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Bioremediation of slaughterhouse wastewater: Evaluation of nitrogen pollutant removal using constructed wetlands

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Abstract: The increasing generation of slaughterhouses wastewater is posing a serious threat to ecosystems and human health due to high levels of contaminants such as total nitrogen (TN), NH_4^+, NO_3^- and NO_2^-, which, if not properly treated, can cause environmental problems such as eutrophication. Constructed wetlands (CWs) have emerged as an effective and cost-efficient alternative to conventional wastewater treatment technologies, including slaughterhouse wastewater. This study evaluated the efficiency of CWs in the removal of nitrogenous compounds in slaughterhouse wastewater using the plant species *Typha latifolia* and *Heliconia latispatha*. Eight horizontal subsurface flow constructed wetlands (HSF-CWs) units were implemented with different species combinations, and nitrogen pollutant concentrations were monitored over a one-year period. Results showed that the polyculture of *T. latifolia* and *H. latispatha* achieved the highest TN (64-65%) and NH_4^+ (89%) removal efficiency, while systems with individual *T. latifolia* and *H. latispatha* showed intermediate efficiencies of 57% and 48%, respectively. For nitrate (NO_3-), the polyculture achieved an efficiency of 91%, while *T. latifolia* reached 78% and *H. latispatha* 68%. Finally, in nitrite removal (NO_2^-), the polycultures showed a removal efficiency of 92%, while *T. latifolia* and *H. latispatha* achieved efficiencies of 82% and 73%, respectively. In comparison, the control presented the lowest efficiencies in all parameters (33-65%). This study concludes that HCs, planted with *T. latifolia* + *H. latispatha*, are a viable and economical alternative for the treatment of slaughterhouse wastewater, offering a more sustainable option compared to more complex and expensive technologies, such as electrocoagulation or nanofiltration systems. The use of polycultures and the combination of technologies is recommended to maximize treatment efficiency.

Keywords: slaughterhouse wastewater, nitrogenous contaminants; constructed wetlands, ornamental plants, bioremediation

Introduction

The rapid generation of industrial wastewater, especially in meat production, is placing increasing pressure on communities and ecosystems, threatening not only environmental balance but also the health and future of entire generations (Boukouvalas *et al*., 2024). Wastewater from slaughterhouses contains high concentrations of pollutants, including total nitrogen (TN), ammonium (NH_4^+), nitrates (NO_3^-), and nitrites (NO_2^-). If these compounds are not properly treated, they can have significant environmental impacts, such as eutrophication and contamination of surface and groundwater bodies (Brennan *et al*., 2021; Silveira *et al*., 2021). In addition, nitrogen compounds have been identified as one of the main causes of water quality degradation in various regions of the world (Wang *et al*., 2024).

Proper treatment of effluents from slaughterhouses is crucial to mitigate their environmental impact. Nature-based solutions (NBS), such as constructed wetlands (CWs), have emerged as an effective and sustainable alternative to conventional wastewater treatment methods (Agaton & Guila, 2023). These systems are attractive not only because of their operational simplicity and low cost, but also because of their high effectiveness in removing pollutants such as nitrogen, which is one of the main causes of eutrophication in water bodies (Waly *et al*., 2022).

CWs have been widely used to treat various types of wastewaters, demonstrating their versatility and effectiveness in a range of contexts. They have proven effective in treating domestic and industrial wastewater (Herazo *et al*., 2021), as well as in removing contaminants from specific industries, such as heavy metals (Yu *et al*., 2022) and textile dyes (Patil *et al*., 2022). They have also been applied in the treatment of agricultural runoff and pesticides (McCalla *et al*., 2022), petroleum hydrocarbons (Taha *et al*., 2023), and pharmaceuticals (Hu *et al*., 2021), obtaining favorable results in reducing nitrogen compounds present in the effluents studied.

However, although CW shave proven to be efficient in treating various types of wastewater, their ability to treat slaughterhouse effluents, which have a higher organic load and nitrogen concentration, has not yet been fully explored, particularly regarding nitrogen removal performance (Fang *et al*., 2022).

This study focused on evaluating the efficiency of a CW system for treating slaughterhouse wastewater, with special attention to the removal of nitrogenous contaminants such as TN, NH_4^+, NO_3^-, and NO_2^-. To this end, macrophytes such as *Typha latifolia* and *Heliconia latispatha* were used, species that have demonstrated a great capacity to improve filtration and nutrient removal in wetland systems (Aguilar-Cortés *et al*., 2025; Sandoval-Herazo *et al*., 2021). In addition, the results obtained were compared with other conventional treatment methods to position constructed wetlands as a viable and economical solution for treating effluents from slaughterhouses.

