After this period, the fish were removed from the net-tank and placed in depuration systems. The experiment was carried out using four depuration times (1, 3, 5 and 7 days) and two environments (saline at a concentration of 7 g of salt  $\text{L}^{-1}$  and freshwater of 0 g of salt  $\text{L}^{-1}$ ). Six depuration systems were used for each environment. Depuration was carried out in recirculation systems with water that was collected from an artesian well, with maturation of the biological filters of the depuration systems for 30 days prior to the tests. Natural sea salt for animal consumption was used for the salinization of the environment (SalMos®, Mossoró-RN, Brasil). Salinity was determined with a portable refractometer (RTS-28/Instrutherm®). A total of 240 fish were captured from the net-tank and randomly distributed among the two depuration systems, (six for each environment). Each depuration system received 20 adult fish, which were initially kept in a useful volume of 180 liters. The systems were connected to a circulation pump (2000 L of water  $\text{h}^{-1}$ ) and equipped with aeration, heaters and biological and mechanical filters.

Two fish were sampled from each depuration system, saline and freshwater, at each of the four depuration times ( $n = 12$  fish/treatment) for the analysis of performance and proximate composition. An additional treatment was performed without depuration of the animals (pre-depuration). Another set of fish was used ( $n = 10$  fish/treatment) for *rigor mortis* analysis, which was performed only with pre-depuration animals and animals with seven days of depuration. As fish were removed from the systems, water was removed to maintain an initial stocking density of 60 kg fish  $\text{m}^{-3}$ . The fish were slaughtered by stunning on ice and then their spinal cord was sectioned. Filleting took place by removing the skin and obtaining the fillet. The fillets were then vacuum packed and kept at  $-20$  °C. Performance and *rigor mortis* were analyzed immediately after slaughter. Water quality was evaluated for the depuration systems of both environments.

### 2.3 Performance indices

Performance indices were evaluated pre- and post-depuration. Average weight, average length, condition factor [CF = total body weight (g)/(total fish length)<sup>3</sup> ( $\text{cm}^3$ )  $\times 100$ ], hepatosomatic index [HSI = (liver weight/total body weight)  $\times 100$ ], viscerosomatic index [VSI = (viscera weight/total body weight)  $\times 100$ ] and fillet yield were analyzed using the method described by Akbary & Jahanbakhshi (2016).

### 2.4 Proximate composition

Crude protein, moisture and ash contents of fillets were determined according to the methods of the Association of Official Analytical Chemists (Association of Official Analytical Chemists, 2010). Crude protein content (N  $\times 6.25$ ) was determined by the micro Kjeldahl method, after acid digestion, followed by ammonia nitrogen distillation (TE-0363, Tecnal®, Brazil) and hydrochloric acid titration (N = 0.02). Crude lipid content was determined by the rapid extraction and purification method proposed by Bligh & Dyer (1959), based on the extraction of fat from a sample (3 g) by adding chloroform, methanol and distilled water, followed by agitation, filtration, drying and weighing. Moisture content was determined by drying oven at 105 °C (410/3NDR, Nova Ética®, Brazil) until constant weight. Ash content was determined by incineration using a digital muffle furnace at 600 °C for 4 h (NT380, Nova Técnica®, Brazil). Gross energy was calculated from the protein and fat composition of fillets; conversion factors of 4.27 kcal/g and 9.02 kcal/g were used for protein and fat, respectively, as described for calculating gross energy in meat products (United States Department of Agriculture, 1987).

### 2.5 Rigor mortis

The rigor index was determined immediately after slaughter during pre-depuration and on the seventh day of depuration in saline and freshwater environments. Measurements were made every 15 min in the first hour and then every hour for up to 9 h, posteriorly, with daily measurements up to 408 h (17 days). The rigor index (RI) was calculated according to the methodology described by Bito et al. (1983) using the following equation:  $RI = (Lo - Lt)/Lo \times 100$ , where  $L$  corresponds to vertical tail drop (cm);  $Lo$  is tail drop at the beginning of the experiment and  $Lt$  is measurements over time, measured using a ruler and a square. Fish tail inclination measurements were performed by placing the fish on the flat surface of a table, supported up to the level of the pelvic fin and leaving the caudal part of the body free. The animals were kept in a cooler with ice during the collection intervals.

