

EVALUATING GROWTH AND NUTRIENT COMPOSITION OF AFRICAN CATFISH UNDER DIFFERENT SALINITIES

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ABSTRACT:

An investigation was carried out to explore the effect of salinity on the growth and flesh quality of African catfish (*Clarias gariepinus*), a significant species in global aquaculture. Fifty juvenile catfish of weight $24.25 \pm 0.4g$ were collected from the Chenab River in Pakistan and acclimatized in controlled aquaculture conditions at different salinity levels NaCl (T1; 0, T2; 2, T3; 4; T4; 6, and T5; 8 ppt) over 60 days. Growth performance indicators, including final weights, weight gain, growth rate, and feed conversion ratio (FCR), were measured. The statistical result indicated that the fish in lower salinity environments (0-2 ppt) exhibited significant ($p < 0.05$) final weights, weight gain, and growth rates, with performance declining significantly as salinity increased. The best FCR 1.2 ± 0.01 was indicated at 0 ppt, while the highest salinity (8 ppt) was revealed in the FCR 1.7 ± 0.04 . Proximate composition analysis indicated that crude protein and moisture content elevated with salinity while crude fat and ash content decreased. These results suggest that *C. gariepinus* thrives in slightly brackish conditions, with optimal growth at low salinity levels. However, higher salinities adversely impact physiological health and growth performance.

KEYWORDS: *Clarias gariepinus*, Growth performance, Proximate composition, Salinity levels.

1. INTRODUCTION

African catfish (*Clarias gariepinus*) is a vital freshwater fish famous around the globe due to their delicious taste, rapid growth, and large size with few spines in their flesh, withstand a wide range of environmental conditions and disease resistance; withstand stress which makes them most predominant for food and commercial purposes in aquaculture (Hewitt et al., 2009; Adewolu et al., 2009; Furness, 2016; Seale et al., 2024). In the aquaculture industry, after carp and tilapia, catfish recently attracted the attention of fish culturists towards prolific production (Phan et al., 2009; Adeleke et al., 2020). *C. gariepinus*, belonging to Family Clariidae is widely distributed in Southeast Asia and Africa and has the highest generic diversity used as locally cultured fishes (Cavaco et al., 2001; Chibwana et al., 2013). The *C. gariepinus* survives at various salinity levels, with affected growth, and possesses a limited capacity to survive an increase in salinity (Borode et al., 2002). *C. gariepinus* is extensively found in swamps, lakes, and rivers (Tembo et al., 2023). It has a large body and the capacity to live in stagnant environments due to the occurrence of mucous and pseudo-lungs (Asriqah et al., 2018).

Salinity is a primary factor affecting the growth, survival, health, and distribution of aquatic organisms due to the restriction of freshwater resources, underground brackish water has been an alternative way for extensive fish production in aquaculture; it is necessary to understand the optimum salinity of specific species that will contribute in expansion of aquaculture by utilizing of brackish waters (Semra, 2013; Imsland et al., 2001). The growth performance of *C. gariepinus* shows higher growth in brackish media consumed more food than the freshwater fishes, which largely depends on feed consumption and its assimilation and conversion into body tissues (Nyadjeu et al., 2021). However, salinity directly affects endocrine factors by feeding, growth, and altering the ionic and osmotic regulation. Therefore, it can be an

environmental stressor (Boeuf & Payan, 2001; Varsamos et al., 2004). Limited research has been conducted to evaluate the effects of salinity on growth performance and nutritional qualities of *C. gariepinus*. However, this research aims to assess the different concentrations of salinities on this species' growth and proximate composition. The levels of salinities selected in this study were based on the presence of substantial brackish and saline water resources.

2. MATERIALS AND METHODS

Study area

The Chenab River, an essential watercourse in the Punjab province of Pakistan, has its source in the Indian Himalayas and traverses the Jammu region before entering Pakistan (Figure 1). This river plays a crucial role in the irrigation system, providing essential agricultural nourishment and supporting many aquatic organisms. Water resources play a vital role in agriculture and preserving environmental equilibrium, rendering them an invaluable asset for humans and ecological stability within the vicinity.



Figure 1: Sampling location of fish collection along the River Chenab, Pakistan

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Ethical approval statement

The study protocols encompassed the ethics of laboratory animals and strictly adhered to the regulations established by the Department of Fishers, Saline Water Aquaculture Research Center (SWARC), Pakistan.