Bioremediation using CWs is an innovative and environmentally friendly option for treating slaughterhouse wastewater. This study highlights the significant potential of this nature-based solution (NBS) within the agro-industrial sector, addressing one of today's greatest environmental challenges. By focusing on the efficiency of constructed wetlands in removing nitrogen from slaughterhouse effluents, this research not only expands knowledge about their applicability but also provides key guidelines for their implementation, positioning them as a sustainable, economical, and high-impact alternative for the meat industry.

Materials and Methods

Study site

This study was conducted at the facilities of the National Technological Institute of Mexico (TecNM), Misantla Campus, Veracruz, Mexico. The research was carried out during 2023, from January to December. The area where the work was carried out is characterized by a warm and humid climate, with an average annual temperature of approximately 22.7 °C. In addition, the region receives an average annual rainfall of 2036.4 mm, which influences the environmental conditions of the site. The campus is located 300 meters above sea level (Figure 1), which provides a relevant geographical context for the study of constructed wetlands and their capacity to treat slaughterhouse wastewater.

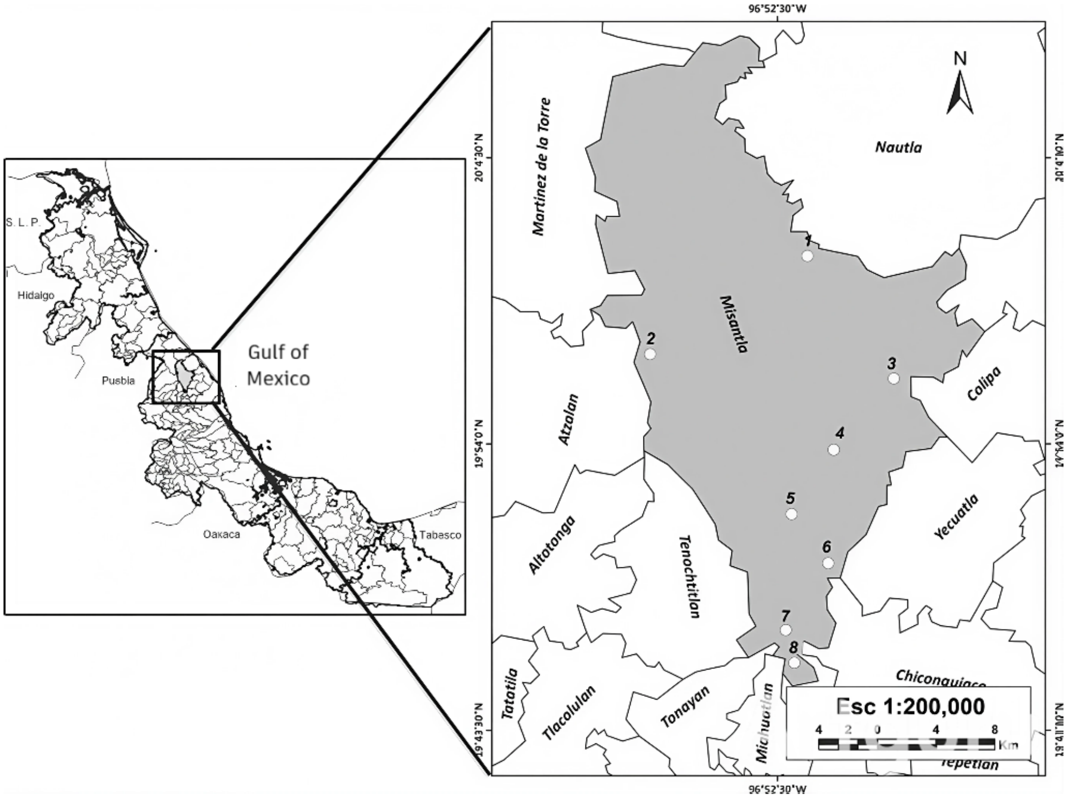


Figure 1. Geographical location of the study area

Description of the experimental units

The wastewater used in this study was obtained from wastewater from a slaughterhouse located 1.2 km from the experimental area. This water was collected in a 1,200-liter sedimentation tank, which was placed 50 cm above the level of the experimental units to ensure gravity flow to the constructed wetlands.

The treatment system consisted of eight subsurface horizontal flow wetland units (HSF-CWs), constructed on a mesocosm scale using reinforced concrete. The dimensions of each experimental unit were 150 cm long, 40 cm wide, and 100 cm deep, with a water saturation depth set at 65 cm. The units were arranged in a linear alignment, with each contiguous cell being a direct replica of the previous one.

The units were assigned according to the type of experimental treatment: a control cell (without vegetation, but with the same filter medium as the others), a cell with polyculture of the species *Typha latifolia* and *Heliconia latispatha*, a cell with monoculture of *Typha latifolia*, and a cell with monoculture of *Heliconia latispatha*. This experimental design allowed for a detailed and replicated assessment of the impact of different plant configurations on the performance of wetlands for contaminant removal.

