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Comparative study of growth and survival of Nile tilapia (*Oreochromis niloticus*) and Three Spotted tilapia (*Oreochromis andersonii*) overwintered in concrete ponds in Zambia

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DOI 10.53974/unza.jabs.8.3.1289

ABSTRACT

This study aims to provide information on the growth of tilapia in winter nurseries. This will help alleviate the adverse effects of the cold season on aquaculture in Zambia. In this study, *Oreochromis niloticus* (T1) and *Oreochromis andersonii* (T2), with initial mean weights of 2.12 ± 1.03 and 2.20 ± 1.03 g respectively were used. The fingerlings were stocked in six concrete ponds (7×5 m) at 6 fish/m² (180 fish/pond), in three replications, which resulted in a total of 1080 experimental fish. After 12 weeks of culture, the average weight gain (AWG) was 24.37 ± 3.67 g for *O. niloticus* and 20.49 ± 1.50 for *O. andersonii*. There were no significant differences ($P > 0.05$) in the final weights (FW), average weight gain (AWG), specific growth rate (SGR), and survival rates of the two species. However, the food conversion ratio (FCR) for *O. niloticus* of 1.07 ± 0.11 was significantly better ($P < 0.05$) than for *O. andersonii* of 1.62 ± 0.10 . The findings showed that *O. andersonii* can exhibit similar attributes as *O. niloticus* in terms of growth and survival in semi-intensive culture and by the end of the cold season, both species were overwintered to weights suitable for grow-out culture. Overwintering of late spawn tilapia may help provide ready-to-stock fingerlings at the onset of the fish growing season in Zambia

performance, Oreochromis andersonii, Oreochromis niloticus, Zambia

INTRODUCTION

Zambia produced 159,043 metric tons of fish in 2021, a 22.5 percent increase compared to the 129,822 metric tons produced in 2019. Most of the fish in this production were tilapia species from aquaculture and fisheries. Fish production from aquaculture increased from 38,000 metric tons in 2019 to 59,679 metric tons in 2021, a 57.1 percent increase (MOFL, 2022). Described as the "Aquatic Chicken" (Maclean 1984), the Nile tilapia (*Oreochromis niloticus*) is the most commonly farmed species of tilapia in the world. Because of its high fertility, rapid development rate, and wide tolerance to environmental conditions, Nile tilapia has become a popular aquaculture species (Zale & Gregory, 1989; Avella et al., 1993). Although these traits are advantageous for aquaculture, they also increase the invasiveness of the species (Costa Pierce, 2003; Zengeya, 2015; Deiness et al., 2014).

Nile tilapia is regarded as a non-indigenous species in Zambia, and legislation attempts to limit its spread to the Kafue and Zambezi basins, where it developed a niche population following its escape from aquaculture facilities in the mid-1990s (Schwank, 1995). The latter reflects the application of a biodiversity approach that avoids invasiveness by restricting aquaculture species use to watersheds where they are either native or otherwise endemic (Hoevenaars and Ng'ambi, 2019; WorldFish & AbacusBio, 2021).

KEYWORDS: *Over-wintering, Growth*

The Three spotted tilapia (*Oreochromis andersonii*), Green-headed Tilapia (*Oreochromis macrochir*), Red-breasted Tilapia (*Coptodon rendalli*), and Tanganyika Tilapia (*Oreochromis tanganyicae*) are among the native tilapia species raised in Zambia (Zhang et al., 2024). *O. andersonii* commonly known as Kafue bream or Njinji (Fishbase, 2024), is the most preferred fish to raise in areas where *O. niloticus* is prohibited because the species is considered to have strong aquaculture production potential within Zambia (Cayron-Thomas, 2010; Musuka & Musonda, 2012; Kefi & Mwango, 2018). The Zambian government recently launched a genetic improvement initiative (GIP) for *O. andersonii* with assistance from WorldFish, which serves as a technical advisor (WorldFish & AbacusBio, 2021).

The growth performance of *O. andersonii* compared to *O. niloticus* is not well documented. While certain research investigations (Cayron-Thomas, 2010; Kefi & Mwango, 2018; Mupeyo, 2005) find comparable growth performance, others (Day et al., 2016; Basiita et al., 2022) report greater growth performance for *O. niloticus*. Although Simataa & Musuka (2013) did not perform a comparative growth analysis, they did indicate that the withdrawal of *O. niloticus* from aquaculture negatively impacted small-scale farmers in Zambia, in part because other farmed tilapia species did not grow as fast as *O. niloticus*.

This study aimed to compare the overwintering growth performance and survival of *O. andersonii* and *O. niloticus* fingerlings cultured in concrete ponds. The study will offer essential information on the early growth of tilapia fingerlings in the cold season, which can be utilized in winter nurseries to help mitigate the recurring shortage of good-quality fingerlings at the commencement of the aquaculture season in Zambia. Additionally, the study will help establish a baseline against which genetically enhanced strains of *O. andersonii* can be evaluated (WorldFish & AbacusBio., 2021).

MATERIALS AND METHODS

Study area

The study was carried out at a private farm in Ngwerere, Lusaka (15°17'39.70"S, 28°18'39.84"E), for 12 weeks (84 days)

during winter from June to August 2024. The experiment was conducted in six tarpaulin-lined concrete ponds (7 m× 5 m× 1.2 m). Water was maintained at 70 cm depth, and the ponds were covered with greenhouse plastic to mitigate against the cold temperature.

Experimental design

The experimental fish were monosex Nile tilapia (*Oreochromis niloticus*) (T1) and Three spotted tilapia (*Oreochromis andersonii*) (T2), acquired from Alia Aqua Hatchery in Lusaka. The fingerlings were acclimatized for two weeks before being stocked at an initial weight of 2.12 ± 1.03 and 2.20 ± 1.03 g for T1 and T2, respectively. The fingerlings were stocked in 6 concrete ponds at 6 fish/m² (180 fish/pond), in three replications, which resulted in a total of 1080 experimental fish. A biweekly sampling of about 30% of stocked fish from each experimental pond was done by using a seine net. The sampled weight was used to adjust the feeding rate for the next fortnight. The weights of sampled fish were recorded by using an electronic balance (Model SF-400).