Collection of Fish and Experimental Studies

Fifty catfishes, with an initial weight of 24.25 ± 0.4 g, were collected from the Chenab River. These fish were transported to the SWARC (coordinates: N $32^{\circ}41'92''$; E $71^{\circ}43'96''$) in Muzaffargarh, South Punjab, Pakistan. The main objective of this research center is to investigate and advance the progress of aquaculture (Owais et al., 2023). The fish were transferred to a glass aquarium ($140 \times 35 \times 52$ cm³). The data measured included salinity (0- parts per thousand (ppt)), temperature (26.2 °C), pH (8.13), and dissolved oxygen (7.32 mg/L); the fish were given seven days to acclimatize to the new conditions throughout this time, the pond was continuously supplied with aeration. After the acclimation, the aquaria were assigned to five treatment groups: T0 (control), T1, T2, T3, and T4, representing salinity levels of 0, 2, 4, 6, and 8 ppt, respectively. Each treatment had two replicates, with fish stocked at a density of 5 fish per aquarium. The fish were fed a commercial diet containing 30% crude protein, comprising 13.8% fish meal, 6.75% rice protein, 6% mustard oil cake, 2.4% rice bran, and 2% wheat bran, plus vitamin premix, twice daily at 5% of their body weight (Mandal et al., 2020). The feed quantity was adjusted according to the average body weight of the fish, aligning with the new biomass. The water was exchanged every other day, with daily removal of detritus and uneaten feed by siphoning, as described by Xia et al. (2013). The fish were exposed to salinity for 60 days.

Physicochemical Parameters

The physicochemical parameters were meticulously monitored using an Apera 8500 EC meter, an Apera 8500 pH meter, and a P-512 dissolved oxygen meter daily. Aeration was provided to maintain dissolved oxygen levels, and salinity was regularly tested and adjusted as needed over the 60-day experimental period.

Measurement of growth performances

After sixteen days of exposing the fish to different levels of salinities, fish were deprived of food for one night and got free access to water. On the following day, the fish were measured by using Digital Balance Adventure (Model; A&D HT-120), and the growth performance parameters were measured, including feed intake (the amount of food consumed by fish over a specific period.), weight gain (the increase in body weight over time), growth rate percentage (evaluate the increase in their weight over a period), and feed conversion ratio (FCR) (the efficiency of converting feed into body mass), were measured as per the methodologies described by Jewel et al. (2024).

The growth parameters were calculated using the formulas provided below, based on the method outlined by Al Sulivany (2024).

$$WG = FW - IW$$

$$FCR = FG / WG$$

$$GR = (WG / IW) \times 100$$

$$FI = FCR \times WG$$

The abbreviation WG is Weight gain (g/day). FW; Final weight (g), IW; Initial weight (g). FCR; Feed Conversion Ratio (%). GR; Growth Rate (%) FI; Feed Intake (FI; g/day)

Proximate Composition Analysis

After measuring the growth performance parameters, the fish were caught from each aquarium and euthanized by a sharp

blow on the head; then, the meat (muscle) was cut and washed with distilled water (Zhao et al., 2023). The proximate composition of meat was analyzed following the AOAC (1990) guidelines. Five grams of fish meat were dried in an oven at 105 °C to determine the moisture content (Lee et al., 2023). The dried samples were finely crushed using a mortar and pestle, and lipid content was determined using chloroform-methanol (Olayemi et al., 2022). Crude protein content was assessed using the standard micro-Kjeldahl method (Abd El-Karim et al., 2021). For ash content determination, a 2 g dried sample was combusted in a muffle furnace at 550 °C until complete combustion, and the remaining white residue was evaluated as ash content (Zhang et al., 2021).

The proximate composition analysis was calculated using the following formulas (Zhang et al., 2021; Abd El-Karim et al., 2021; Olayemi et al., 2022; Lee et al., 2023).

$$MC = (IW - DW / IW) \times 100$$

$$LC = (W. \text{ of extracted lipid} / SW) \times 100$$

$$CP = NC \times 6.25$$

$$AC = WA / SW \times 100$$

The abbreviation MC is Moisture content (%). DW; Dry weight. LC; Lipid content (%). SW; Sample weight. CP; Crude protein (%). NC; Nitrogen content. AC; Ash Content (%). SW; sample weight

Statistical Analysis

The data obtained from the experiment was utilized for statistical analysis by using GraphPad Prism 9, Analysis of Variance (ANOVA). Duncan's Multiple Range Test mean comparisons were used to find significant differences between groups. A ($P < 0.05$) was used as the indicator for statistical significance. The means and standard errors are presented for each result.

