



Size and diet of *Oreochromis niloticus* in an urban drainage channel in Santos, State of São Paulo, Brazil

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ABSTRACT. The introduction of non-native species can significantly impact ecosystems, with the Nile tilapia (*Oreochromis niloticus*) serving as a notable example. Despite its widespread presence in various rivers and reservoirs, its occurrence, particularly its dietary habits in drainage channels, remains insufficiently explored. Therefore, the present study investigated the ecological dynamics of *O. niloticus* within an urban drainage channel system in Santos, São Paulo, Brazil. Sampling of 56 individuals across four campaigns revealed a dynamic population structure, characterized by seasonal variation in size, notably the prevalence of mature females during periods of increased rainfall. The sex ratio did not deviate significantly from the expected ratio (1:1). Analyses of size and weight demonstrated significant fluctuations across sampling months, suggesting potential variations in reproductive activities and population dynamics. The condition factor remained remarkably consistent, indicating stable health conditions despite intra-annual variations. Dietary analysis indicated a distinct preference for detritus, consistent with the species' characteristic foraging behavior on the channel bed. The correlations between water turbidity, stomach food item richness, and preference for detritus suggest that, in murkier waters, foraging on sediment-rich organic matter becomes more productive. Regarding shifts in diet composition, this species exhibited opportunistic feeding habits throughout all seasons, highlighting its adaptability to urban environments.

Keywords: freshwater fish; invasive species; trophic ecology; Nile tilapia.

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Introduction

The presence of the Nile tilapia, *Oreochromis niloticus* (Linnaeus, 1758), a fish species native to Africa, in non-native waters has been shown to produce far-reaching ecological consequences (Bittencourt et al., 2014). This species, known for its year-round reproduction and opportunistic habits, has found its way into various rivers (Linde et al., 2008), reservoirs (Figueredo & Giani, 2005), and even high-salinity environments (Franco et al., 2023) throughout Brazil because of its robust tolerance to environmental fluctuations (Lima et al., 2020). The introduction of *Oreochromis niloticus* is particularly impactful, not only leading to competitive displacement and predation of native species, but also potentially introducing pathogens and parasites and acting as a catalyst for broader environmental changes (Vicente & Fonseca-Alves, 2013).

In the coastal area of the municipality of Santos, State of São Paulo, Brazil, a drainage system consisting of nine channels has been constructed to manage stormwater. This system, which traverses the plain, functions to direct water outward from the island. The central portion of the system is elevated, and the water is partly directed into the Santos Bay and partly into the Santos Channel (Coelho et al., 2012). However, there remains a significant gap in our understanding of the ecological consequences of the presence of Nile tilapia in these channels. Tilapias frequently outcompete other species in habitats affected by pollution or other human impacts. Their competitive dominance can have a substantial impact on native species, potentially leading to their extinction from the invaded environments (Attayde et al., 2007). Furthermore, the introduction of such non-native species has the potential to render ecosystems vulnerable to subsequent invasions, thereby altering the biotic and abiotic characteristics of the invaded habitats (Orsi et al., 2016). In

addition, this territorial species exhibits competitive behavior over resources such as food sources and mating partners. Individuals of *O. niloticus* exhibit reproductive behaviors typical of cichlids, characterized by aggression, dominance in territorial disputes, nest construction, and competition for mating (Medeiros et al., 2005).

An investigation into the dietary habits of species, particularly in habitats impacted by human activities, such as these channels, is imperative. For instance, Channel 4 on Siqueira Campos Avenue, a Santos channel, has been found to contain elevated levels of contamination indicators such as *Escherichia coli*, indicating significant sewage system pollution in the area (Coelho et al., 2012). The study also documented high turbidity, conductivity, and ammonia nitrogen concentrations in this channel (Coelho et al., 2012). Consequently, the present study aimed to investigate the body size and diet of *O. niloticus* within these artificial habitats, offering insights into the behavior of the species and its impact on the Santos channels.