### 2.6 Water quality

Water quality parameters were evaluated during depuration. Temperature (°C), electrical conductivity ( $\mu\text{S}/\text{cm}$ ) and pH were measured with a combo multiparameter probe (HI98130/Hanna instruments®); dissolved oxygen (DO) with a multiparameter probe (HI9146-04/Hanna instruments®); and total ammonia and alkalinity with a photometer (HI83200-01/Hanna Photometer®).

## 2.7 Statistical analysis

Data are expressed as mean  $\pm$  standard deviation. The evaluated parameters were submitted to the Cramér-von Mises test for normality of errors and to the Levene's test to verify homoscedasticity of variances. When necessary, the data were transformed into the arcsine square root of the variable expressed as a percentage and submitted to two-way ANOVA followed by Tukey's test ( $p < 0.05$ ). Student's "t" test was applied to the compare pre-depuration group and the depuration groups. Data were processed using the *Statistical Analysis System-SAS* program, version 8.0.

## 3 Results

Results for the water quality parameters are shown in Table 1. Levels of dissolved oxygen, pH, temperature and non-ionized ammonia concentration did not differ significantly ( $p > 0.05$ ) between environments. Electrical conductivity, total dissolved solids and total alkalinity were higher in the saline than in the freshwater environment ( $p < 0.05$ ).

**Table 1.** Water quality parameters (mean  $\pm$  standard deviation) of the depuration systems in saline (7 g of salt L<sup>-1</sup>) and freshwater environments of the average of all the depuration period.

Parameters	Salinity 0 g of salt L <sup>-1</sup>	Salinity 7 g of salt L <sup>-1</sup>
Dissolved oxygen (mg L <sup>-1</sup> )	4.44 $\pm$ 0.29	4.59 $\pm$ 0.40
Electric conductivity ( $\mu$ S cm <sup>-1</sup> )	0.07 $\pm$ 0.01 <sup>B</sup>	11.74 $\pm$ 0.19 <sup>A</sup>
Total dissolved solids (ppt)	0.03 $\pm$ 0.01 <sup>A</sup>	5.87 $\pm$ 0.08 <sup>A</sup>
pH	7.17 $\pm$ 0.20	7.05 $\pm$ 0.37
Temperature (C)	26.10 $\pm$ 1.62	25.08 $\pm$ 0.63
Un-ionized ammonia ( $\mu$ g NH <sub>3</sub> L <sup>-1</sup> )	1.14 $\pm$ 0.79	1.47 $\pm$ 0.50
Total alkalinity (mg CaCO <sub>3</sub> L <sup>-1</sup> )	14.0 $\pm$ 3.2 <sup>B</sup>	31.7 $\pm$ 5.37 <sup>A</sup>

Different uppercase letter in the same line indicates significant difference ( $p < 0.05$ ) between the environments, according to the Tukey's test.

