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Biometric and Survival Responses of Sex-Reversed Nile Tilapia (*Oreochromis niloticus* L.) Fingerlings to Extended Acclimatization Periods

Adrian C. Dela Cruz[1] and Alvin T. Reyes[1, 2]*

[1]College of Fisheries-Central Luzon State University, 3119 Science City of Muñoz, Nueva Ecija, Philippines

[2]Freshwater Aquaculture Center-Central Luzon State University, 3119 Science City of Muñoz, Nueva Ecija, Philippines

[*]**Corresponding Author: alvin.reyes@clsu2.edu.ph**

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ABSTRACT

Evaluating biometric and survival responses is essential in aquaculture for assessing fish health and performance, particularly in species like the sex-reversed Nile tilapia (*Oreochromis niloticus* L.), where understanding the effects of extended acclimatization can improve handling protocols, reduce stress-related losses, and enhance overall rearing efficiency. Therefore, this study was conducted to investigate the effects of extended acclimatization periods on the biometric and survival responses of the sex-reversed Nile tilapia fingerlings. A total of nine experimental units were installed in a single pond, representing three acclimatization durations: 20 minutes (T1), 30 minutes (T2), and 40 minutes (T3), each replicated thrice. Over a 15-day rearing period, biometric parameters such as mean length gain (MLG), mean weight gain (MWG), length-weight relationship (LWR), Fulton's condition factor (K), and relative condition factor (Kn) were evaluated alongside survival rates and water quality. The results revealed no significant differences in MWG, MLG, K, Kn, and survival rates across treatments. All groups exhibited negative allometric growth with moderately strong length-weight correlations (r^2 = 0.75–0.81). Water quality parameters remained within acceptable ranges throughout the study, suggesting that environmental conditions were not limiting factors. Histograms of biometric parameters indicated consistent distributions with minimal outliers. These findings demonstrate that the sex-reversed Nile tilapia fingerlings are resilient to varying acclimatization durations, with no adverse effects on growth or survival, supporting the flexibility of acclimatization protocols in hatchery and grow-out operations. Among the tested durations, a 20-minute acclimatization period remains a practical option to minimize time and labor, and the results may also be applicable to other aquaculture species. Future studies are encouraged to include post-stocking fingerling counts and longer culture periods to better assess the long-term effects of acclimatization strategies.

INTRODUCTION

In aquaculture, the evaluation of biometric and survival responses serves as a cornerstone for assessing the health, performance, and adaptability of cultured species under varying environmental and management conditions **(Akinkuolie *et al*., 2021)**. Biometric parameters, such as length, weight, and condition factors, not only reflect the

individual growth potential of fish but also provide valuable insights into the efficiency of management practices **(Ibrahim *et al.*, 2025)**. When combined with survival rate analysis, these indicators help determine the overall effectiveness of rearing strategies and the resilience of fish stocks to handling and environmental stressors **(Ndiaye *et al.*, 2015)**.

Among commonly cultured freshwater species, the Nile tilapia (*Oreochromis niloticus* L.) remains a dominant choice due to its fast growth, adaptability, and high market demand **(Munguti *et al.*, 2022)**. The use of sex-reversed male fingerlings has become widespread in commercial hatcheries to achieve uniform growth and better production outputs **(Chouhan *et al.*, 2025)**. However, these fingerlings are particularly sensitive during the post-nursery phase, especially when transferred to new environments where acclimatization is required to reduce handling stress and physiological shock **(Schreck & Tort, 2016)**.

Acclimatization, when poorly managed, can result in elevated mortality rates and suboptimal growth. While short acclimatization periods are common in practice, limited research has been conducted on the effects of extended acclimatization on biometric traits and survival. Understanding how prolonged acclimatization influences these responses is essential for improving juvenile handling protocols, minimizing stress-related losses, and ensuring consistent performance during grow-out **(Paixão *et al.*, 2024)**.

This study was conducted to assess the biometric and survival responses of the sex-reversed Nile tilapia fingerlings subjected to varying acclimatization durations. By determining how these responses change with extended acclimatization periods, the research aimed to inform best practices for improving fish welfare and production outcomes in tilapia aquaculture.