The filter medium used in all units was red tezontle, with a particle diameter between 5 and 8 mm and a porosity of 0.5, which facilitated adequate interaction between the water and plant roots. All experimental units were operated with a hydraulic retention time (HRT) of 5 days, which allowed the simulation of adequate conditions for the bioremediation process of wastewater in the constructed wetland system.

Vegetation development

Monitoring plant development was essential to understanding the functional role of plants in removing nitrogen compounds within the constructed wetland system. To this end, periodic assessments of the growth of the plant species used were carried out from the initial establishment of the plants until the end of the experimental period.

The variables considered included plant height and plant density. The first parameter was determined using standardized measurements with an UltraTech® digital caliper, while the number of flowers was recorded manually through direct inspection in the field. Measurements were taken every two months, allowing growth to be recorded over time and correlated with nitrogen removal efficiency parameters.

This continuous monitoring provided key data on the vigor and phenological status of the plants, which are fundamental elements in the efficiency of the bioremediation process, since greater biomass and plant activity are usually associated with a greater capacity to assimilate and transform nitrogen compounds present in wastewater.

Monitoring of water quality parameters

For a complete characterization of the quality of the collected wastewater, physicochemical analyses were performed to determine the levels of various key parameters, such as total nitrogen (TN), ammonium (NH_4^+), nitrate (NO_3^-), and nitrite (NO_2^-). The analyses were performed using an HI801 IRIS™ visible spectrophotometer (Hanna Instruments, USA) following the procedures established in standard water analysis methods (APHA/AWWA/WEF, 2012) to ensure the accuracy and reliability of the results obtained.

In addition, water environmental conditions, including temperature and pH, were monitored using a Hanna HI9811-51 portable multiparameter meter (HANNA® Instruments, Woonsocket, RI, USA). These parameters are essential for evaluating the environment in which bioremediation reactions take place and the behavior of contaminants. Likewise, dissolved oxygen (DO) was quantified using a specific Hanna HI9146-04 meter (HANNA® Instruments), which is crucial for understanding the dynamics of biodegradation and redox processes in the wetland. Environmental parameters were recorded daily using a DAVIS™ 6152 Vantage Pro2 automatic weather station.

These analyses provided essential data for interpreting the system's efficiency in removing nitrogenous contaminants, allowing correlations to be established between physicochemical conditions and the plants' ability to assimilate and transform the compounds present in wastewater.

Statistical analysis

To evaluate the system's effectiveness in removing nitrogenous contaminants, a randomized experimental design was used, which allowed for control of the variability of the experimental conditions and ensured the validity of the results obtained. The data collected during the study were analyzed using analysis of variance (ANOVA) to identify significant differences between the treatments and conditions evaluated. Subsequently, a comparison of means test was performed using Tukey's method, with a significance level of $p \leq 0.05$. This analysis provided detailed information on the differences in nitrogen removal efficiency between the different experimental groups, allowing the authors to determine which treatments were statistically superior in terms of their performance in the bioremediation of slaughterhouse wastewater. Statistical analysis was performed using IBM® SPSS® Statistics software, which ensured robustness and accuracy in the handling and processing of data obtained throughout the experimental period.

Results and Discussion

Characterization of the effluent

The results obtained in this study provide valuable information on the quality of the influent and the concentrations of nitrogenous pollutants present in slaughterhouse wastewater. Table 2 shows the physicochemical characterization of the influent. The differences between the average concentrations observed during the study period were significant for NH_4^+-N and NO_2^--N, but not significant for the other parameters analyzed ($p < 0.001$).

Regarding TN concentrations (250.3 ± 8.7 mg/L), the values found were within the ranges reported by other authors who used different conventional treatments in various countries (Bazrafshan *et al*., 2022; Fatima *et al*., 2021; Teo *et al*., 2023). For NH_4^+−N, the values obtained were similar to those reported by Phan *et al*. (2020), while for NO_3^-−N and NO_2^-−N, considerably higher values were recorded compared to those reported by the same author.

Table 1**.** Average concentrations of nitrogen contaminants in influent and effluent

Parameter	**Units**	**Influent**	***Typha latifolia***	***Heliconia latispatha***	**Polyculture**	**Control**
TN	mg/L	250.299 ± 8.669	106.502 ± 2.374	127.487 ± 5.085	88.494 ± 2.342	165.728 ± 6.212
NH_4^+−N	mg/L	172.801 ± 10.275	38.474 ± 1.915	58.239 ± 2.994	18.127 ± 1.056	92.315 ± 4.175
NO_3^-−N	mg/L	22.553 ± 1.240	4.790 ± 0.199	7.036 ± 0.436	2.077 ± 0.111	9.047 ± 0.385
NO_2^-−N	mg/L	7.721 ± 0.263	1.401 ± 0.034	2.032 ± 0.076	0.573 ± 0.024	3.280 ± 0.205

All values are expressed as mean ± standard error (n=12).