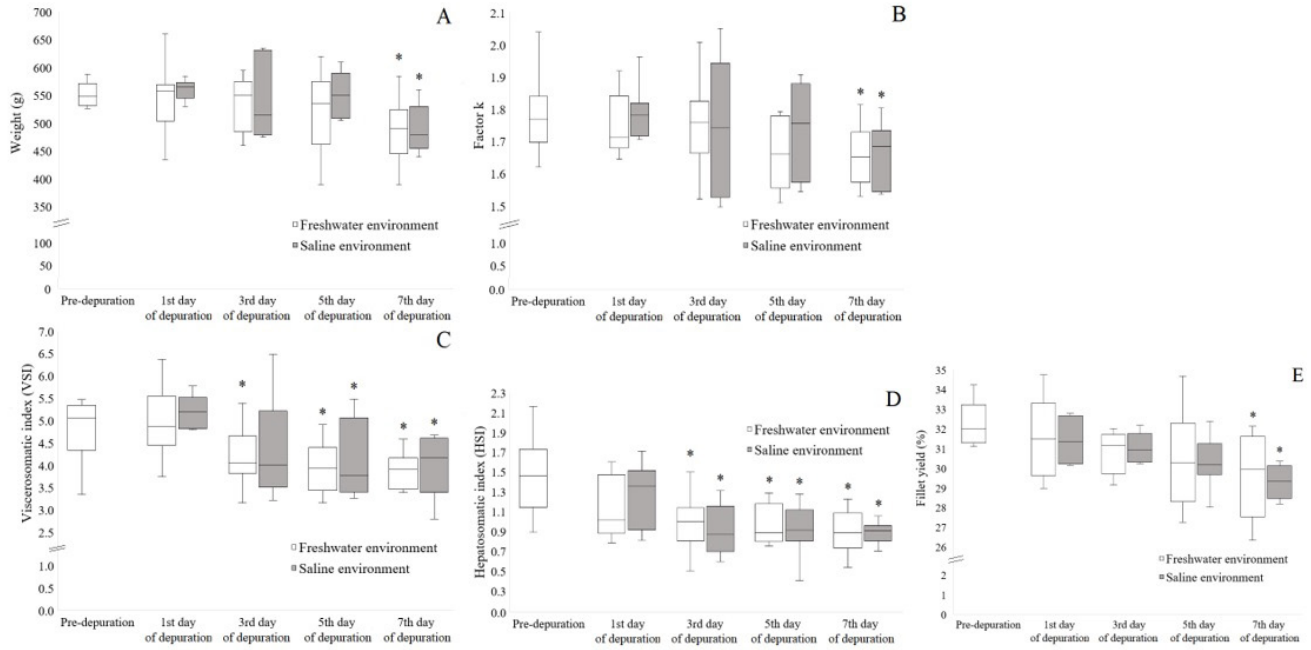
Performance parameters of the animals (weight, length, CF, VSI, HSI and fillet yield) showed no significant differences between saline and freshwater groups (Table 2). However, the animals showed reductions ( $p > 0.05$ ) in weight, VSI and HSI with increasing depuration time. The difference was observed between the pre-depuration group with the seven-day depuration groups of each environment, with a significant reduction ( $p < 0.05$ ) of all performance parameters, except for length (Figure 1). The average weights of animals depurated for seven days reduced from 11.3% to 11.6% in relation to the pre-depuration group, while the difference for fillet yield was reduced from 8.2% to 9.1%. VSI did not differ significantly ( $p > 0.05$ ) between the one-day depuration groups with the pre-depuration group. However, beginning on the third day of depuration, fish in freshwater experienced a significant reduction ( $p < 0.05$ ), while those in the saline environment only experienced a significant reduction after the fifth day. HSI was reduced beginning on the third day of depuration in both environments and differed significantly ( $p < 0.05$ ) from the pre-depuration group. The lowest values ( $p < 0.05$ ) of HSI were observed on the seventh day of depuration.

Results for proximate composition of tilapia fillets are shown in Table 3. Crude protein did not vary as a function of depuration time in the saline environment (Table 3) and did not show any difference in relation to the pre-depuration group (Figure 2). However, there was a significant difference depending on the environment, with freshwater depuration experiencing a reduction in protein levels ( $p < 0.05$ ). Total lipids were influenced by depuration time and environment, as shown in Table 3 and Table 4. Total lipids were gradually reduced ( $p < 0.05$ ) in both depuration environments, with effects being observed from the first day to the seventh day of depuration compared to pre-depuration (Table 4). Beginning on the third day of depuration, the percentage of lipids was significantly higher ( $p < 0.05$ ) for fish depurated in the saline environment than for those depurated in freshwater (Table 4). The lowest percentage ( $p < 0.05$ ) of lipids was observed on the seventh day of depuration; however, with a significantly greater reduction in the freshwater environment compared to the saline environment. There was an increase in the

**Table 2.** Performance of Nile tilapia, *p-values* and means values ( $\pm$  SD) of weight (g), length (cm), factor k, viscerosomatic index (VSI), hepatosomatic index (HSI) and fillet yield (%), in saline and freshwater environments in the depuration period (1, 3, 5 and 7 days).