Experimental Feed

The experimental fish diet (sinking pellets) was formulated with 33% crude protein (Table 1). The feed ingredients were purchased from the local markets in Lusaka. The feed ingredients were weighed and mixed from the University of Zambia, Animal Science Laboratory. The feed was then pre-cooked by adding hot water to it making a paste and was then pelleted using a hand mincer No. 22 with a 2mm screen and the pellets were left to sun dry for three days. The pellets were then kept in air-tight labelled bags and stored at room temperature. Proximate analysis of the formulated feed was done at Livestock Services, located at the Lusaka showgrounds. Feeding was done at 5% of the total biomass of the fish twice a day at 10:00 and 14:00 hrs every day.

Water quality parameters

Water temperature, dissolved oxygen (DO), pH, electrical conductivity, total dissolved salts (TDS) in the experimental ponds were monitored three times daily at 6:00 am, and afternoons at 12:00 and 6:00 pm. The DO, temperature, and TDS of water were measured using a multi-functional water quality meter. The Secchi disc was used to determine turbidity while pH, alkalinity, ammonia, nitrites and nitrates were monitored using test strips. Mortality was checked and recorded daily, and dead fish were removed immediately from the

ponds.

Determination of growth, survival and feed utilization parameters

Fish growth performance was assessed based on body weight gain (BWG), average daily gain (ADG), specific growth rate (SGR), feed conversion ratio (FCR), and survival rate (SR). The calculations used to evaluate these parameters are provided below:

- Body Weight Gain (BWG) : is the total body weight increase (in g) of the fish during the trial, where Wf is the mean final body weight and W0 is the mean initial body weight.

$$BWG [g] = Wf [g] - W0 [g]$$

- Percent weight gain (PWG) % = $(Wf [g] - W0 [g]) / W0 [g] \times 100$

- Average daily gain (ADG): is a measure of how much an animal's weight increases each day over a specific period of time.

$$ADG [g] = (Wf [g] - W0 [g]) / t \text{ (days)}$$

- Specific Growth Rate (SGR): is the growth rate given as % increase per day with t as number of feeding days. It is a good growth measure for young fish, as their weight gain is still in exponential phase.

$$SGR (\%) = (\ln Wf [g] - \ln W0 [g]) / t \text{ (days)} \times 100$$

$\ln Wf$ = the natural logarithm of the final weight

$\ln W0$ = the natural logarithm of the initial weight

t = time (days) between $\ln Wf$ and $\ln W0$

- Feed Conversion Ratio (FCR): is the ratio between Feed Intake (FI) and fish body weight gain, where BWG is the total body weight gain over the trial time.

$$FCR = FI [g] / BWG [g]$$

- Survival rate (SR): is the ratio between total number of living fish at end of the trial (Nt) and living fish at the beginning of the trial (N0).

$$SR (\%) = Nt / N0 \times 100$$

Data Analysis and Management

The weights of the fish obtained at the end of the experiment were used to calculate the body weight gain (BWG), specific growth rate (SGR), and survival rate. The collected data were subjected to a one-way analysis of variance (ANOVA) to compare the treatment means at a 5% significance level using the Statistical Package for Social Sciences (SPSS) version 16. Microsoft Excel was used

to create graphs and figure.

RESULTS

Table (2) provides information on the effects of overwintering of *O. niloticus* (T1) and *O. andersonii* (T2) in concrete ponds on growth parameters, including feed conversion ratio (FCR), weight gain (g and%), total weight gain (WG g), daily gain (DWG g/fish/day), and specific growth rate (SGR %/day). Figs. 1 – 4 graphically depict these effects. Physio-chemical parameters during the experimental period are presented in Table 2 and Figs. 6 and 7 graphically depicts the effects of temperature during the study. Table 2: Growth parameters of *O. niloticus* (T1) and *O. andersonii* (T2) after 84 days of culture represented as Mean \pm Standard Deviation.

Fish Growth Performance

Final Weight

O. niloticus (T1) and *O. andersonii* (T2) had mean final weight (FW) gains of 25.21 ± 10.38 g and 22.57 ± 8.44 g, respectively (Table 1). The starting and final weights of T1 and T2 did not differ significantly ($P > 0.05$). T1 and T2 had mean final length gains of 10.99 ± 1.57 cm and 10.83 ± 1.55 cm, respectively, with no significant differences ($P > 0.05$).

Weight Gain

The average weight gain of the tilapia was 20.49 ± 1.50 g for T2 and 24.37 ± 3.67 g for T1 (Table 1). The two treatments performed similarly in terms of weight gain over almost all of the sampling phases, but by week 12, T1 had overtaken T2, (Fig. 1), but without any significant difference ($P > 0.05$) between the treatments. The spider diagrams for weight and length (Fig. 3) illustrates the growth pattern of the two species. The fish had similar distribution of weight but in terms of length, T2 was slightly longer until week 12.

Percent Weight Gain (%)

The mean percent weight gains of fish were 1155 ± 159.46 % and 936.41 ± 67.50 % for T1 and T2, respectively, with no significant differences ($P > 0.05$) among the treatments.

Specific Growth Rate (SGR % per day)

The specific growth rates (SGR) of tilapia in T1 and T2 were found to be 3.12 ± 0.32 % and 2.80 ± 0.07 % respectively with no significant difference ($P > 0.05$) between the treatments.

Feed Conversion Ratio (FCR)

The total amount of feed utilized in the experiment was accounted for when calculating the feed conversion ratio. T1 and T2 had respective FCRs of 1.07 ± 0.11 and 1.62 ± 0.10 . There was a significant difference ($P < 0.05$) between T1 and T2.

Total Production (g)

The survival rates for T1 and T2 were $81.33 \pm 14.5\%$ and $66.67 \pm 0.58\%$, respectively, and there were no significant differences between the treatments ($P > 0.05$). Total production at the end of the study (after 84 days) was 10.31 ± 0.67 Kg and 8.24 ± 0.06 Kg in T1 and T2, respectively with no significant differences among the treatments ($P > 0.05$).