Material and methods

Study area and sampling

Sampling was conducted in Santos, State of São Paulo, Brazil, specifically on Channel 4 (Figure 1), one of the city channels. This channel spans from the brackish waters of the port channel to the saline waters of the beach. This artificial channel measures approximately 2.5 km long and 11 m wide and varies in depth from 0.1 m during dry periods to 2.0 m during rainfall events or high tides. Fish were captured during four sampling campaigns (March, May, November 2022, and March 2023) using cast nets during low-tide events. To ensure consistency in the method, the same researcher cast the net (7 cm mesh size) five times on each occasion. The selection of low tide as the time point for sampling was driven by constraints; high tide was not a viable option because of the substantial increase in water volume, which significantly hindered the visibility of individual specimens. Cast nets were used as the sampling method because the channel is an urban environment with steep banks, which makes sampling within it impractical. This was further compounded by the accumulation of garbage at the bottom.

Captured fish were then anesthetized using a eugenol solution, comprising 1–1.5 mL of clove oil solution per liter of water (5 mL of clove oil + 95 mL of 96–99° GL alcohol). Total length (cm) and weight (g) of each individual were measured. The total length was adopted instead of the standard length because all specimens had caudal fins under excellent conditions. Necessary authorizations for the collection were obtained from SISBIO (permit number: 66427-1) and the Municipal Government of Santos (Process number: 068490/2021-68). All animal handling procedures were approved by the Ethical Committee on Animal Use at UNISANTA (CEUA 01/2021).

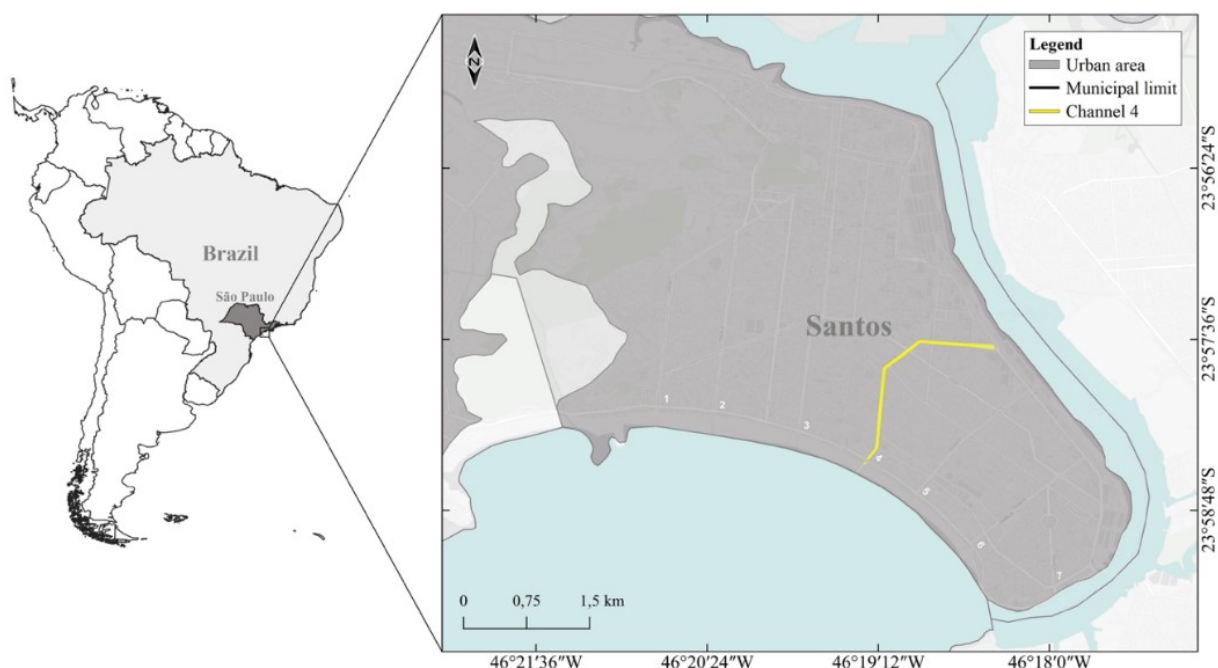


Figure 1. Location of the sampling site, Channel 4, in the municipality of Santos, State of São Paulo, Brazil.