MATERIALS AND METHODS

1. Experimental pond and units

The research was carried out at the Freshwater Aquaculture Center (FAC) of Central Luzon State University, located in Science City of Muñoz, Nueva Ecija, Philippines. A single 1,000m² pond served as the study site and was prepared according to the standard procedures of the research center. Nine experimental units (hapa nets) were set up within the pond, representing three different acclimatization durations (Treatment 1: 20 minutes, Treatment 2: 30 minutes, and Treatment 3: 40 minutes), each with three replicates. Each hapa net measured 2.5 x 2 x 1 m. To protect the fish from bird predation, B-netting was installed over every unit. The study design followed a randomized complete block design (RCBD).

Water samples were collected from five different areas of the experimental pond, both before and after the rearing period, to assess its suitability for culture. Water quality analyses included measurements of alkalinity, hardness, nitrite, total ammonia-nitrogen (TAN), and phosphorus. Alkalinity and hardness were measured using laboratory titration, while nitrite, TAN, and phosphorus were measured using spectrophotometry.

2. Experimental animal

The sex-reversed Nile tilapia fingerlings (size #22) used in the experiment were sourced from the pond production system of FAC-CLSU. These fingerlings underwent sex-reversal at the research center before the experiment began. They were initially collected from nursery hapas and held in a conditioning tank for 24 hours before being transported. Nine plastic bags were used during transport, each containing approximately 1,100 fingerlings. The quantity of fish per bag was estimated based on weight, with 130g equivalent to roughly 1,100 fingerlings. Packing was done in the morning, and the transport duration lasted four hours to replicate typical delivery conditions experienced by the FAC's clientele.

3. Stocking and acclimatization procedure

Stocking was carried out between 3:00 pm and 4:00 pm. Prior to introducing the fingerlings into the hapas, temperature, dissolved oxygen, and pH were measured using a multiparameter device (for temperature and DO) and a pH meter. One sealed plastic bag of fingerlings was assigned to each of the nine experimental units. These bags were floated on the water surface for acclimatization: 15 minutes for Treatment 1, 25 minutes for Treatment 2, and 35 minutes for Treatment 3. After the floating period, the bags were gradually opened and acclimated by adding one cup of pond water every minute for the final five minutes.

4. Monitoring and feeding

The fingerlings were monitored and fed over a 15-day period. Water quality parameters (temperature, DO, and pH) were measured twice daily, once in the morning and once in the afternoon. The daily feeding rate was maintained at 20% of the average body weight (ABW), with feed given four times a day at 8:00 a.m., 11:00 a.m., 1:00 p.m., and 4:00 p.m., at approximately three-hour intervals.

5. Fish sampling

At the end of the 15-day period, fingerlings were harvested from each unit using a bamboo pole to gather them at one end of the hapa net. A strainer was then used to collect the fish, which were transferred into small basins for weighing. The total biomass and number of fingerlings per unit were recorded to determine the survival rate. Additionally,

50 fish were randomly selected from each unit, sedated using Tricaine methanesulfonate (MS-222), and individually weighed and measured for length-weight relationship (LWR) analysis. A digital weighing scale was used for weight, while total length was measured using a caliper. The following parameters, as used by other researchers, were considered to evaluate the biometric and survival responses of the fingerlings:

Mean weight gain (**Panase & Mengumphan, 2015**)

MWG (g) = Final weight (g) – Initial weight (g)

Mean length gain (**Panase & Mengumphan, 2015**)

MLG (cm) = Final length (cm) – Initial length (cm)

Length-weight relationship (Panase & Mengumphan, 2015)

$W = aL^b$

where W is weight, L is total length, *a* is the intercept, and *b* is the growth coefficient.

Fulton's condition factor (*for isometric growth assumption*) **(Ragheb, 2023)**

Condition factor (K) = (100*W) / L^3

where, W is the weight in g, and L is the total length in cm

Relative condition factor *(for actual growth pattern*) **(Le Cren, 1951)**

Relative condition factor (Kn) = Wo / Wc

where Wo is the observed weight, and Wc is the calculated weight

Survival rate (Venkatachalam *et al*., 2017)

SR (%) = (No. of stocks harvested/Initial no. of stocks) x 100

6. Statistical analysis

The data on the different biometric parameters and survival rates were analyzed using the analysis of variance (ANOVA) single factor. Microsoft Excel's analysis toolpak was used for all the analyses at a 5% level of significance. Furthermore, this Microsoft Excel add-in was used for the determination of *b* values for the length-weight relationship analysis.