Weight and length variation

Based on their final weight range, the fish were divided into six categories: 1–10 g, 11–20 g, 21–30 g, 31–40 g, 41–50 g, 51–60 g, and 61–70 g. The size difference between the two species after 12 weeks of culture is depicted in Fig. 4 below. The weight distribution of the two species was comparable, with around 40 and 46.3 percent T1 and T2, falling between 21 and 30 grams.

The length distribution of T1 and T2 at the end of the culture period was comparable (Fig. 5). Fifty-eight (58) percent of T1 and 46.4 percent of T2 had ultimate lengths between 11 and 12 centimetres.

Water Quality Parameters

At $22.12 \pm 1.68^\circ\text{C}$, the average daily temperature was 5.12 degrees warmer than the typical winter temperature of open ponds, which was estimated to be about 17°C (Cayron-Thomas, 2010). The ponds were covered with greenhouse plastic, which helped to keep the water temperatures stable during the winter. Temperatures were on average $16.88 \pm 2.8^\circ\text{C}$ in the morning (6 hours), $23.63 \pm 2.02^\circ\text{C}$ in the afternoon (12 hours), and $26.0 \pm 2.79^\circ\text{C}$ in the evening (18 hours). Fig. 6 illustrates how the average morning temperatures decreased as August approached, but the afternoon and night-time temperatures higher. There was no correlation between the culture period and average temperature during (Fig. 7; $R^2 = 0.04$), though there was a slight drop in temperature towards August.

DISCUSSION

After 12 weeks (84 days) of culture, *O. niloticus* (T1) and *O. andersonii* (T2) had final weights (FW) of 25.21 ± 10.38 and 22.57 ± 8.44 grams, respectively. The body weight gains (BWG) were of 24.37 g and 20.49 g, respectively, with weight gain percentages of 1155% and 936% from an initial weight of 2.12 ± 1.03 and 2.20 ± 1.03 grams, respectively. These figures are quite low when compared to Ahmed et al., 2023, who found that after 70 days of culture, Nile tilapia grew from an initial weight (IW) of 15.9 grams to 123 and 111 grams, with weight gain percentages of 12348% and 11181%. Rahman et al., 2023, achieved final weights of 50.70 and 69.24 grams for mixed and mono-sex tilapia respectively, in 90 days from an initial weight of 0.80 and 1.12 respectively.

According to research, tilapia's weight gain might vary greatly depending on the initial weight at stocking. Larger fish typically perform better in terms of growth and smaller fish may perform better under particular circumstances, as seen by the higher percentages of weight increase and specific growth rates that smaller fish can achieve. Assessing growth outcomes depending on stocking size requires careful consideration of certain environmental conditions and management approaches (Shamsaddin et al 2022). Additionally, the genetic makeup of tilapia strains influences their growth performance, which differs depending on the geographic area (Hossain, 2015). The body weight gain (BWG) of the *O. andersonii* strain used in this study was better than that of the strains reported by DOF 2020 after 14 weeks of culture; Zambezi (13.87 ± 0.28 g), Kalimba (13.11 ± 0.31 g) and Luangwa (4.99 ± 0.5 g). *O. andersonii* showed remarkable growth performance and was only overtaken by *O. niloticus* at about 12 weeks of culture. The percent distribution of weights and the growth patterns were similar, showing that *O. andersonii* has the potential to perform as well as *O. niloticus* in culture, which is in agreement with other authors (Cayron-Thomas, 2010; Kefi & Mwangi, 2018).

The Average daily gain (ADG) of T2 (0.244 g d⁻¹) was comparable to that of Day et al., (2016) and Basiita et al., (2022), who reported ADGs of 0.185 and 0.385 g d⁻¹, respectively. However, in comparison to other authors (Basiita et al. 2022; Day et al. 2016; Bahnasawy et al. 2003; 0.9 g d⁻¹); and others, the ADG value for T1 (0.292 g d⁻¹) in this study was very low. Several researchers investigating tilapia growing in fertilized earthen ponds achieved significantly higher ADG values (Diana et al., 1995; El-Shebly., 1998; Brown et al.,

2002; Yi et al. 2002b). Diana et al., (1995) fed the fish (starting body weight: 15 g) to satiation, to obtain ADG values ranging from 1.6 to 3.04 g d⁻¹. Using tilapia weighing between 33.0 and 66.5 g and providing supplemental food (25% crude protein) twice a day in addition to fertilization, El-Shebly (1998) achieved higher ADG values (1.56 - 3.47 g d⁻¹). The higher initial weight of the stocked fish and the higher quality of the supplemental feed could have caused the higher ADG of fish reported by the authors cited above. These findings may have implications on the profitability of pond fish culture in Zambia since most small-scale fish farmers stock fry (1 gram) in grow-out ponds.

The present study's SGR values (3.12 % and 2.8 % for T1 and T2, respectively) are significantly higher than the 1.40 – 1.81 % values for overwintering mono-sex tilapia fry reported by Dan and Little (2000b). In contrast to the current work, Dan and Little's (2000b) reduced SGR may have resulted from lower temperature (11.0 to 23.0°C). Diana et al. (1996) found that the SGR of *O. niloticus* using feed and fertilizer in Thailand was 3.10%, while Green (1992) found that the SGR of tilapia using feed and fertilizer in Honduras was 2.03%. For GIFT fed with formulated feed (30.09% protein), Hossain et al., (2004) estimated the SGR to be 2.04 - 2.03% in Bangladesh. Ahmed et al., (2013) found that the SGR of monosex tilapia was 3.09% using prepared feed (55.24% protein) and 2.97% using commercially available feed. Body weight determines SGR, which was first shown to follow a negative potential function in its declining pattern (Jobling, 1983). In addition, SGR is temperature dependent because fish are ectothermic, yet temperature is not mentioned in the mathematical expression of SGR (Márquez et al., 2024). As the SGR and FCR in this study are consistent with the findings of other authors in other regions, we can say that the study's overwintering temperature and management settings were within a suitable range for both T1 and T2. However, the fish could not reach optimal growth because the average morning temperature was very low. This is further discussed under water quality.