3. RESULTS

The effect of varying salinity levels on the growth performance of African catfish (*C. gariepinus*) is shown in (Table 1 and Figure 1; A, B, C, D, E, and F). The data observed significant variation across treatments during 60 days. Statistical analysis demonstrated that salinity had a profound effect on the parameters of the growth, such as final weight (FW), weight gain (WG), growth rate (GR), and feed conversion ratio (FCR), with a clear trend of decreasing in the growth as salinity levels elevated.

Catfish in the T2 group, exposed to 2 ppt salinity, obtained the highest FW, with mean and SEM ranging between (43.10 ± 0.26 g), closely followed by the T1 group (41.86 ± 0.78 g). Conversely, the fish in the T5 group (with a salinity of 8 ppt) exhibited the highest salinity level. They attained the lowest final weight of 37.80 ± 0.31 g, deemed statistically significant ($p < 0.05$).

T1 and T2 revealed the highest WG (18.62 ± 0.44 g and 18.42 ± 0.16 g, respectively), which was consistent with this trend., while T5 had the lowest WG at 13.81 ± 0.38 g ($p < 0.05$). The GR also demonstrated a similar trend, with T1 achieving a GR of $80.12 \pm 1.33\%$ compared to the significantly lower rate of $57.59 \pm 2.17\%$ in T5 ($p < 0.05$).

Feed intake (FI) remained comparatively constant across all salinity exposures, indicating that the reduced growth performance in higher salinity treatments was not due to decreased feeding but likely due to physiological stress. The FCR was optimal in T1 (1.2 ± 0.01) and experienced a gradual decline progressively with elevated salinity, peaking at (1.7 ± 0.04) in the T5 group ($p < 0.05$).

Table 1: Growth performance (Mean ± SEM) of *Clarias gariepinus* at different salinity levels during the 60-day experiment.

Growth parameters	Treatment				
	T1(0-ppt)	T2 (2-ppt)	T3 (4-ppt)	T4 (6-ppt)	T5 (8-ppt)
Initial weight (g)	23.24±0.40 ^a	24.68±0.24 ^b	24.60±0.40 ^b	24.44±0.24 ^b	24.32±0.32 ^{ab}
Final weight(g)	41.86±0.78 ^{cd}	43.10±0.26 ^d	41.40±0.24 ^c	39.46±0.5 ^b	37.8±0.31 ^a
Weight gain(g)	18.62±0.44 ^d	18.42±0.16 ^d	16.86±0.22 ^c	15.06±0.15 ^b	13.81±0.38 ^a
Feed intake(g)	23.54±0.38 ^a	24.21±0.22 ^a	24.31±0.31 ^a	23.44±0.23 ^a	23.56±0.23 ^a
Growth Rate(g)	80.12±1.33 ^d	74.83±1.40 ^c	68.40±1.80 ^b	61.76±1.19 ^a	57.59±2.17 ^a
FCR	1.2±0.01 ^a	1.3±0.06 ^a	1.4±0.01 ^b	1.5±0.01 ^c	1.7±0.04 ^d