Environmental conditions and plant growth

The study was conducted in a tropical climate, which was reflected in the values of the environmental parameters recorded during the experimental period (Table 2). The climatic conditions observed indicate that both species were able to develop favorably, suggesting that environmental variability within these parameters did not represent a significant constraint on their growth.

On the one hand, *Heliconia latispatha* found a favorable environment for its growth, as the average temperature during the study period was within its optimal development range, which varies between 18 and 34 °C. Likewise, the relative humidity recorded exceeded the minimum threshold of 50% necessary for its proper development. In addition, high solar radiation, together with intense lighting, favored its growth, given that this species requires high levels of light for optimal physiological performance (Jagtap *et al*., 2024; Malakar & Biswas, 2022).

Table 2. Environmental parameters measured throughout the study

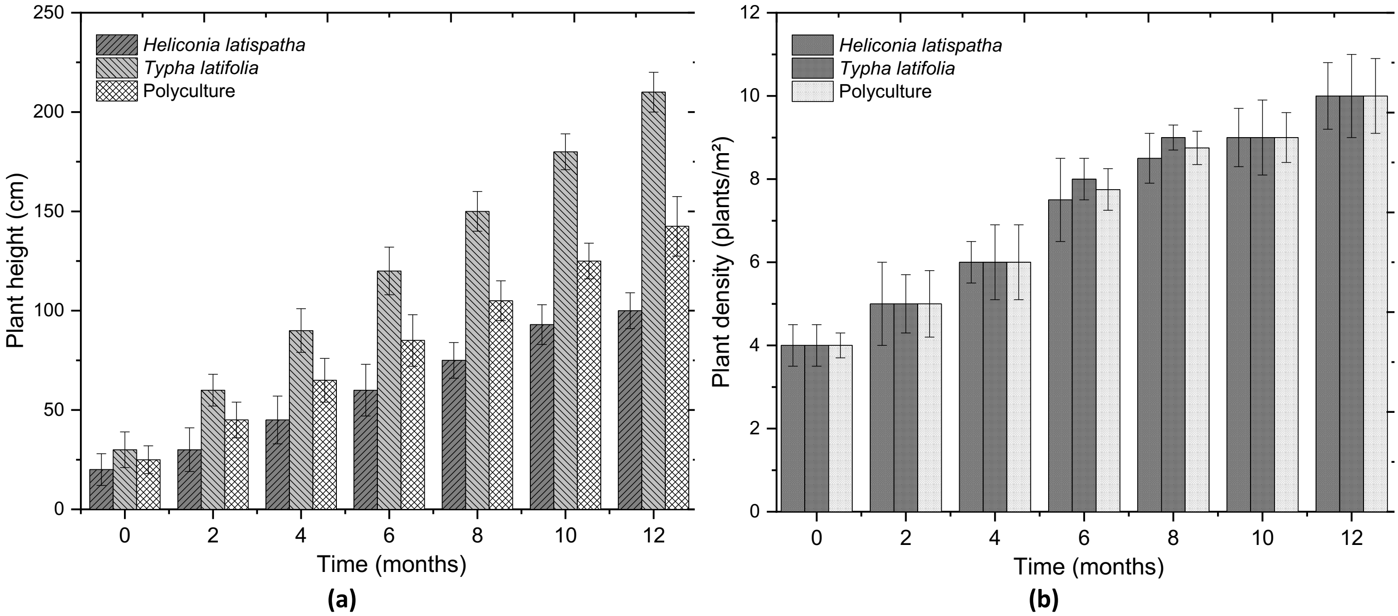
Parameter	**Average ± SD***
Ambient temperature (°C)	28.6 ± 5.6
Precipitation (mm)	120.0 ± 3.2
Solar radiation (W/m^2)	99.0 ± 11.1
Relative humidity (%)	95.1 ± 6.8
Light intensity (lux)	54748.7 ± 5937.5

*Standard Deviation, these values correspond to the entire study period (n=365).

On the other hand, *Typha latifolia*, known for its ability to adapt to different climates, also found suitable conditions for its development (Sesin *et al*., 2021). However, its growth is favored in tropical areas, as this species tolerates temperatures of up to 30 °C; therefore, the values recorded during the study remained within its upper tolerance limit. In addition, its ability to adapt to different levels of relative humidity allowed it to thrive in the environmental conditions recorded. Similarly, although it can develop under different levels of solar radiation, the values obtained coincide with those reported in other tropical regions where its successful growth has been documented (Wasko *et al*., 2022).