After collection, fish were euthanized by increasing the dose of anesthesia, placed in labeled plastic bags, and preserved in 10% formalin for approximately 48 hours. Samples were then transported to the Marine and Coastal Organism Biology Laboratory (LABOMAC) of UNISANTA, where they were stored in containers of 70% alcohol for further analysis. Simultaneously with fish sampling, water quality parameters - pH, dissolved oxygen, conductivity, turbidity, temperature, and salinity - were measured using a multi-parameter probe (Horiba U-52G).

Laboratory procedures

Following the dissection, the stomach repletion degree (SRD) was recorded, based on a scale where 1 represents an empty stomach, 2 denotes a partially filled stomach, and 3 indicates a completely full stomach. Stomachs containing food were extracted and preserved in 70% alcohol for content analysis. The sex and gonadal maturation stage of each specimen was determined macroscopically and classified as follows: A for immature gonads (juveniles), B for maturing or resting gonads (adults), C for mature gonads (breeding individuals), and D for spent gonads (post-breeding individuals) (Vazzoler, 1996).

Stomach contents were examined under a stereomicroscope and identified to the lowest taxonomic level possible, using specialized literature (Bicudo & Menezes, 2006; Costa et al., 2006; Mugnai et al., 2010; Triplehorn & Johnson, 2011). The Food Preference Degree (FPD) method (Braga, 1999) was used to quantify dietary preference. In the FPD classification, if only one specific food item is ingested, it is assigned a value of 4. If multiple items are present, the predominant item is assigned a value of 3, the intermediate item is assigned a value of 2, and the least represented item is assigned a value of 1 in each stomach analyzed. Dietary preference was then determined using the $S_{(i)}/N$ equation. Here, $S_{(i)}$ represents the accumulation of values assigned to a particular component i , and N represents the number of stomachs examined. An item is considered absolutely preferred when $FPD = 4$, highly preferred when $3 < FPD < 4$, preferred with the presence of others when $2 < FPD < 3$, secondary when $1 < FPD < 2$, and occasional when $FPD < 1$ (Braga, 1999).

Data analysis

The Fulton condition factor (K) was calculated using the following equation: $K = \frac{10^5 \times \text{weight (g)}}{\text{total length (mm)}^3}$. A chi-square test was used to assess whether the sex ratio of the species differed from the expected 1:1 ratio. A Student t-test was used to determine whether there were significant differences in body size between males and females. To detect significant differences in total length, weight, and condition factor between sampling months, one-way analysis of variance (ANOVA) was performed for each case. Spearman's rank correlation was used to associate environmental factors with dietary richness and detritus FPD because the residuals of the model did not meet the normality assumption.

A Jaccard dissimilarity matrix was constructed using the following formula $J = \frac{a}{(a+b+c)}$, where a is the number of food items present in both samples, b represents items present only in the first sample, and c represents items present only in the second sample. A permutational multivariate analysis of variance (PERMANOVA) was then applied to this Jaccard-based distance matrix to investigate whether diet composition changed between sampling months. PERMANOVA was run with a comprehensive set of 999 permutations to ensure a rigorous evaluation of statistical significance.

Results

A total of 56 *O. niloticus* individuals were collected in March 2022 ($N = 10$), May 2022 ($N = 12$), November 2022 ($N = 16$), and March 2023 ($N = 18$). The sample consisted of 22 female, 22 male, and 12 immature (undetermined) specimens. The sex ratio did not deviate significantly from the expected ratio (1:1) ($\chi^2 = 0$, $df = 1$, $p = 1$). A notable presence of mature females was observed in March of both 2022 and 2023, particularly in March 2023, when 8 out of 18 collected individuals were classified as mature females. Conversely, May 2022 exhibited nine immature specimens, with only three individuals that could be determined for their sex and maturity stage (Figure 2).

The average total length (TL) of the specimens was 25.89 cm, with a standard deviation (SD) of 6.67 cm, and the average weight was 395.58 g (SD = 274.71 g). A subsequent analysis revealed no statistically significant differences in body size between males and females ($t = -1.43$, $p = 0.16$). However, there was a significant difference in TL across the four collection periods ($F_3 = 15.2$, $p < 0.01$), with May 2022 differing significantly

from the other collections due to its lower values ($p < 0.01$) (Figure 3A). The weight also showed a significant variation across samples ($F_3 = 6.71$, $p < 0.01$), with May 2022 being the only month that differed significantly from all other samples ($p < 0.05$) (Figure 3B). However, the condition factor remained consistent across all samples ($F_3 = 1.69$, $p = 0.18$) regardless of the weight and length measurements (Figure 3C).