In this investigation, the FCRs for T1 and T2 were 1.07 ± 0.11 and 1.62 ± 0.10 , respectively. Comparable FCR values have

been found for the same species (Basiita et al., 2022). While Day et al., 2016, found a higher value (2.53) for T2, they also found similar results for T1. According to Rana and Hassan (2013), the reported FCR values for tilapia range greatly, from 1.0 to 1.71 in cage habitats and from 1.5 to 2.5 in pond environments. In freshwater and saltwater pond settings, Thoa et al. (2016) found that the FCR values were 1.08 and 1.89, respectively. Fertilized ponds with supplementary feeding had FCRs of 1.08 to 1.60, according to Diana et al., (1994). FCR is acceptable if it is below the recommended limit of 2 (Craig 2009). In this investigation, the FCR of both T1 and T2's FCRs were within the permissible range. Despite this, the FCR of T2 was affected by the quantity of recorded mortality. Mortality and individual variations in fish's ability to convert feed into biomass are the main factors influencing FCR at the production unit level. These factors are heavily impacted by the environment (de Verdal et al. 2018). *O. niloticus* showed better FCR during this study due partly to its vigorous feeding habits compared to *O. andersonii*.

The mean survival for T1 and T2 in this investigation was 81.33 ± 14.5 % and 66.67 ± 0.58 %, respectively. The current study's fish survival (%) is better than the 33-54 % survival rate for mono-sex over-wintered fry reported by Dan and Little (2000b) and lower than the 94 –100 % survival rate for over-wintering brood tilapia (Dan and Little, 2000a). The effect of fish size on cold tolerance is either significant or insignificant, according to several authors (Chaani et al., 2000; Frei et al., 2007; Siddick et al., 2014). Fingerlings measuring 5.8 g on average are more susceptible to cold stress than fingerlings weighing 9.69 g on average, according to Hofer and Watts (2002). According to Charo-Karisa et al. (2005), Nile tilapia juveniles should weigh at least 5 g to improve their overwinter survival. Therefore, the disparity in the survival rate (%) of the present investigation could be due to the smaller size of the fry used for stocking in both treatments. Again T1 showed resilience by recording a higher survival rate. Though this difference was not significantly different, it confirms that *O. niloticus* can easily adapt to adverse environmental and cultural conditions compared to *O. andersonii*.

Throughout the trial, key water quality measures were continuously observed. The acceptable standard range for tilapia culture was fulfilled by the mean observed values of the important water parameters (Dissolved Oxygen: 6 mg/l, pH: 7.1, Nitrites: 0.73 mg/l, and Nitrates 16.6 mg/l).

Temperature was the main area of concern because it negatively impacts fish across Zambia during the cold winter season. The average morning temperature was 16.88 ± 2.8 °C (6 hours), the afternoon temperature was 23.63 ± 2.02 °C (12 hours), and the evening temperature was 26.0 ± 2.79 °C (18 hours). As summer drew nearer, the average morning temperatures surprisingly dropped, and the afternoon and evening temperatures marginally rose, as seen in (Figure 6). However, the average daily temperature during this investigation was within a suitable range (20–30 °C) for tilapia culture (Mjoun et al. 2010; Mirea 2013). On the other hand, *O. niloticus* grows best at 26–30 degrees Celsius (Azaza et al. 2008; Chervinski 1982). According to Nehemia et al., (2012), fish growth is about 30 % of its maximum at temperatures between 20 and 22 °C. The activity and feeding of fish declines when the temperature falls below 20°C and ceases entirely at about 16°C. The survival and growth of tilapia raised in aquaculture ponds in Egypt were adversely affected when the winter temperature fell to 16 °C or lower (Siddik et al., 2014; Soltan et al., 2015; Maher et al 2023). Thus, in this study, the growth and survival of the fingerlings may have been adversely impacted by the low morning temperatures of about 16°C, which may have contributed to reduced final weights and weight gains of both species.

CONCLUSION

The findings of this study demonstrated that the fingerlings of *O. niloticus* and *O. andersonii* raised in concrete ponds covered with greenhouse plastic had comparable growth performance averages (weight gain, total weight gain, daily weight gain, survival and specific growth rate). *O. niloticus* had a much lower FCR, indicating its ability to utilize feed efficiently under adverse conditions. However, as noted by WorldFish & AbacusBio. (2021), we cannot be positive that this study included a comparison between *O. andersonii* and a genetically modified strain of *O. niloticus* because other research has shown that *O. niloticus* had clear growth rate advantages (Basiita et al., 2022; Day et al 2016). Both *O. niloticus* and *O. andersonii* species in Zambia have not undergone any genetic characterization and hence the strains obtained from hatcheries are

unknown. The strain of *O. andersonii* employed in this investigation demonstrated the same potential for use in aquaculture as *O. niloticus*, which is mostly used for commercial fish farming in Zambia.