It is important to note that the recorded precipitation may have contributed to maintaining favorable water conditions for both species, especially for *Typha latifolia*, which thrives in aquatic ecosystems (Haldan *et al*., 2022). Taken together, these environmental factors suggest that the climatic conditions during the study were suitable for the development of both species, with environmental variability not representing a limiting factor. However, it is important to note that no specific analysis was conducted on the possible effect of precipitation on effluent dilution. Although daily monitoring of environmental parameters was carried out to ensure relatively stable conditions, it is recommended that future research include more detailed monitoring of the impact of factors such as precipitation, evapotranspiration, and other environmental parameters on treatment efficiency, especially in systems exposed to tropical climatic conditions.

However, in terms of plant development, *Typha latifolia* had the highest average height, reaching a maximum of 200 cm in monoculture and 210 cm in polyculture (Figure 2a).



(a) **(b)**

Figure 2. Vegetative development of plant species in monoculture and polyculture, (a) Height (b) Plant density

The maximum height values for *H. latispatha* ranged from 90 cm in polyculture to 124 cm in monoculture, which is characteristic of this species. However, in terms of plant density, *T. latifolia* showed the greatest increase in monoculture (Figure 2b); nevertheless, *H. latispatha* showed very similar results. Likewise, in polyculture, *H. latispatha* showed overall growth and plant density comparable to those of *T. latifolia*.

Monitoring of control parameters (pH, water temperature (T), and dissolved oxygen (DO))

Figures 3–5 illustrate the behavior of the control parameters (pH, T, and DO) monitored during the study period.
In this study, the pH values recorded in the influent throughout the experiment ranged from 7.9 to 8.1, with an average of 8.01 ± 0.6. The outlets of the HSF-CWs systems recorded an average value of 7.42 ± 0.45. Likewise, values of 8.13 ± 0.58 in the influent and 7.15 ± 0.24 in the effluent was also observed.

The pH analysis showed significant differences between the influent and the systems with vegetation (Figure 3). However, no significant differences were observed between the systems with vegetation or between these and the control. The results in the planted systems were similar, although *H. latispatha* differed significantly from the control.
The pH values recorded in the influent (7.9–8.1) and effluent (7.15–7.42) are within the ranges reported in similar studies for HSF-CWs systems. For example, previous research has documented pH values between 6.1 and 7.9 in wetlands planted with ornamental and common vegetation, such as *Canna indica* and *Typha domingensis* (Faisal *et al*., 2023), as well as *Juncus effusus* (Meng *et al*., 2024). The pH analysis showed that the CWs managed to reduce the pH of the influent to more neutral values in the effluent, which is beneficial for its discharge into receiving bodies or for its possible reuse (Sánchez *et al*., 2021). The significant differences observed between the influent and the systems with vegetation highlight the contribution of the biological and physicochemical processes inherent in these systems, such as microbial activity, plant absorption, and chemical precipitation processes (Banc *et al*., 2021).

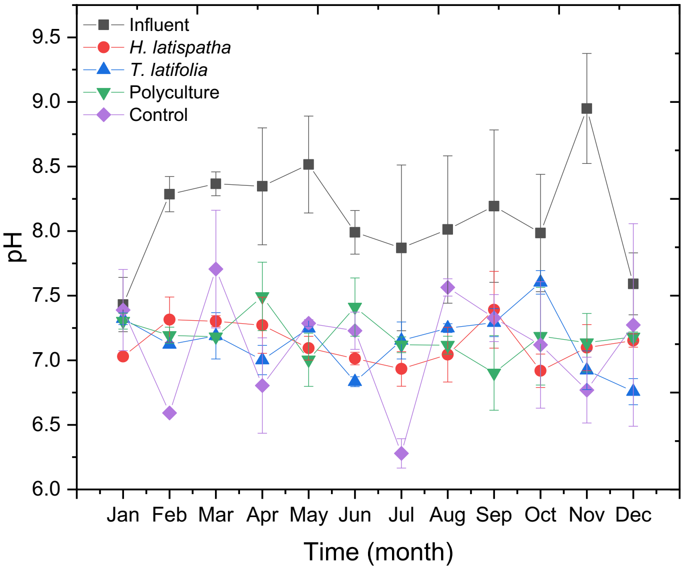


Figure 3. pH behavior throughout the experiment

In the case of temperature, no significant differences were observed between the different treatments used (Figure 4). *H. latispatha* and *T. latifolia* showed homogeneous behavior in thermal regulation, with temperatures within a similar range.