RESULTS

1. Biometric performance, length-weight relationship analysis, and condition factors

The biometric analysis (Table 1) showed consistent growth patterns among all treatment groups regardless of the acclimatization duration. There were no statistically significant differences in weight gain and length gain for the sex-reversed fingerlings.

The analysis of the length-weight relationship (Fig. 1) further supported this, as all groups exhibited negative allometric growth ($b < 3$). The correlation between length and weight remained moderately strong and uniform across treatments. Condition factor

results were also uniform across all treatments. Both the Fulton's and relative condition factors remained close to 1.00.

Table 1. Biometric parameters of the sex-reversed Nile tilapia fingerlings subjected to different acclimatization periods

Parameter	Acclimatization Periods		
	T1	T2	T3
	20 minutes	30 minutes	40 minutes
MWG (g)	0.46±0.01[a]	0.48±0.07[a]	0.43±0.02[a]
MLG (cm)	0.72±0.05[a]	0.74±0.13[a]	0.66±0.01[a]
b value	2.71	2.43	2.66
Growth behavior	Negative allometry	Negative allometry	Negative allometry
r^2 value	0.78	0.75	0.81
K value	1.66±0.04[a]	1.72±0.15[a]	1.69±0.04[a]
Kn value	1.02±0.02[a]	1.02±0.08[a]	1.02±0.02[a]

*Means with the same superscript letter are not significantly different at the 0.05 level. Values are presented as mean ± standard deviation.

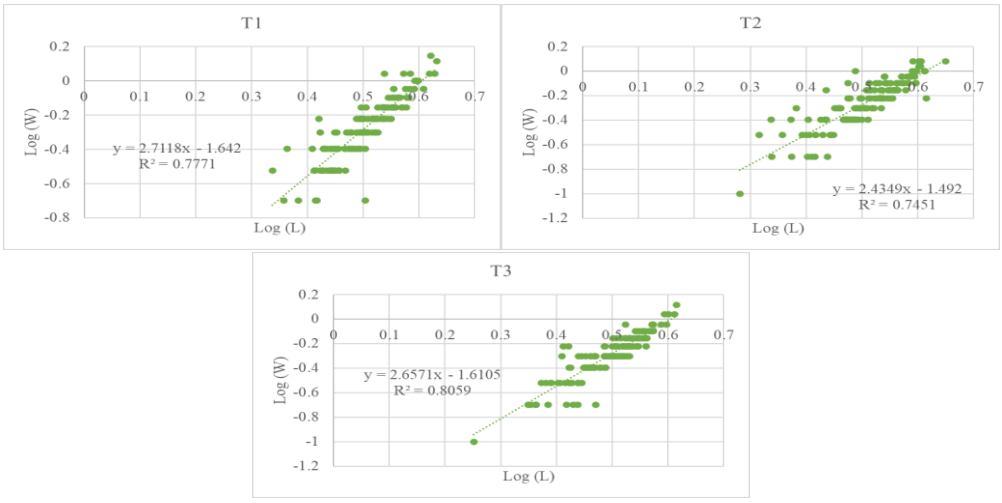

Fig. 1. Linear regression of the length-weight relationship of the acclimatized sex-reversed Nile tilapia fingerlings after the 15-day culture period

2. Histogram distributions

Histograms of the biometric distributions revealed that fish lengths followed a near-normal distribution (Fig. 2) across all treatments, while weight distributions displayed a slight rightward skew (Fig. 3). The condition factor (K) and relative condition factor (Kn) values were closely clustered across treatments. Only a few outliers were observed in the distributions, but these did not affect the overall data pattern (Figs. 4, 5).