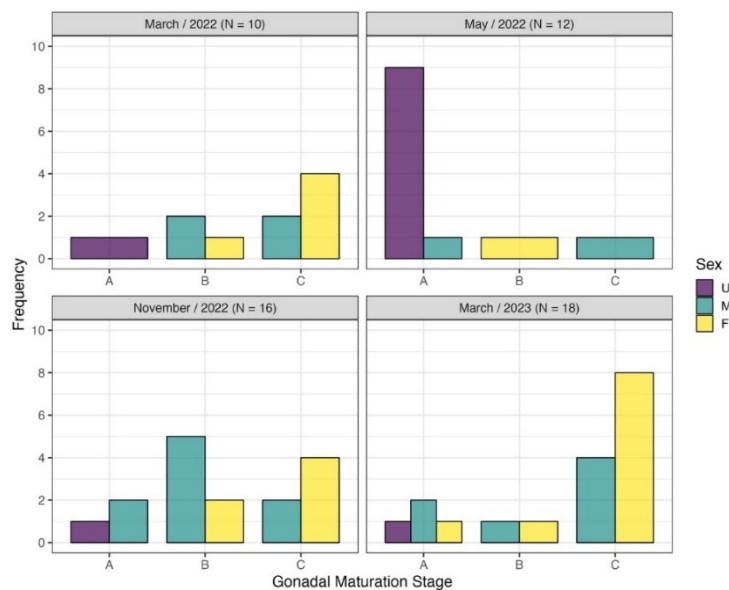


Figure 2. Frequency of occurrence of gonadal maturity stages of males and females of *Oreochromis niloticus* in all sampling months. U = Undetermined. M = Male. F = Female.

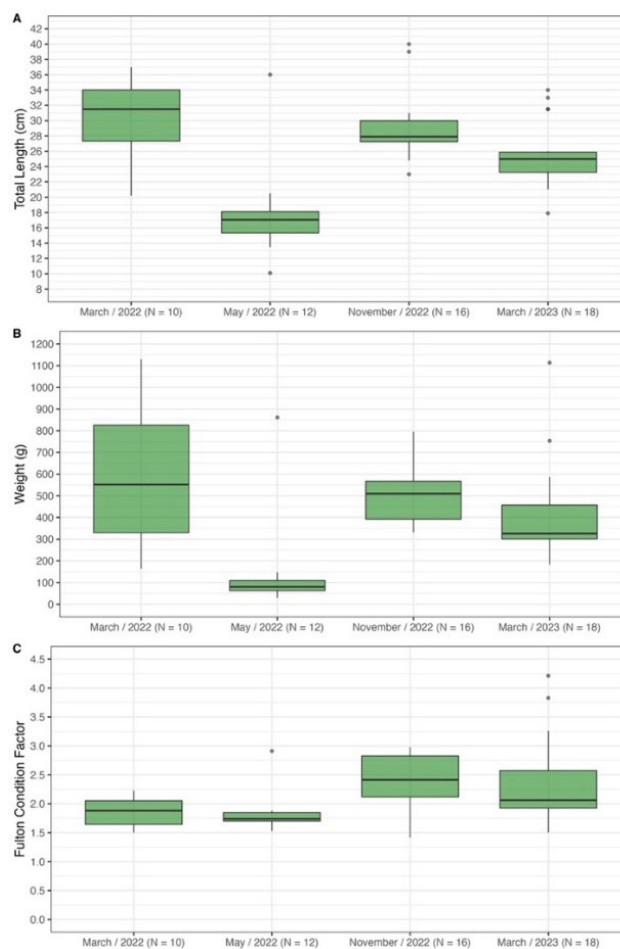


Figure 3. Boxplot containing the minimum, maximum, median, and quartile values of the total length (A), weight (B), and Fulton condition factor (C) of *Oreochromis niloticus* sampled in the Santos channel.