The observed temperature range (20–30 °C) is optimal for microbial activity and processes such as nitrification and denitrification. Amiri *et al*. (2022) observed that CWs planted with typical wetland species, such as *Phragmites australis* and *Typha latifolia*, showed a greater capacity to regulate temperature. On the other hand, studies such as that by Stefanatou *et al*. (2023) have shown that the use of ornamental plants in CWs not only improves aesthetics but also contributes to the regulation of parameters such as temperature and dissolved oxygen.

Regarding DO, when comparing the results of the study period, all systems with vegetation showed significant differences with respect to the influent and the system without vegetation, as well as among themselves (Figure 5). The planted systems showed significant differences compared to the influent and the control. The influent had an average DO of 0.7 ± 0.09 mg/L, with values between 0.50 and 0.85 mg/L, indicating initial conditions of low oxygenation, typical of slaughterhouse wastewater due to its high organic load. In contrast, the systems with vegetation showed a significant increase in DO values, ranging between 1.51 and 2.08 mg/L. On the other hand, systems without vegetation (control) presented lower values (0.52 to 1.15 mg/L), confirming that vegetation plays a key role in improving DO through mechanisms such as oxygen transfer through plant tissues and the generation of aerobic microzones in the rhizome. When comparing systems planted with *Typha latifolia* (a typical wetland species) and ornamental species such as *H. latispatha*, as well as a combination of both in polyculture, the results showed significant differences in oxygenation capacity. This behavior may be related to differences in the ability of plants to transfer oxygen through their rhizosphere. Previous studies have shown that ornamental species such as *Zantedeschia aethiopica* and *Canna indica* can also play an important role in oxygen transfer, although their efficiency may be lower compared to typical plants such as *T. latifolia* and *Cyperus papyrus* (Singh & Chakraborty, 2021).

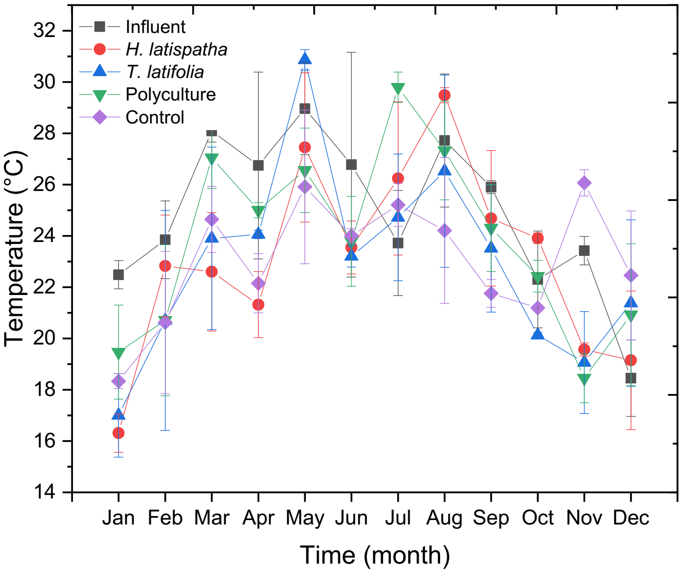


Figure 4. Temperature behavior throughout the experiment

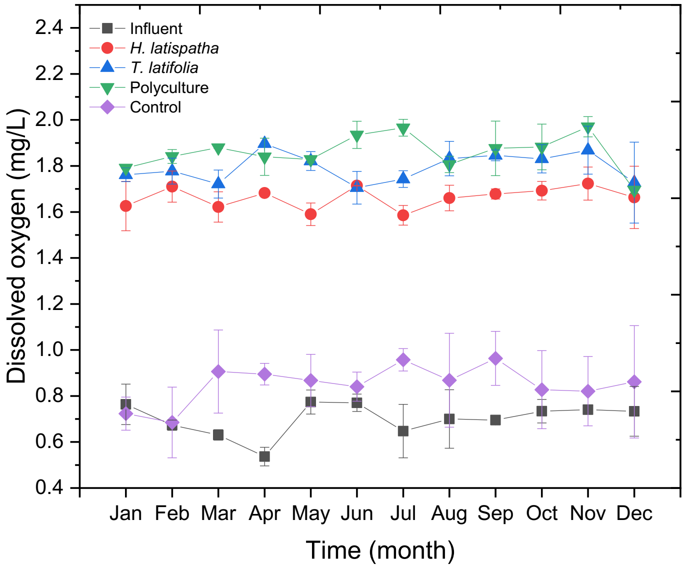


Figure 5. DO behavior throughout the experiment

Removal efficiencies of TN, NH_4^+, NO_3^-, and NO_2^-

Figures 6-9 show the inlet and outlet concentrations, as well as the removal efficiencies for the nitrogen compounds analyzed in this study (TN, NH_4^+, NO_2^-, and NO_3^--N).