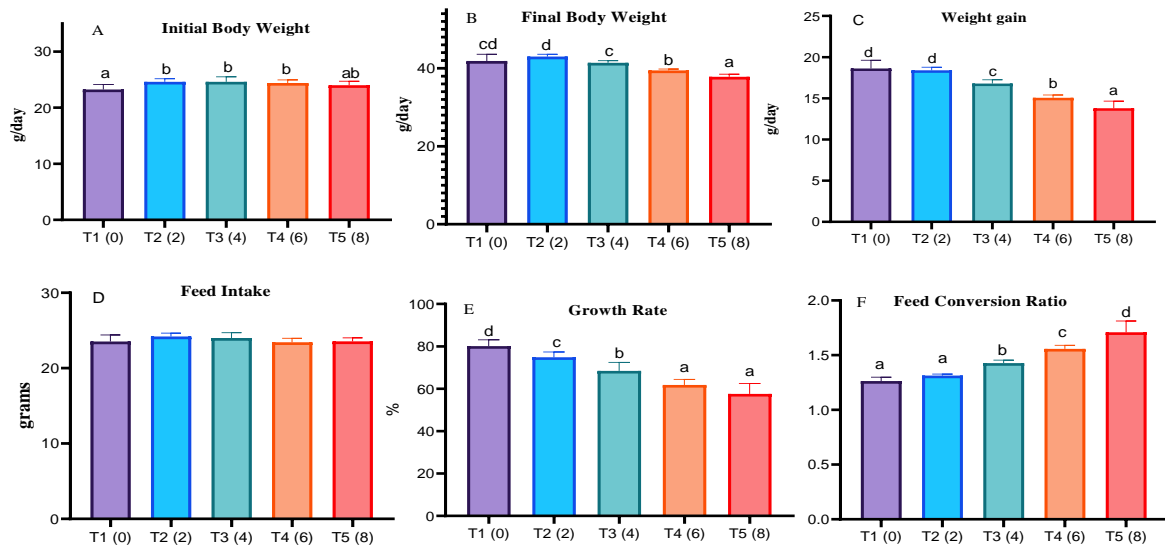


Figure 1: The effect of varying levels of salinities on the growth performance of *C. gariepinus*. During the 60-day experiment. Note that data are displayed as means with standard error (SE). Significant differences ($P < 0.05$ - 0.01) are indicated by distinct superscripts (a, b, c, and d). A; for Initial body weight, B; for final body weight, C; for weight growth, D; for feed intake, E; for growth rate, and F; for feed conversion ratio.

The experimental procedure of the effects of different concentrations of salinity on the proximate composition is presented in (Table 2 and Figure 2; A, B, C, and D). The crude protein data experienced a significant increase with the elevation of salinity levels. Beginning from (15.31 ± 0.07%) in the T1 (0 ppt), it gradually elevated to (19.78 ± 0.17%) at the high concentration of the salinity level (T5; 8 ppt). This signifies a substantial rise ($p < 0.05$) of approximately 29%. On the other hand, crude fat revealed an inverse relationship with salinity. The highest content of fat was observed in the control group T0 at (8.62 ± 0.07%), which progressively diminished to (2.94 ±

0.30%) in T5, marking a substantial reduction of about 66% ($p < 0.05$).

The moisture content in the flesh of fish also has been shown to exhibit a positive correlation with salinity levels. It raised from (72.98 ± 0.20%) in the T1 group to (78.72 ± 0.63%) in T5, representing a significant elevation of nearly 8% ($p < 0.05$). In contrast, ash content exhibited a downward trend with increased salinity. The group (T1) exhibited a significant ash content of (2.02 ± 0.04%), which subsequently decreased to (1.16 ± 0.02%) in T5, indicating a significant reduction of approximately 43% ($p < 0.05$).

Table 2: Effect of different salinity levels on proximate composition of freshwater *C. gariepinus* at different salinity levels (0-ppt, 2-ppt, 4-ppt, 6-ppt, and 8-ppt) during the 60-day experiment.

PC Parameters	Treatments				
	T1 (0-ppt)	T2 (2-ppt)	T3 (4-ppt)	T4 (6-ppt)	T5 (8-ppt)
Crude Protein (%)	15.31±0.07 ^a	16.28±0.24 ^b	17.44±0.21 ^c	18.34±0.09 ^d	19.78±0.17 ^d
Crude fat (%)	8.62±0.07 ^e	6.92±0.31 ^d	5.51±0.07 ^c	4.46±0.12 ^b	2.94±0.30 ^a
Moisture (%)	72.98±0.20 ^a	74.24±0.05 ^b	75.34±0.11 ^c	76.54±0.11 ^d	78.72±0.63 ^e
Ash (%)	2.02±0.04 ^e	1.74±0.02 ^d	1.52±0.03 ^c	1.28±0.02 ^b	1.16±0.02 ^a

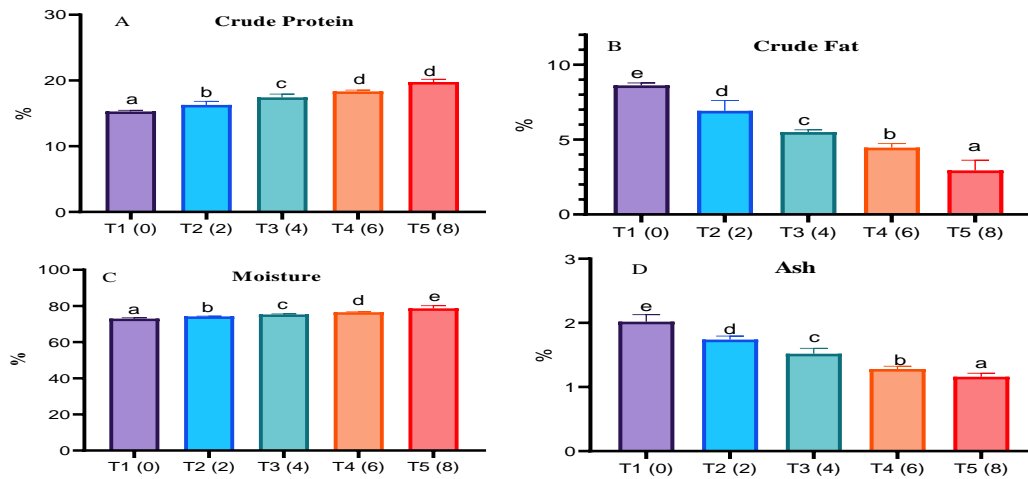


Figure 2: The effect of varying levels of salinities on the proximate composition of *C. gariepinus*. During the 60-day experiment. Note, data are displayed as means with standard error (SE). Significant differences ($P < 0.05$ - 0.01) are indicated by distinct superscripts (a, b, c, and d). A; for crude protein. B; for Crude fat. C; for moisture. D; for Ash.