Regarding the diet, 49.1% of the individuals had a stomach repletion degree (SRD) of 3, 27.3% had an SRD of 2, and 23.6% had an SRD of 1. A total of 42 stomachs contained food (SRD 2 and 3), and nine food items were identified. Most of these items were classified as occasional, with only algae as secondary, and detritus as preferred with the presence of others, according to the Food Preference Degree (FPD) (Table 1). The diet composition exhibited no significant alterations between the sampling periods ($F_{3,52} = 1.539$, $p = 0.13$). A negative correlation was observed between increased water turbidity and the richness of food items ($S = 42,460$, $p < 0.01$, $\rho = -0.451$) (Figure 4A), and a positive correlation was identified between water turbidity and the FPD attributed to detritus ($S = 16,513$, $p < 0.01$, $\rho = 0.435$) (Figure 4B). Means and standard deviations for each environmental parameter are listed in Table 2.

Table 1. Food items found in the stomachs of *Oreochromis niloticus* from Channel 4 in Santos, State of São Paulo, along with their respective Food Preference Degree (FPD) values (Braga, 1999), and classification.

Items	FPD	Classification
Hymenoptera		
Formicidae	0.089	Occasional
Diptera (larvae)		
Chironomidae	0.125	Occasional
Diptera (pupae)	0.017	Occasional
Diptera (adults)	0.053	Occasional
Gastropoda	0.017	Occasional
Teleostei		
<i>Menticirrhus littoralis</i>	0.017	Occasional
Algae	1.607	Secondary
Plant material	0.714	Occasional
Detritus	2.696	Preferred with the presence of others

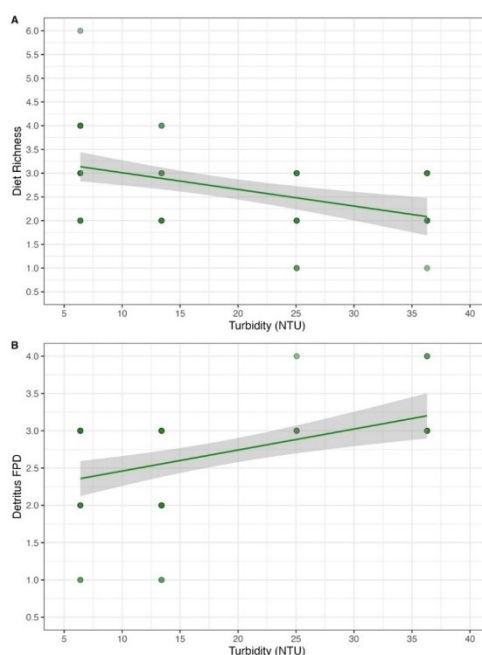


Figure 4. Relationship between turbidity (NTU) and dietary richness found in each stomach (A), and between turbidity (NTU) and the Food Preference Degree (FPD) attributed to detritus (B) found in the diet of *Oreochromis niloticus*.

Table 2. Mean and standard deviation of the environmental variables measured in Channel 4 in Santos, State of São Paulo, simultaneously with *Oreochromis niloticus* sampling.

Sample	Variable	Mean	Standard deviation
March 2022	Conductivity (mS cm ⁻¹)	1.66	0.08
	DO (mg L ⁻¹)	6.11	2.96
	pH	5.71	0.09
	Salinity (ppt)	0.88	0.40
	Temperature (°C)	28.95	0.29
	Turbidity (NTU)	26.9	7.83

May 2022	Conductivity (mS cm ⁻¹)	1.03	0.02
	DO (mg L ⁻¹)	5.98	2.50
	pH	7.83	0.07
	Salinity (ppt)	0	0
	Temperature (°C)	25.29	3.61
	Turbidity (NTU)	36.30	16.82
November 2022	Conductivity (mS cm ⁻¹)	1.42	0.47
	DO (mg L ⁻¹)	6.37	1.37
	pH	7.02	0.36
	Salinity (ppt)	0.7	0.28
	Temperature (°C)	26.95	1.09
	Turbidity (NTU)	6.23	1.08
March 2023	Conductivity (mS cm ⁻¹)	15.45	9.60
	DO (mg L ⁻¹)	7.03	2.13
	pH	6.46	0.25
	Salinity (ppt)	9.15	5.90
	Temperature (°C)	29.04	0.15
	Turbidity (NTU)	13.35	0.65