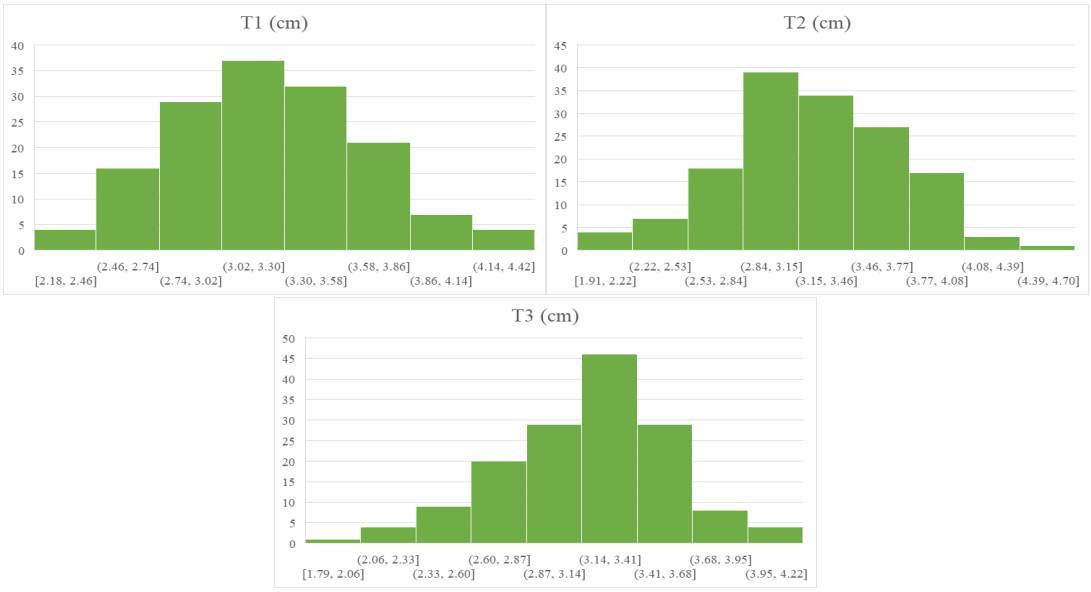

Fig. 2. Length distribution histogram of the sampled Nile tilapia fingerlings after the 15-day culture period.

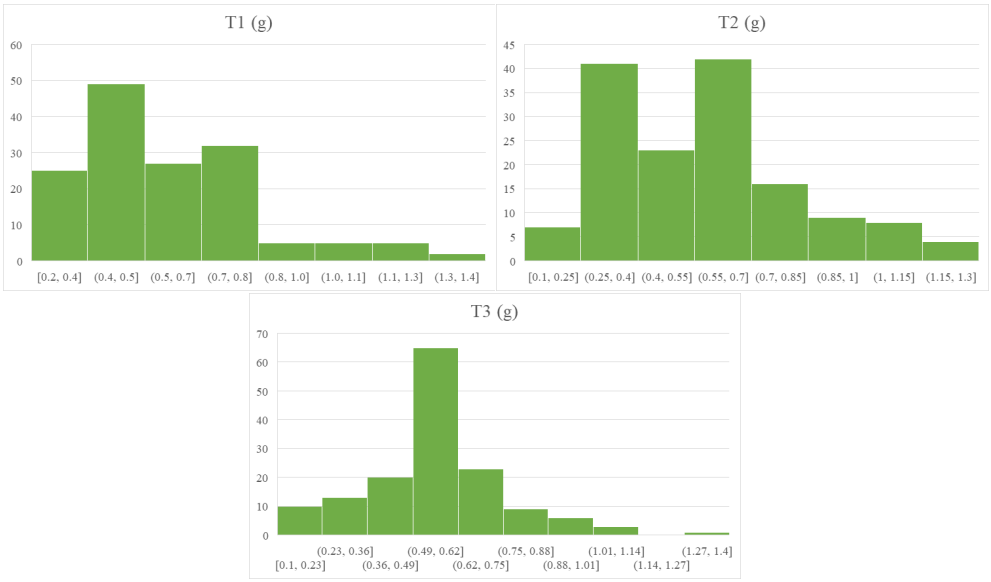

Fig. 3. Weight distribution histogram of the sampled Nile tilapia fingerlings after the 15-day culture period.