Statistical	<i>p values</i>					
	Weight	Length	Factor k	VSI	HSI	Fillet yield
Environment (S)	0.46	0.70	0.62	0.77	0.95	0.58
Time of depuration (T)	0.04	0.54	0.10	< 0.01	< 0.01	0.09
Interaction S x T	0.99	0.59	0.97	0.97	0.66	0.99
Treatments	Means for S					
S0(0 g of salt L <sup>-1</sup> )	522.42 $\pm$ 64.65	31.6 $\pm$ 1.7	1.71 $\pm$ 0.14	4.31 $\pm$ 0.78	1.00 $\pm$ 0.26	30.82 $\pm$ 2.10
S7(7 g of salt L <sup>-1</sup> )	534.35 $\pm$ 66.37	31.4 $\pm$ 1.7	1.73 $\pm$ 0.14	4.36 $\pm$ 0.89	1.01 $\pm$ 0.28	30.53 $\pm$ 1.19
	Means for T					
T1 (1 <sup>st</sup> Day of depuration)	553.57 $\pm$ 49.86 <sup>a</sup>	31.8 $\pm$ 1.7	1.77 $\pm$ 0.09	5.04 $\pm$ 0.69 <sup>a</sup>	1.20 $\pm$ 0.31 <sup>a</sup>	31.52 $\pm$ 1.76
T3 (3 <sup>rd</sup> Day of depuration)	538.85 $\pm$ 59.52 <sup>ab</sup>	31.4 $\pm$ 1.7	1.75 $\pm$ 0.17	4.26 $\pm$ 0.83 <sup>b</sup>	0.96 $\pm$ 0.25 <sup>b</sup>	31.05 $\pm$ 1.41
T5 (5 <sup>th</sup> Day of depuration)	529.61 $\pm$ 69.87 <sup>ab</sup>	31.7 $\pm$ 2.0	1.72 $\pm$ 0.17	4.02 $\pm$ 0.73 <sup>b</sup>	0.95 $\pm$ 0.21 <sup>b</sup>	30.41 $\pm$ 1.88
T7 (7 <sup>th</sup> Day of depuration)	488.93 $\pm$ 53.28 <sup>b</sup>	31.0 $\pm$ 1.3	1.66 $\pm$ 0.10	3.94 $\pm$ 0.58 <sup>b</sup>	0.89 $\pm$ 0.17 <sup>b</sup>	29.53 $\pm$ 1.81

N = 12 fish/group. Statistical analysis used was two-way ANOVA for interaction between environment and time of depuration. Different lowercase letter indicates significant difference between the groups in the same column, according to the Tukey's test ( $p < 0.05$ ).



**Figure 1.** Performance of Nile tilapia between the pre-depuration and depurated groups in saline (7 g of salt L<sup>-1</sup>) and freshwater (0 g of salt L<sup>-1</sup>) environments in the depuration period (1, 3, 5 and 7 days). (A) Weight; (B) Factor k; (C) VSI; (D) HSI; (E) Fillet yield. N = 12 fish/group. Asterisk (\*) indicates significant difference (*p* < 0.05) of each depurated group in relation to the pre-depuration group, according to the Student's "t" test.

**Table 3.** Proximate composition of Nile tilapia fillet, p-values and means values (± SD) of crude protein (%), crude fat (%), moisture (%), ash (%) and energy kcal 100 g<sup>-1</sup>, in saline (7 g of salt L<sup>-1</sup>) and freshwater environments in the depuration period (1, 3, 5 and 7 days).