For TN, the polyculture of *H. latispatha* and *T. latifolia* showed the highest removal efficiency, with average values of 64–65% (Figure 6). Effluent concentrations were also the lowest, reaching 88 ± 2.342 mg/L. This superior performance can be attributed to the synergy between the two species, which combine their different capacities for absorption, tolerance, and promotion of microbiological processes in the rhizosphere.

Individual systems showed intermediate efficiencies, with *T. latifolia* outperforming *H. latispatha* throughout the monitoring period (57% vs. 48%). This result may be due to the fact that *T. latifolia* is a typical wetland plant with a greater capacity to promote nitrification and denitrification, processes that are essential for TN removal.

The control system, without plants, showed the lowest removal efficiencies (33%) and the highest effluent concentrations (166 ± 6.212 mg/L). This highlights the crucial role of plants in TN removal, as they not only act as passive filters, but also facilitate key biological processes through oxygenation of the root zone.

No significant differences were observed whitin each treatment across the study period, suggesting stability in the performance of the systems studied. However, between different treatments, all comparisons showed significant differences, highlighting the differential effectiveness of each system. Polyculture was significantly more efficient than individual systems with *H. latispatha* or *T. latifolia*, which in turn were more effective than the control system. This pattern suggests that species diversification in constructed wetlands may be an optimal strategy for maximizing TN removal in wastewater treatment systems.

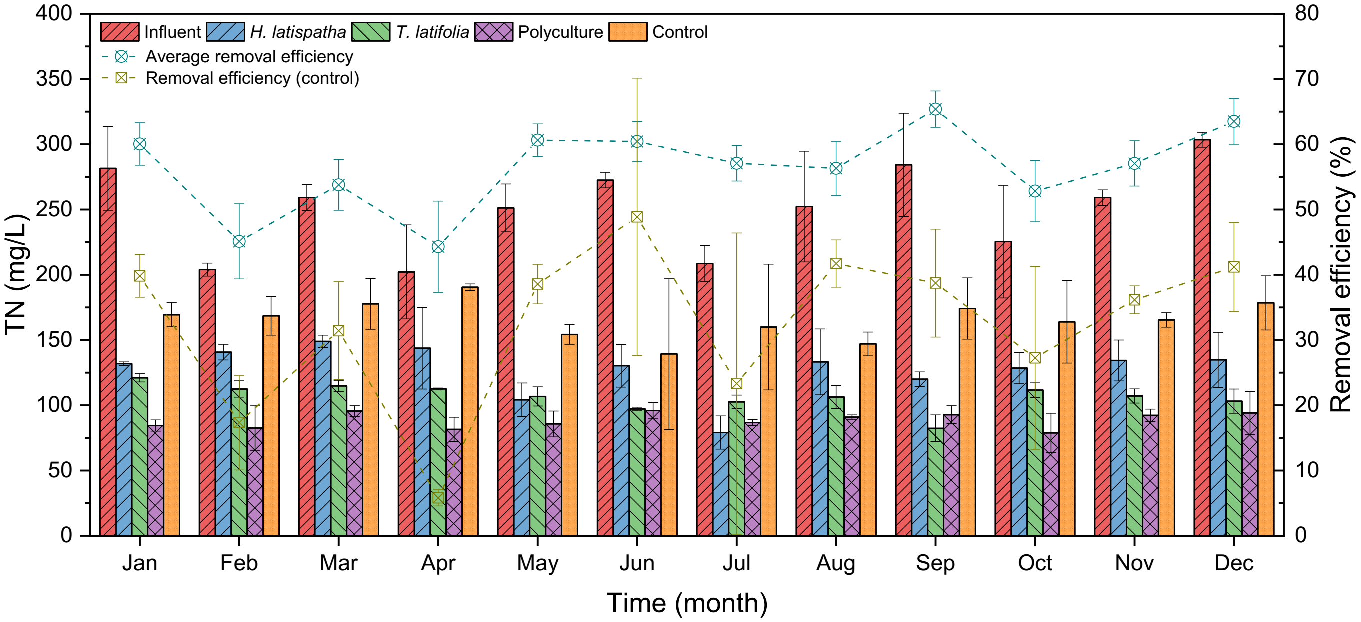


Figure 6. Concentrations at inlet, outlet, and removal efficiencies of TN in CWs

The results of this study highlight the effectiveness of CWs systems in eliminating TN, with superior performance in polyculture. The ability of *T. latifolia* to promote denitrification and its positive interaction with *H. latispatha* appear to be key factors in this effectiveness. In contrast, systems without plants (control) were significantly less efficient, highlighting the importance of plants as active components in constructed wetlands. These findings reinforce the importance of designing systems that optimize the combination of plant species to maximize the removal of pollutants in wastewater with high nitrogen content, such as that from slaughterhouses. The stability observed between years suggests that these systems can offer sustainable and reliable solutions for wastewater treatment.