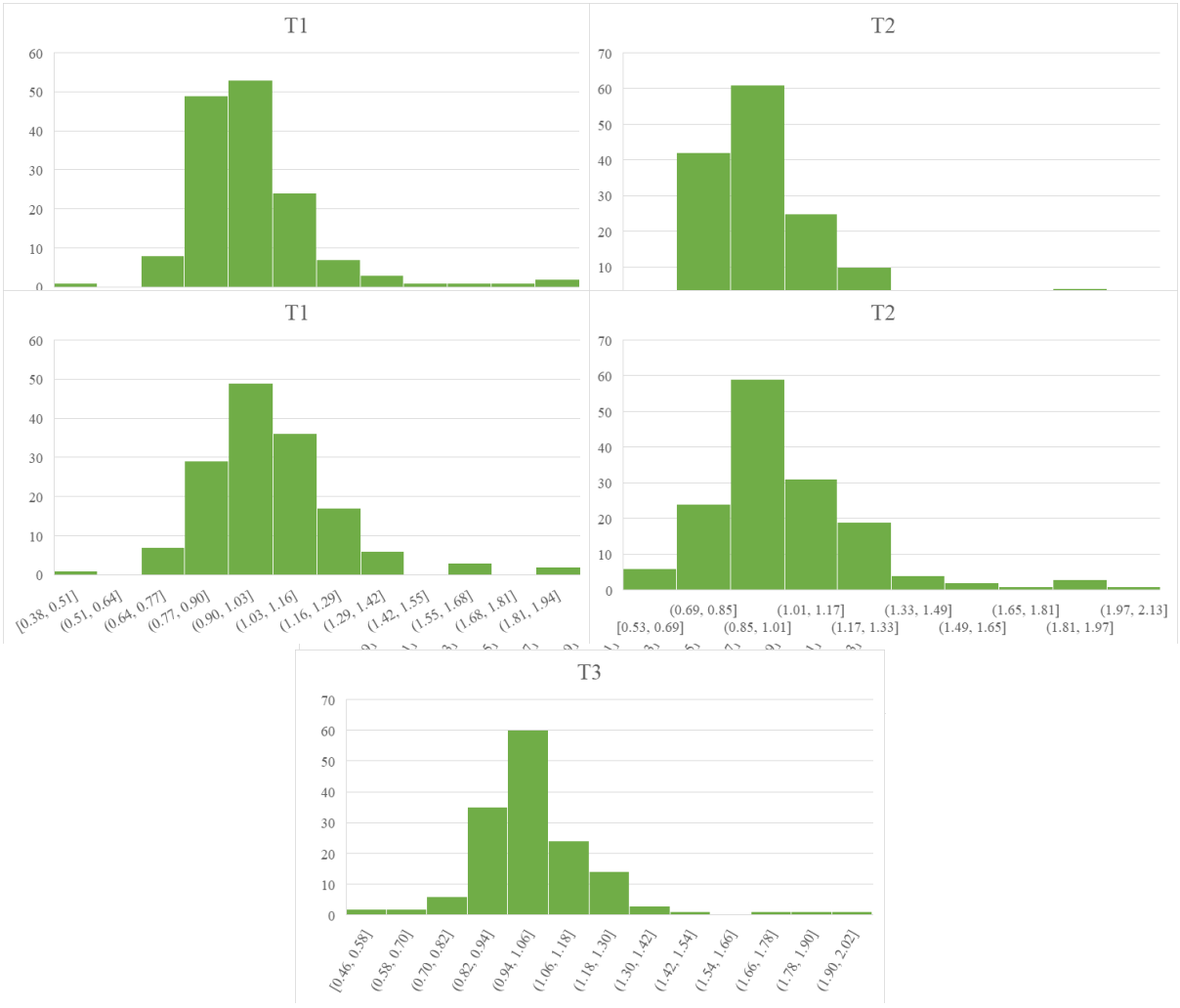

Fig. 5. Kn distribution histogram of the sampled Nile tilapia fingerlings after the 15-day culture period.

3. Water quality in the experimental pond

Prior to stocking, all treatments recorded DO levels well above the recommended minimum of 5 mg/L **(Riche & Garling, 2003)**. Temperature readings were slightly above the recommended range of 27 to 32°C **(Pandit & Nakamura, 2010)**. While pH levels were near the upper limit, they were still within the optimal range of 6.7 to 9.5 **(Santhosh & Singh, 2007)**. These values are summarized in Table (2).