Statistical	<i>p</i> values				
	Crude Protein	Crude fat	Moisture	Ash	Energy kcal
<b>Environment (S)</b>	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01
<b>Time of depuration (T)</b>	0.51	< 0.01	0.35	0.12	< 0.01
<b>Interaction S x T</b>	0.93	< 0.01	0.55	0.55	0.17
Treatments		Means for S			
<b>S0 (0 g of salt L<sup>-1</sup>)</b>	17.88% ± 0.76% <sup>b</sup>	0.63% ± 0.21% <sup>b</sup>	80.31% ± 1.00% <sup>a</sup>	0.99% ± 0.04% <sup>b</sup>	81.59 ± 4.3 <sup>b</sup>
<b>S7 (7 g of salt L<sup>-1</sup>)</b>	18.64% ± 0.88% <sup>a</sup>	0.80% ± 0.12% <sup>a</sup>	79.27% ± 1.12% <sup>b</sup>	1.06% ± 0.05% <sup>a</sup>	86.24 ± 3.3 <sup>a</sup>
		Means for T			
<b>T1 (1<sup>st</sup> Day of depuration)</b>	18.51% ± 0.54%	0.91% ± 0.10% <sup>a</sup>	79.30% ± 1.22%	1.01% ± 0.05%	87.30 ± 2.6 <sup>a</sup>
<b>T3 (3<sup>rd</sup> Day of depuration)</b>	18.35% ± 0.92%	0.72% ± 0.11% <sup>b</sup>	79.99% ± 1.23%	1.02% ± 0.07%	84.26 ± 3.6 <sup>ab</sup>
<b>T5 (5<sup>th</sup> Day of depuration)</b>	18.16% ± 0.96%	0.66% ± 0.17% <sup>bc</sup>	79.96% ± 1.18%	1.00% ± 0.07%	82.55 ± 4.2 <sup>b</sup>
<b>T7 (7<sup>th</sup> Day of depuration)</b>	18.02% ± 1.12%	0.55% ± 0.15% <sup>c</sup>	79.98% ± 1.05%	1.05% ± 0.05%	81.53 ± 5.4 <sup>b</sup>

N = 12 fish/group. Statistical analysis used was two-way ANOVA for interaction between environment and time of depuration. Different lowercase letter indicates significant difference between the groups in the same column, according to the Tukey's test (*p* < 0.05).

**Table 4.** Interaction (S × T) means values (± standard deviation SD) of crude fat (%), in saline (7 g of salt L<sup>-1</sup>) and freshwater environments in the depuration S period (1, 3, 5 and 7 days).