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APPENDICES

Appendix A. Figures

Appendix A.1

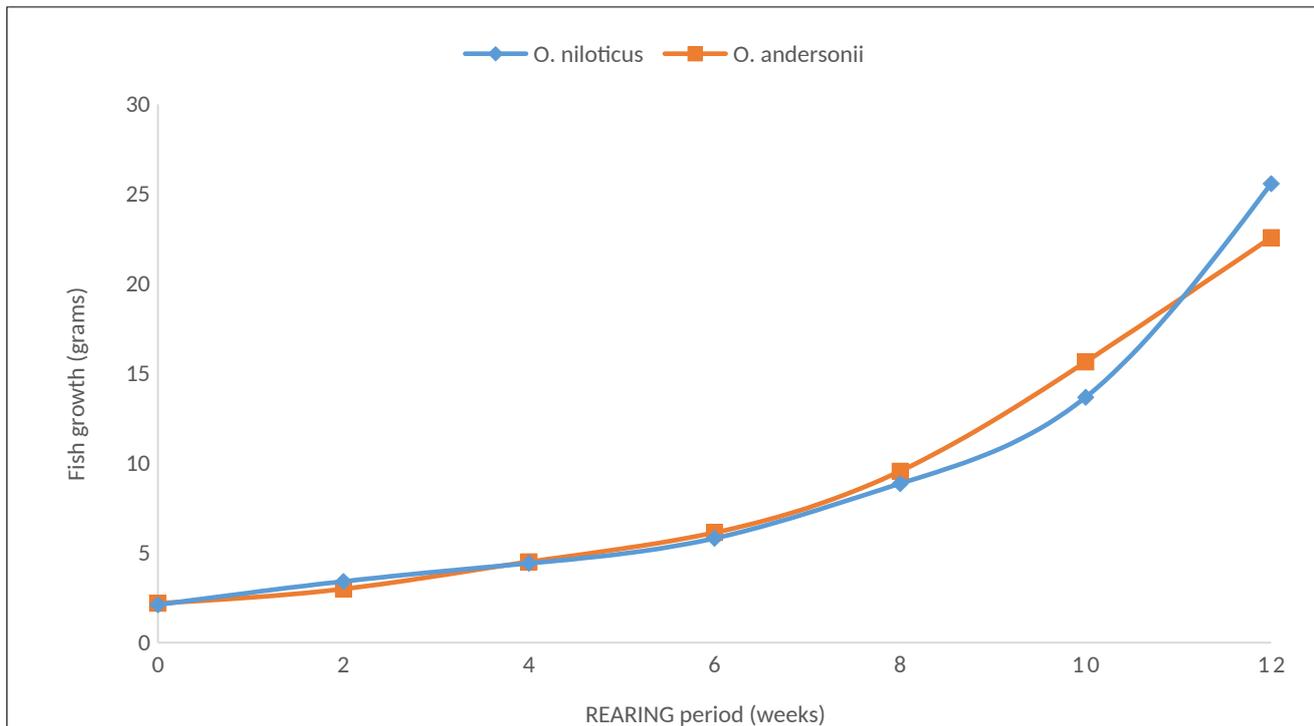


Figure 1: Changes in live body weight (g) of T1 (*O. niloticus*) and T2 (*O. andersonii*) during the experimental period

Appendix A.2

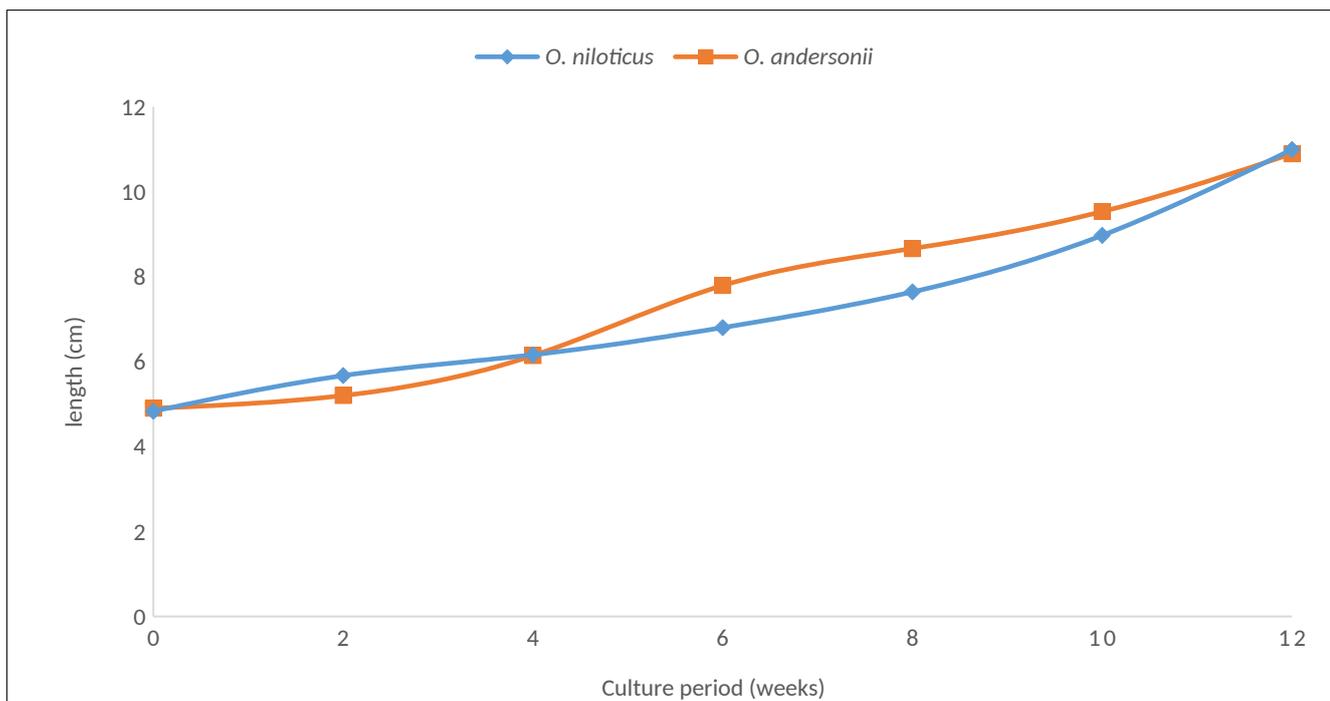


Figure 2: Changes in total length of T1 (*O. niloticus*) and T2 (*O. andersonii*) during the experimental period