During the 15-day culture period, water quality remained within acceptable ranges (Table 3). Morning (am) readings showed slightly lower DO and temperature values. In contrast, afternoon (pm) measurements reflected increased values, especially for DO

(Table 3). Despite these daily fluctuations, the overall values remained consistent across treatments and stayed within or close to the recommended thresholds.

Table 2. Average (±SD) values of dissolved oxygen, temperature, and pH in the pond water prior to stocking the Nile tilapia fingerlings

Treatment	Dissolved Oxygen (mg/L)	Temperature (°C)	pH
1	11.53±0.015	34.5±0.012	9.21±0.015
2	11.49±0.025	34.3±0.064	9.32±0.015
3	11.45±0.040	34.8±0.115	9.40±0.017

Table 3. Average (±SD) daily readings of water quality parameters recorded over the 15-day culture period.

Treatment	Dissolved Oxygen (mg/L)	Temperature (°C)	pH
	Am		
1	7.52±1.16	31.04±0.99	9.47±0.23
2	7.40±1.15	31.10±0.99	9.40±0.32
3	7.43±1.14	31.15±1.00	9.43±0.31
	Pm		
1	9.38±0.49	32.40±1.32	9.31±0.44
2	9.36±0.49	32.39±1.29	9.32±0.43
3	9.35±0.49	32.35±1.26	9.30±0.47

Furthermore, the results of the laboratory water quality analysis revealed some changes in key parameters before and after the study (Table 4). Total alkalinity slightly decreased over the course of the experiment but remained within the recommended range of 75 to 200 mg/L, as outlined by **Bhatnagar *et al.* (2013)**. Similarly, total hardness also declined, though it still fell within the acceptable range of 50 to 150mg/ L according to **Swain *et al.* (2020)**.

Nitrite levels showed a slight increase but stayed well below the recommended upper limit of 0.3mg/ L **(Rebouças *et al*., 2016)**. On the other hand, TAN levels increased slightly, though they remained within the safe limit of less than 1mg/ L, as recommended by **Emerenciano *et al.* (2017)**. Phosphorus levels experienced a minor increase by the end of the study period, yet they remained well below the threshold of 0.5mg/ L, as cited by **Boyd (2001)**.

Table 4. Mean of water analyses (±SD) on alkalinity, hardness, nitrite, toal ammonia-nitrogen, and phosphorus before and after the conduct of the study

Period	Total Alkalinity (mg/L)	Total Hardness (mg/L)	Nitrite (mg/L)	TAN (mg/L)	Phosphorus (mg/L)
Before	139.60±3.05	90.89±6.73	0.02±0.001	0.16±0.04	0.017±0.003
After	126.20±2.59	83.28±2.69	0.05±0.022	0.32±0.03	0.021±0.001

4. Survival rates

Survival rates remained consistently high across all treatments, with no significant differences observed (Table 5). No sharp decline in survival was noted, suggesting that the varying acclimatization periods did not impose excessive stress on the fingerlings. This stability indicates that the fish adjusted well after transport and tolerated the acclimatization durations effectively.

Table 5. Survival rates of the sex-reversed Nile tilapia fingerlings subjected to different acclimatization periods

Parameter	Acclimatization Periods		
	T1 20 minutes	T2 30 minutes	T3 40 minutes
Survival rate (%)	86±2.75 [a]	83±4.86 [a]	84±3.23 [a]

*Means with the same superscript letter are not significantly different at the 0.05 level. Values are presented as mean ± standard deviation.

DISCUSSION

1. Biometric responses

No significant difference was observed in the mean length gain (MLG) or mean weight gain (MWG) values among the three treatments. This implies that the fish recovered from handling and transportation stress after all acclimatization periods, enabling normal growth to continue. Their established reputation as a robust aquaculture species is supported by the fact that the Nile tilapia fingerlings can grow similarly under different acclimatization durations, demonstrating their adaptability and hardiness **(Prabu *et al.*, 2019; El-Sayed & Fitzsimmons, 2023).**

2. Length-weight relationship

With *b* values less than 3, the length-weight relationship (LWR) analysis showed a consistent negative allometric growth pattern across all acclimatization treatments. This suggests that over the 15-day culture period, the fish length developed more quickly than its weight, as **De Castro Silva *et al.* (2015)** remark. Studies by **Mitra *et al.* (2014)** and

Abdulkarim *et al.* (2019) have also demonstrated that this trend is common in juvenile fish, where elongation frequently occurs prior to notable weight growth.