Discussion

The prevalence of mature female tilapia, particularly in March, can be attributed to seasonal rain. It is plausible that these rains led to an increase in the water level of the Santos Channel, resulting in an augmented volume of food and a deeper water column that is conducive to increased habitat availability. The mean weight and length of the sampled individuals were comparable to those documented in the literature for invasive *O. niloticus* populations (Bittencourt et al., 2014). However, a notable variation in *O. niloticus* size and weight was observed between samplings, with May exhibiting significant disparities, likely attributable to the prevalence of immature specimens in that month, resulting in smaller sizes and lighter weights. Notably, despite these variations in size and weight, the condition factor of *O. niloticus* remained consistent across all collections. This suggests that, irrespective of seasonal or intra-annual variations, the health conditions of these individuals in the urban channel remain relatively stable. Nevertheless, these findings should be interpreted with caution, as they were based on a relatively small sample size of 56 individuals. Although this sample provides valuable insights, it may not fully capture the population's variability across different environmental conditions.

In terms of diet, the strong preference for detritus is likely due to the tendency of tilapia to forage on the substrate of water bodies (Oso et al., 2006). This behavior may be more pronounced in murkier waters, where visibility is limited and foraging in sediment-rich organic matter becomes more fruitful. The correlation between water turbidity and the richness of food items in the stomach further supports this notion. The bioturbation process of the sediment at the bottom of the channel, which may occur because of this foraging, could even release contaminants, as already reported by Coelho et al. (2012). In clear waters, tilapia may have greater opportunities to feed on items from the surrounding vegetation, even in urban areas, such as ants and gastropods (Oso et al., 2006). The presence of various items, such as plant material, Gastropoda, Hymenoptera, and adult Diptera in the drainage channel may be attributed to the impact of rainwater runoff from the surrounding urban area. Notably, the only autochthonous items identified were Diptera larvae and algae. This finding aligns with the documented resilience of these macroinvertebrates to variations in water quality (Machado et al., 2015). On the other hand, the presence of algae may be related to the confluence of fresh and saltwater, as it is an important part of the species' diet in natural environments (Teshfahun & Temesgen, 2018).

A critical revelation was the discovery of *O. niloticus* preying on native Scianidae fry, specifically *Menticirrhus littoralis* (Holbrook, 1847), although it was recorded in only one stomach. This species is of significant value for both commercial and recreational fisheries in Brazil (Braun & Fontoura, 2004). This finding emphasizes the potential ecological impact of this invasive species, even on native marine species, and highlights the need for urgent monitoring and management of its presence in these channels. The diet composition remained unchanged throughout the study, suggesting a generalistic feeding habit for *O. niloticus* throughout the year. This is consistent with the resilience of this invasive species that feeds on almost any item possible in such harsh environments as urban channels (Khallaf et al., 2003).

Moreover, the challenging environmental conditions encountered, including low dissolved oxygen levels in all months sampled, elevated temperatures, and high salinity in March 2023, demonstrate the remarkable

resilience of the species and indicate that this species poses a challenge to the drainage ecosystems of the Santos channels. Previous studies have documented short-term occurrences of this species in regions with elevated salinity (Franco et al., 2023). Although the salinity levels recorded in March 2023 are not exceptionally high, ongoing monitoring of these channels is imperative to gain a more complete understanding of potential species incursions. In conclusion, the results presented highlight the need for further research. Investigating the long-term ecological impacts and evaluating potential management strategies to control this invasive species in channels are promising directions for future studies.

Conclusion

The presence of *O. niloticus* in the urban drainage channels of Santos highlights the remarkable adaptability of the species to harsh environments, including low oxygen levels, elevated salinity, and pollution. Despite seasonal fluctuations in the size and weight of individuals, the condition factor remained stable, indicating that the species maintained its health under urban conditions. The species' opportunistic diet and its potential impact on native species, such as predation on *M. littoralis* larvae, underscore the ecological concerns associated with its introduction. Therefore, this study emphasizes the urgent need for monitoring and managing *O. niloticus* in these channels to mitigate its long-term ecological impact.

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