Appendix A.3

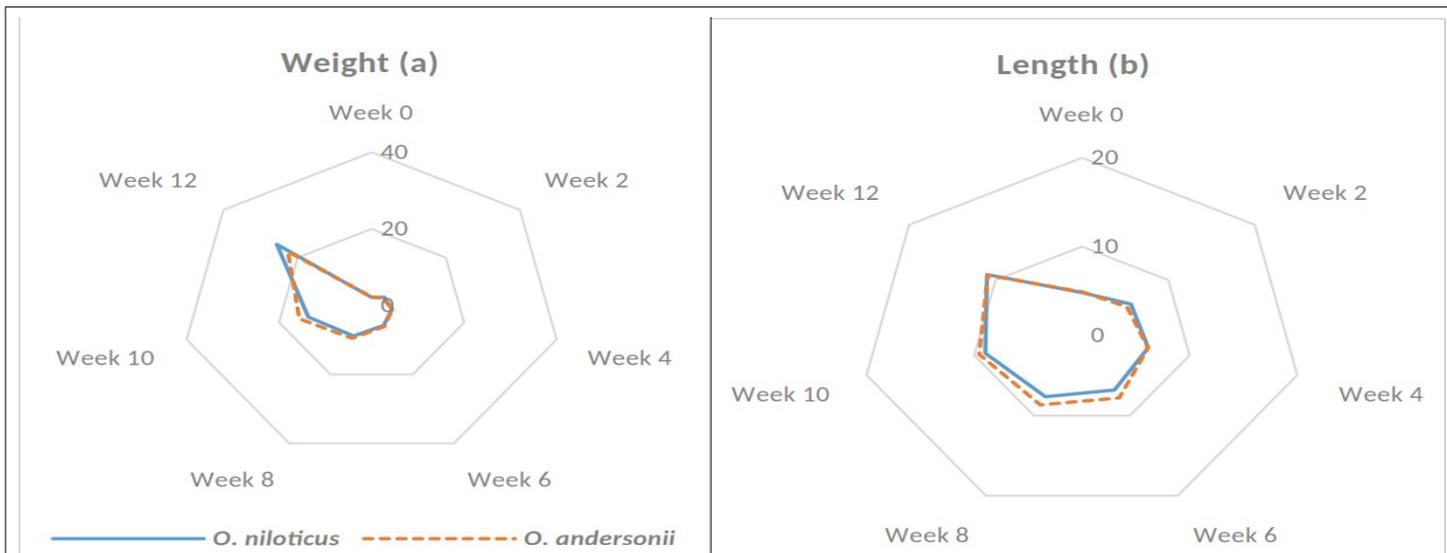


Figure 3: Radar diagrams showing the biweekly distribution weight (grams) (a) and length (cm) (b) for T1 (*O. niloticus*) and T2 (*O. andersonii*) during the experimental period.

Appendix A.4

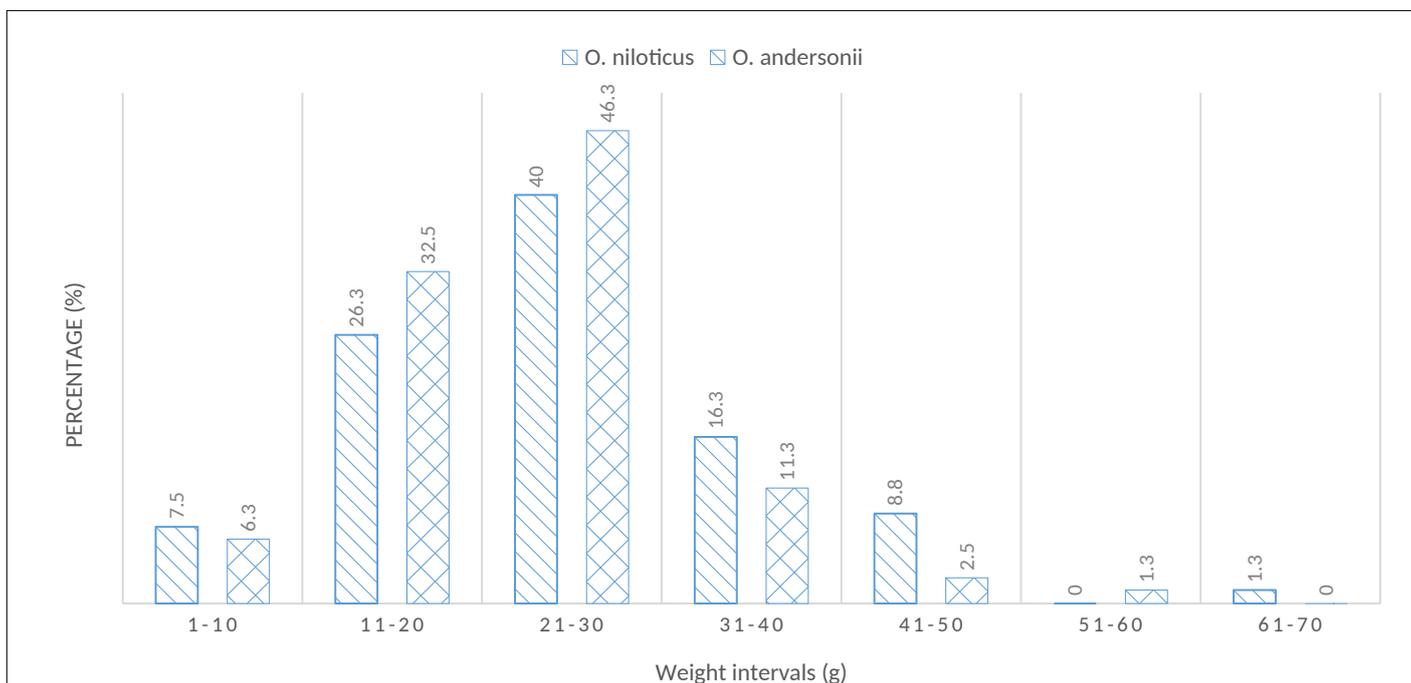


Figure 4: Weight intervals showing percentage distribution of live body weight (g) of T1 (*O. niloticus*) and T2 (*O. andersonii*) at the end of the experimental period