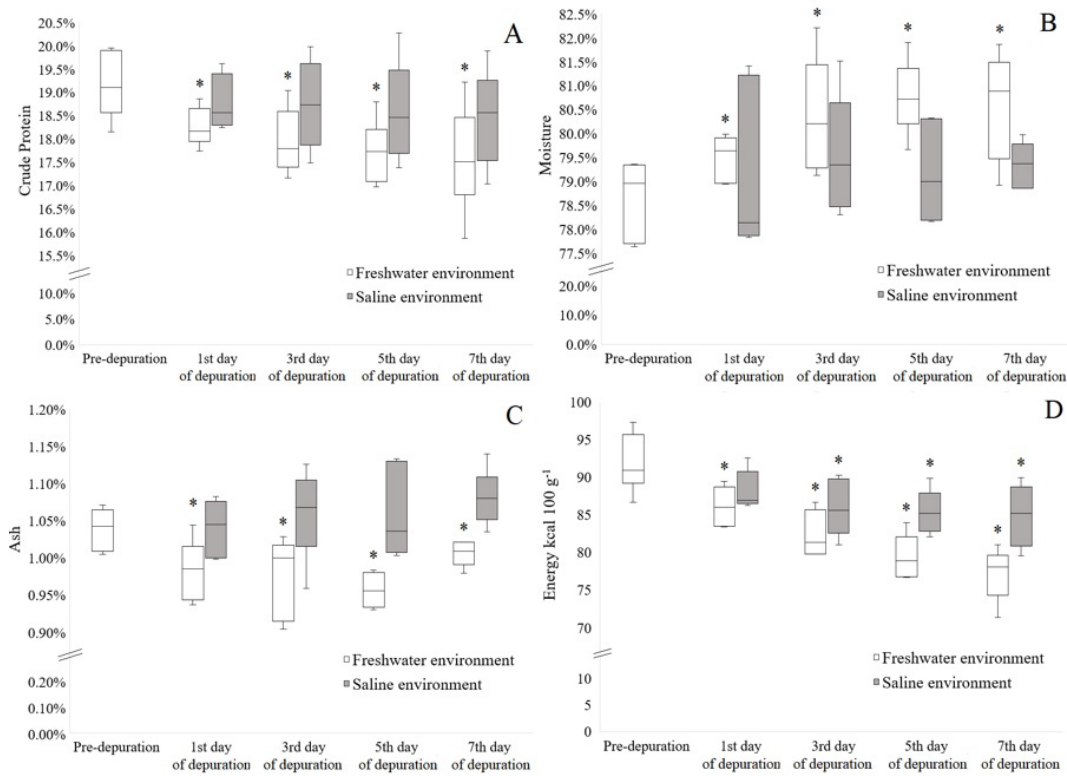
Parameter	Pre depuration	1 <sup>st</sup> Day of depuration	3 <sup>rd</sup> Day of depuration	5 <sup>th</sup> Day of depuration	7 <sup>th</sup> Day of depuration
<b>Crude fat (0 g of salt L<sup>-1</sup>)</b>	1.12% ± 0.14%	0.92% ± 0.12% <sup>A*</sup>	0.65% ± 0.07% <sup>Bb*</sup>	0.51% ± 0.07% <sup>Cb*</sup>	0.42% ± 0.04% <sup>Cb*</sup>
<b>Crude fat (7 g of salt L<sup>-1</sup>)</b>		0.91% ± 0.08% <sup>A*</sup>	0.80% ± 0.10% <sup>ABa*</sup>	0.81% ± 0.06% <sup>ABa*</sup>	0.69% ± 0.09% <sup>Ba*</sup>

N = 12 fish/group. Different lowercase letter indicates significant difference between the groups in the same column, according to the Tukey's test (*p* < 0.05). Different uppercase letter indicates significant difference between the groups in the same line, according to the Tukey's test (*p* < 0.05). \*Significant difference (*p* < 0.05) of each depurated group in relation to the pre-depuration group, according to the Student's "t" test.

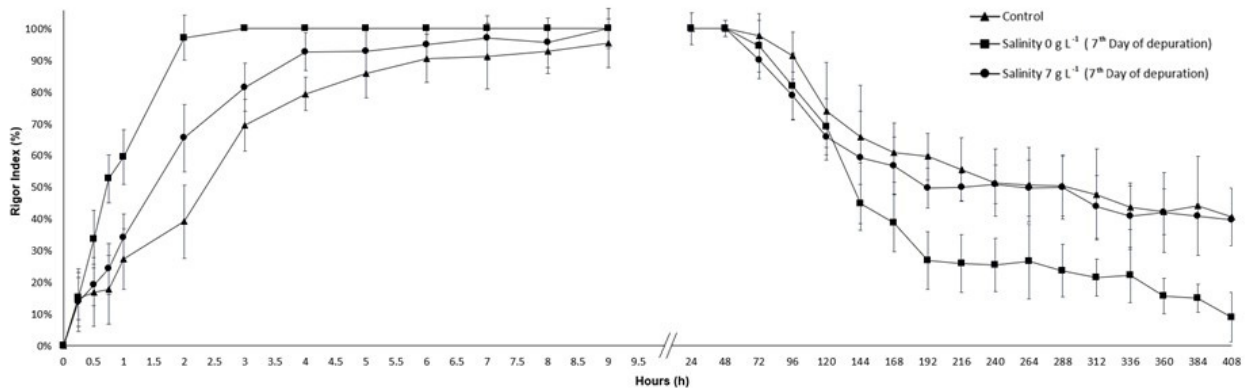
moisture content ( $p < 0.05$ ) in fish depurated in the freshwater environment (Table 3). In comparison to the pre-depurated group with the animals depurated in saline environment did not differ significantly ( $p > 0.05$ ) in moisture content with the depuration time (Figure 2). The percentage of ash reduced significantly ( $p < 0.05$ ) in fish depurated in the freshwater environment in relation to the pre-depurated and saline groups. The percentage of ash in animals depurated in the saline environment did not vary as a function of depuration time and without a significant difference in relation to the pre-depurated group ( $p > 0.05$ ). There was a significant difference in gross energy between fish depurated in