This tendency was evidently reflected in the histogram distributions. The length histograms for all treatments displayed a relatively bell-shaped distribution, indicating that the population's sizes and lengths were uniformly distributed, as **Ravi and Yahaya (2015)** discussed. Nonetheless, the weight histograms showed a rightward tilt, suggesting that more fingerlings were grouped in the lower weight range and fewer were reaching heavier weights, a finding also shown in the study of **Knouft (2004)**.

Furthermore, the negative allometric growth pattern observed in the LWR, where length increases faster than weight, aligns with the length and weight histograms. The bell-shaped length distribution indicates uniform size, while the right-skewed weight distribution reflects slower weight gain relative to length.

The alignment between the LWR results and the histogram patterns strengthens the reliability of the biometric data. It confirms that the observed growth trend was not random or treatment-induced but rather a characteristic developmental stage of the fingerlings during the short rearing period. Furthermore, according to **Saura *et al.* (2018)**, the comparatively high r^2 values across treatments show a steady and predictable LWR, confirming that the fish were growing consistently independent of the period of acclimation.

3. Condition factor analysis

According to **Jisr *et al.* (2018)**, a fish is in good growth condition when its K value is equal to or more than 1, and it is in bad growth condition when its K value is less than 1. For the Kn, **Le Cren (1951)** clarified that a value more than 1 indicates that the fish is healthy, but a value less than 1 indicates that it might be stressed or malnourished.

Both Fulton’s condition factor (K) and the relative condition factor (Kn) were statistically similar across treatments. K values indicated generally good body condition, while Kn values showed that the actual weights closely matched expected values based on fish length. These parameters confirm that fish health and nutritional status were well-maintained throughout the study.

The histograms of K and Kn provided visual confirmation of this uniformity. With only a few small outliers, most fish fell within a small range of values, indicating that each fingerling's growth and condition were consistent. As also noted in the study by **Azaza *et al.* (2013)**, the few extreme values were probably caused by individual variation rather than the effects of acclimatization.

4. Water quality

Water quality throughout the study remained within acceptable ranges and did not vary significantly across treatments. DO, pH, temperature, and other parameters such as TAN, nitrite, and phosphorus remained within or near recommended levels, confirming that environmental conditions did not serve as confounding factors during the experiment. These stable conditions allowed for a clearer evaluation of acclimatization duration effects on biometric and survival responses.

The stable water quality parameters were likely a major reason for the similarities observed in the measured growth parameters. It has been demonstrated in numerous studies that tilapia cultured in various environments with diverse water quality parameters may exhibit variations in growth performance. In the study conducted by **Moses *et al.* (2021)**, for instance, notable differences between tilapia raised in fresh and brackish waters were noted.

5. Survival rates

Survival rates were high and statistically similar among all treatments, ranging from 83% to 86%. The absence of significant differences implies that all acclimatization durations were effective in minimizing stress and supporting successful adjustment to the rearing environment. These findings support existing literature stating that gradual acclimatization through floating and water mixing can significantly reduce post-transport mortality **(Paixão *et al*., 2024)**.

CONCLUSION

The findings of this study indicate that extended acclimatization durations ranging from 20 to 40 minutes did not significantly affect the biometric parameters or survival rates of the sex-reversed Nile tilapia fingerlings. This suggests that flexible acclimatization protocols can be employed in aquaculture operations without compromising fish performance, thereby supporting more efficient and adaptive management strategies. While all tested durations yielded similar results, a 20-minute acclimatization remains a practical choice to minimize time and labor. These findings may also be applicable to other aquaculture species. For a more thorough evaluation of acclimatization effects, future studies are recommended to include post-stocking fingerling counts and to extend the culture period beyond 15 days to assess long-term growth trends.

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