Appendix A.5

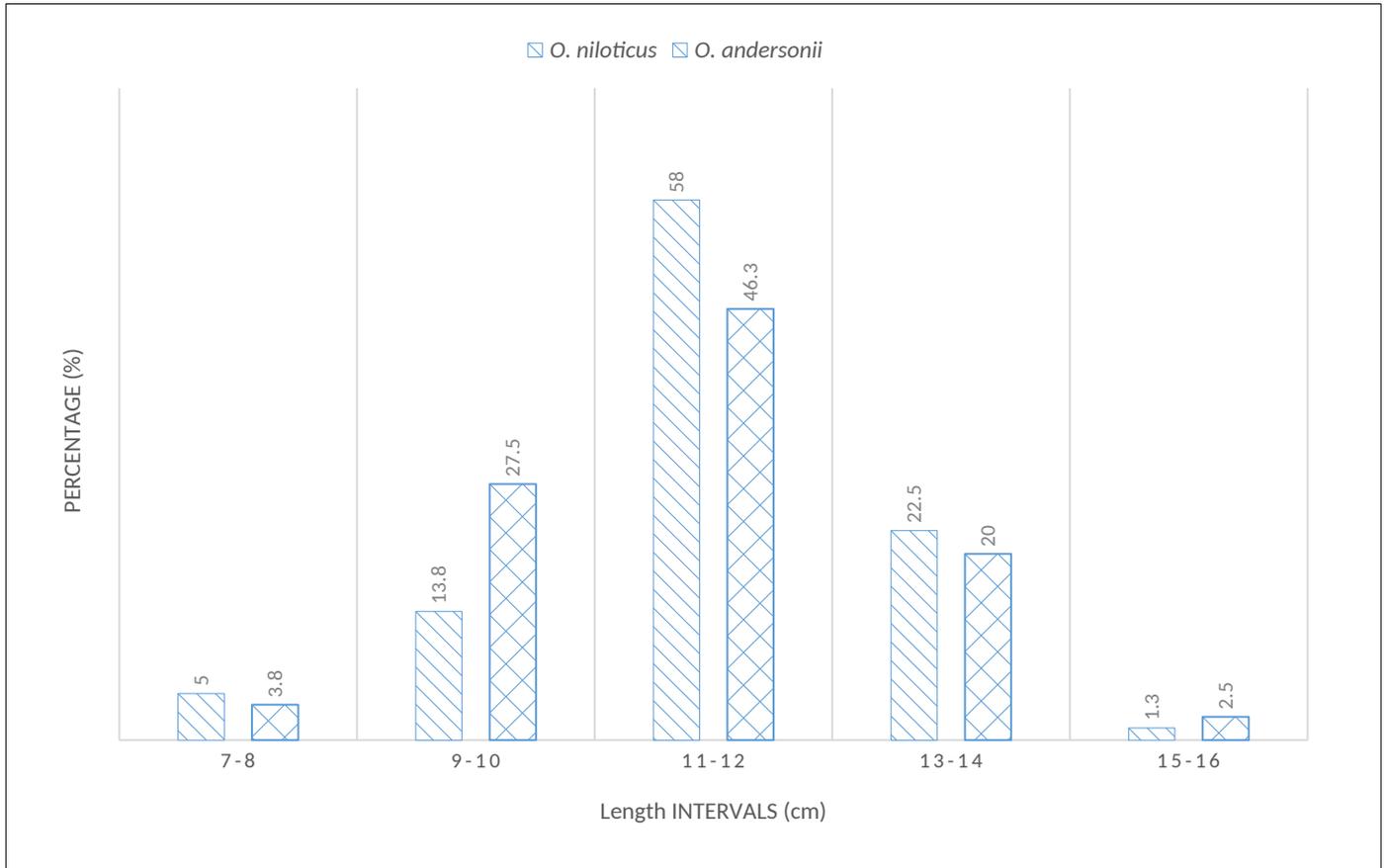


Figure 5: Length intervals showing percentage distribution of total length (cm) of T1 (*O. niloticus*) and T2 (*O. andersonii*) at the end of the experimental period

Appendix A.6

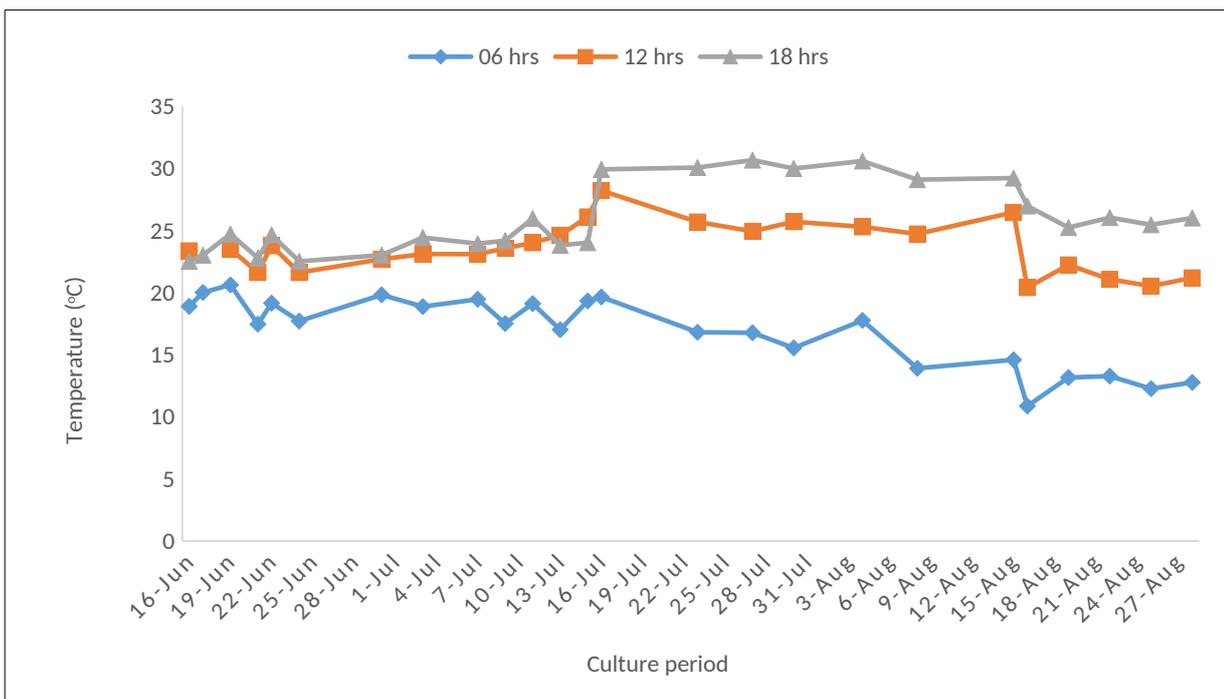


Figure 6: Water temperature at dawn (06hrs), midday (12hrs), and evenings (18 hrs) during the experimental period.