DISCUSSION

The data suggest that *C. gariepinus* exhibits the best growth when reared in moderately saline with a salinity of around 2 ppt. These findings agree with the study by Mugwanya et al. (2023), which suggests that reduced salinity levels can enhance growth rates in fish. The fact that fish performance gradually reduced at a salinity of 0 ppt compared to 2 ppt suggests a potential threshold salinity level exists that is most beneficial for this species. Growth performance diminished when the salinity concentration elevated beyond 2 ppt, with the most pronounced decrease revealed at the 6 ppt and 8 ppt treatments. These findings are consistent with those of Lisachov et al. (2023), who indicated that salinity levels exceeding 2 ppt can reduce the growth of *C. gariepinus*. The reduced growth observed at high salinities can likely be attributed to an increased osmoregulatory stress, which forces the fish to divert energy away from growth and towards maintaining internal balance (Patel et al., 2023).

Data from WG and GR further confirm that *C. gariepinus* grows better in freshwater to slightly brackish conditions. The highest WG and GR recorded in the 0 ppt group, with the 2 ppt group close behind, further demonstrates the species' capacity to thrive in freshwater. These findings are consistent with those of Harika et al. (2024), who reported that increased salinity levels stress fish and negatively impact their growth and physiological functions. Feed intake (FI) remains stable over all groups, suggesting that the indicated differences in growth were not mainly due to the alternations in food consumption. This indication is important because it suggests that the reduced efficiency of the growth at higher salinity levels is likely due to increased metabolic demands for osmoregulation rather than a decrease in appetite. Similar findings were reported by Takagi et al. (2020), who suggested that the stress associated with osmoregulation at higher salinities may decrease the efficiency of food utilization.

The increasing FCR revealed with high salinity levels indicates a decline in the efficiency of fish in converting feed into body mass. This outcome agrees with the findings of Mandal et al. (2020) on *Pangasius hypophthalmus*, who found that FCR increased with increasing salinity. *C. gariepinus* shows distinct salinity tolerance and optimal growth compared to other species. Hassan et al. (2023) and Rodrigues Maciel et al. (2024) observed that striped catfish achieved their highest weight at 9-10% salinities. Similarly, Takagi et al. (2021) indicated that the *Channa striata* exhibited notable growth rates at 0 and 3% salinities. These differences highlight the importance of

conducting species-specific research in aquaculture to optimize growth conditions.

The increase in CP with elevated salinity agrees with results from other studies. Hassan et al. (2023) indicated a similar pattern in *C. gariepinus*, with PC reaching its highest point at 10 ppt. This increase could be a physiological response to the osmotic stress caused by high salinity levels; proteins play a crucial role in osmoregulatory and protect the cell under unfavorable environmental conditions. Conversely, the decrease in CF with salinity elevation is consistent with observations by Kawamura et al. (2017) and Zidan et al. (2022). This diminished lipid content may be attributed to the higher energy expenditure required for osmoregulation in more saline conditions, leading to the depletion of fat reserves. The inverse relationship between protein and fat content suggests a shift in the fish's energy metabolism to adapt to the changing environmental conditions. The observed elevate in moisture content with rising salinity is consistent with findings by Mandal et al. (2020). This pattern could be seen as a mechanism to maintain osmotic equilibrium in more salinity conditions, with the fish retaining additional to counteract the external osmotic pressure. The diminishing ash content with elevating saline water is consistent with the findings of Mandal et al. (2020). This decrease could potentially be attributed to variations in the metabolism of trace elements. The behavioural observations, which include active feeding (8 ppt) salinity and the appearance of stress indicators at higher levels, provide empirical evidence consistent with the findings by Kawamura et al. (2017) and Zidan et al. (2022).

CONCLUSION

This study confirms that *C. gariepinus* performs optimally in freshwater conditions with low salinity, specifically between 0 and 2 ppt. The fish exhibit the best growth rates and feed conversion efficiency at these levels. Conversely, increased salinity negatively affects fish growth, lowering final weight and feed conversion ratios. Elevated salinity also alters the nutritional composition of the fish, increasing protein and moisture content while decreasing fat and ash levels.

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