saline and freshwater environments, with the highest energy being maintained in fish of the saline environment ( $p < 0.05$ ). Energy was influenced by depuration time in both environments, with a reduction throughout the depuration period. Compared to the pre-depurated group, energy contained in tilapia fillets was reduced on the first day of depuration in the freshwater environment and on the third day of depuration in the saline environment.

Pre-rigor was observed during the first nine hours and post-rigor until the 17th day (Figure 3). Rigor was evaluated between the seven-day depuration groups and the pre-depurated group. A rigor index of 100% occurred in up to three hours for fish



**Figure 2.** Proximate composition of Nile tilapia fillet between the pre-depurated and depurated groups in the period (1, 3, 5 and 7 days). (A) Crude protein; (B) Moisture; (C) Ash; (D) Energy.



**Figure 3.** Relationship between percentage of rigor index in the pre-depurated and after seventh day of depuration in saline (7 g of salt L<sup>-1</sup>) and freshwater environments in the period 408 h. N = 10 fish/group.

depurated in the freshwater environment while a longer period was observed for fish of the pre-depuration group, which reached a rigor index of 95% in up to nine hours. A rigor index of 100% occurred in up to nine hours for fish depurated in the saline environment. All groups presented the maximum rigor index during the periods of 24 and 48 h. The post-rigor period began after the 48-hour period in all groups. A difference between groups was observed at 144 h (6 days) with rigor indices of 65.8%, 59.3% and 44.7% for pre-depuration, saline and freshwater groups, respectively. At the end of 408 h (17 days), the rigor index was similar between the pre-depuration group and fish depurated in the saline environment with 40.6% and 39.7%, respectively; the lowest percentage observed for depuration in freshwater was 9.0%.

#### 4 Discussion

Most of the evaluated water quality parameters remained within the range considered adequate for the maintenance of tropical fish species (Arana, 2004). Oxygen, pH, temperature and un-ionized ammonia levels did not differ significantly between environments. The saline environment provided an increase in electrical conductivity, total dissolved solids and total alkalinity by the dissociation of salts contained in the sea salt. Cheng et al. (2022) suggested that environmental salinity and alkalinity effectively improve tilapia meat quality by regulating lipid and amino acid metabolism pathways. The concentrations of 8 to 9 g of L<sup>-1</sup> salt correspond to the sodium content in the blood of fish (Wurts, 1995). Tilapia show lower energy expenditure for osmoregulation when kept at salinities close to or equivalent to their isosmotic point (Febry & Lutz, 1987).

Little or no food is provided to fish during depuration. Food deprivation can induce the mobilization of energy reserves to maintain physiological homeostasis, leading to weight reduction (Zheng et al., 2015). Dias et al. (2016) reported that red tilapia (*Oreochromis* sp.) lost 18% to 45% of body weight in 7 to 200 days of fasting respectively, and seven days of fasting seems to be sufficient for complete emptying of the tilapia intestine, since the reduction in stomach and intestine weight stabilized after this period. In fact, the results of the present study showed that seven days of depuration is a critical time period for the species, even in a saline environment, with a significant reduction in performance indices, with direct impacts on fillet yield and animal welfare by reducing the condition factor. Results of the present study for the viscerosomatic index showed that energy reserves were saved in the saline environment, being significantly modified beginning on the fifth day of depuration, unlike depuration in the freshwater environment whereby the reserves were mobilized on the third day, compared to the pre-depuration group.