Appendix A.7

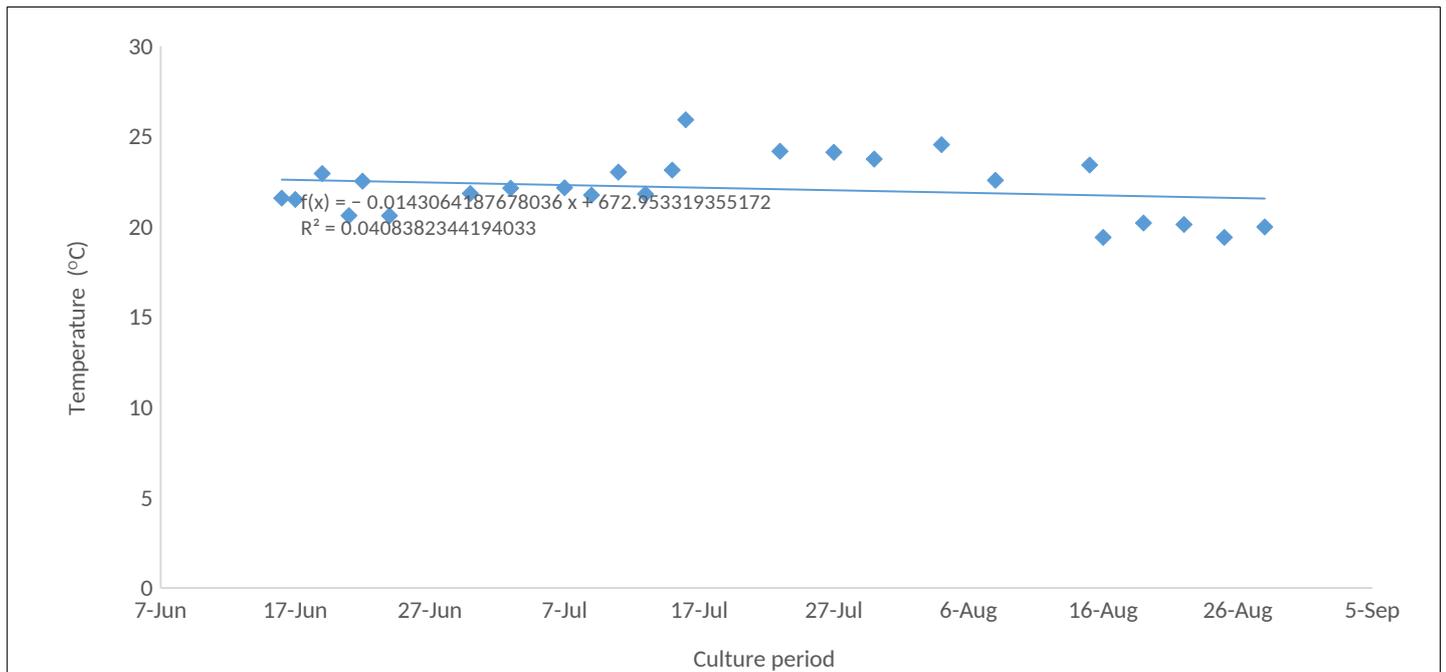


Figure 7: The average daily water temperature was not correlated to culture period (R2 = 0.04), but showed a slightly negative decline towards the end of winter (August).

Appendix B.Tables

Appendix B.1

Table 1: Formulation and proximate composition (on dry matter basis) of the diet fed to the fingerlings during the experimental period.

INGREDIENTS	INCLUSION LEVEL (%)
Fish meal	49
Maize bran	46
Cassava meal	2.5
Palm oil	1.0
Salt	1.0
Vit/Mineral Premix*	1.6
<i>Proximate composition</i>	
Crude Protein	33.01
Crude fat	21.03
Moisture	7.04
Ash	6.62
Phosphorous	1.42
*Broiler Vitamin/Mineral premix content/500g diet: Vitamin A 2,100,000 IU; Vitamin D3 640,000 I; Vitamin E 3,200 IU; Vitamin K 200 mg; Vitamin B1 200 mg; Vitamin B2 1100 mg; Vitamin B6 200 mg; Vitamin B12 2 mg; Biotin 20 mg; Niacin 6600 mg; Pantothenic Acid 3300 mg; Folic Acid 200 mg; Choline Chloride 42,000 mg; Iron (Fe) 3300 mg; Manganese (Mn) 17,000 mg; Copper (Cu) 2000 mg; Zinc (Zn) 15000 mg; Cobalt (Co) 100 mg; Iodine 300 mg; Selenium (Se) 20 mg; Antioxidant.	

Appendix B.2

Table 2: Growth parameters of *O. niloticus* (T1) and *O. andersonii* (T2) after 84 days of culture represented as Mean \pm Standard

Parameter	<i>O. niloticus</i> (mean \pm SD) (g)	<i>O. andersonii</i> (mean \pm SD)	Significance level (p-value)
Initial weight (IW) (g)	2.12 \pm 1.03 ^a	2.20 \pm 1.03 ^a	0.587
Average final weight (g)	25.21 \pm 10.38 ^a	22.57 \pm 8.44 ^a	0.107
Average weight gain (AWG) (g)	24.37 \pm 3.67 ^a	20.49 \pm 1.50 ^a	0.142
Percent Weight gain (PWG) (%)	1155 \pm 159.46 ^a	936.41 \pm 67.50 ^a	0.094
Average Daily Gain (ADG) g	0.292 \pm 0.04 ^a	0.244 \pm 0.02 ^a	0.133
SGR (g/day)	3.12 \pm 0.32 ^a	2.80 \pm 0.07 ^a	0.162
FCR	1.07 \pm 0.11 ^b	1.62 \pm 0.10 ^c	0.035*
Final Biomass (Kg)	10.31 \pm 0.67 ^a	8.24 \pm 0.06 ^a	0.149
Survival (%)	81.33 \pm 14.5 ^a	66.67 \pm 0.58 ^a	0.154

a = no significant difference; b and c = significant difference*, significant at 0.95 confidence limit i.e., P<0.05

Appendix B.3

Table 3: Physio-chemical environmental parameters in the ponds during the

Parameter	Average	*Recommended
Dissolved Oxygen (mg/L)	6	\geq 5
Temperature (°C)	22.1	25°C - 32
pH	7.1	6.5 - 9
Ammonia (mg/L)	0.125	\leq 0.25
Nitrate (mg/L)	16.6	< 100
Nitrite (mg/L)	0.73	0.5 - 1
Conductivity (μ S/cm)	216	< 250
Turbidity (cm)	32.5	20 - 40

experimental period

*Soto-Zarazúa et al., 2010, MAAIF 2020