The results for proximate composition reflected the performance indices and the influence of salt in the depuration process. Han et al. (2011) observed that food deprivation of hybrid tilapia juveniles (*O. niloticus* x *O. aureus*) for four weeks resulted in a reduction of crude protein and fat percentages throughout the body, while moisture and crude ash increased significantly. In contrast, a slight increase in salinity did not affect the nutritional value of grass carp muscle protein (Lv et al., 2018).

Protein stores are spared early during fasting, so proteolysis occurs only when the most readily available energy stores have been amply consumed (Nebo et al., 2018). Periods of food deprivation induce changes in storage reserves in fish, mainly of lipids (Ali et al., 2003). Lipids are often the main source of energy for maintaining fish during winter fasts (Bull & Metcalfe, 1997). There is an inverse relationship between lipid and water content, with catabolized lipids being replaced by an equal volume of water and, under moderate starvation conditions, body weight is maintained through water absorption (Navarro & Gutiérrez, 1995). According to United States Department of Agriculture (2021), raw tilapia has a reference of 96 kcal per 100 grams, which was close to the 91.9 kcal per 100 grams found here for pre-depuration. The present study provided evidence that depuration in a saline environment allowed a better maintenance of proximate composition and energy reserves of the tilapia fillet.

Meat quality and fish welfare can be indicated by *rigor mortis* analysis. According to Huss (1995), tilapia enter maximum rigor in up to 9 h and in up to 1 h under stress conditions. Matos et al. (2010) reported that the onset of *rigor mortis* for sea bream (*Sparus aurata*) was delayed with the use of anesthetics, reaching the highest score at 21 h post-mortem, in contrast to stressed fish that reached rigor at 2 h. Severe stress at the time of slaughter depletes muscle energy, produces more lactic acid, reduces muscle pH, and increases the rate of *rigor mortis* onset (Poli et al., 2005). The results of the present study showed that depuration in the freshwater environment promoted greater depletion of reserves and rapid onset of rigor, while depuration in the saline environment minimized the onset of *rigor mortis*, thus prolonging the pre-rigor period.

At the beginning, pre-rigor, fish muscle is fully relaxed and has a flexible and elastic texture (Huss, 1995). Pre-rigor filleting may be associated with slower bacterial growth, resulting in increased shelf life (Tobiassen et al., 2006). However, *rigor mortis* occurs when muscle extension disappears through the coupling of actin and myosin, characterized by the depletion of ATP and creatine phosphate, producing stiffness of muscle fibers (Wang et al., 2019). In *rigor mortis*, the fish body becomes rigid, making it difficult to obtain clean cuts in processing, thus reducing filleting yield and causing muscle fibers to easily break (Huss, 1995). Post-rigor corresponds to the maturation of the meat in the process that involves the weakening and degradation of Z discs and the degradation of desmin and titin, leading to the destabilization of myofibrils. Among the main enzymes involved in post-mortem proteolysis are calcium-activating calpains and lysosomal cathepsins (Nowak, 2011). At this stage, fish muscle becomes flexible, but not as elastic as before *rigor mortis* (Delbarre-Ladrat et al., 2006). Furthermore, cathepsin is less active in the presence of ATP and strongly inhibited by 5% chloride (Reddi et al., 1972). The resolution of *rigor mortis* was less accentuated in the depuration in saline environment and may be related to the influence of alkalinity in the maintenance of the enzymes involved in proteolysis.

#### 5 Conclusion

Depuration in a saline environment for up to seven days was efficient at maintaining meat quality with regard to proximate

composition and *rigor mortis*, however, depuration until the fifth day is indicated to prohibit change in animal weight. Future studies will be conducted with the objective of evaluating the color, texture, freshness, sensorial aspect and especially the lipid profile of Nile tilapia fillet that occur during depuration in a saline environment.

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