In the current study, *T. latifolia* consistently showed higher TN removal efficiencies than *H. latispatha* during the evaluation period. *T. latifolia* achieved an efficiency of 57%, compared to 48% for *H. latispatha*, with effluent concentrations of 107 ± 2.374 mg/L and 127 ± 5.085 mg/L, respectively. These differences can be attributed to the ability of typical wetland plants such as *T. latifolia* to promote nitrification and denitrification processes in the rhizosphere, due to greater oxygenation and associated microbial activity. In contrast, ornamental plants, although contributing to TN removal, appear to be less efficient in promoting these key processes.

In reference studies, wetland plants such as *Typha latifolia*, *Cyperus papyrus*, and *Canna indica* have demonstrated greater TN removal capacity compared to systems using ornamental plants. For example, Michael *et al*. (2020), in a horizontal subsurface flow system with *Cyperus papyrus*, reported efficiencies of 97.6% for TN in slaughterhouse effluents, while Mohammed & Ismail (2021) achieved an efficiency of 90.2% using *Canna indica* in dairy wastewater. These values far exceed those achieved by *H. latispatha* in the present study.

In systems that use ornamental plants, such as those reported by Almeida-Naranjo *et al*. (2020) with *Heliconia stricta* in hybrid wetlands, TN removal was significantly lower, with values not exceeding 56%. This performance is comparable to that observed in the present study for *H. latispatha* (46–48%).

The difference in TN removal efficiency may be related to the functional characteristics of the plants. Wetland species, such as *T. latifolia*, are adapted to high humidity conditions and have extensive root systems that promote substrate oxygenation and stimulate bacterial activity, facilitating nitrification and denitrification processes. On the other hand, ornamental plants such as *H. latispatha*, although aesthetically attractive and useful in certain applications, have a lower capacity to contribute to these biological processes.

The results of this study, together with data reported in the literature, highlight the superiority of wetland plants over ornamental plants in TN removal. While *H. latispatha* may be useful in specific applications, wetland plants such as *T. latifolia* represent a more efficient option for wastewater treatment systems that require high nitrogen removal rates. These findings underscore the importance of selecting appropriate species based on treatment objectives and wastewater characteristics.

When comparing the results of this study with other technologies that do not use constructed wetlands, it is noteworthy that Tabelini *et al*. (2023) reported TN removal efficiencies of 75–80% when using activated sludge systems and anaerobic lagoons to treat wastewater from the dairy industry. Although these technologies exceed the efficiencies obtained in the present study, it is important to consider that wastewater from slaughterhouses tends to have higher concentrations of nitrogen and organic compounds, which could explain the lower efficiency of constructed wetland-based systems. In addition, activated sludge systems require more infrastructure and energy consumption, while constructed wetlands represent low-cost technologies.

Akarsu *et al*. (2021) achieved 78% efficiency in removing TN from slaughterhouse wastewater using electrocoagulation and electro flotation, resulting in significantly lower effluent concentrations. However, these technologies have limitations related to high operating costs and advanced technical requirements, which can make them unfeasible in rural or resource-constrained regions, where constructed wetlands represent a more sustainable alternative.

Studies such as those by Mkilima *et al*. (2022) and Meiramkulova *et al*. (2020) reported TN removal efficiencies of 98–99% in poultry slaughterhouse wastewater using nanofiltration membranes. Although these technologies produce high-quality effluents, their application in slaughterhouse wastewater faces challenges related to the pretreatment required to prevent membrane clogging, as well as the management of the concentrates generated. In contrast, constructed wetlands offer a simpler and more economical option, albeit with lower removal efficiencies.

Lopes *et al*. (2022) reported a 65% efficiency in TN removal in wastewater from poultry slaughterhouses using combined anaerobic–anoxic–aerobic reactors, an efficiency comparable to that obtained in the polyculture system in this study. However, combined reactors require greater monitoring and technical control, while constructed wetlands are more robust to variations in wastewater quality.

Although advanced technologies offer greater efficiencies in TN removal, constructed wetlands represent a more sustainable, economical, and adaptable alternative for treating slaughterhouse wastewater. The results of this study highlight the importance of polycultures as a strategy for optimizing TN removal, positioning constructed wetlands as a viable option for resource-constrained environments.

With regard to NH_4^+, NO_3^-, and NO_2^-, the results highlight significant differences in performance between the treatments and the control, underscoring the importance of plant species and their combinations in the removal of nitrogen compounds from slaughterhouse wastewater.

According to Figure 7, in the case of NH_4^+, polyculture systems showed the highest removal efficiency (89%), achieving the lowest effluent concentrations (18 mg/L). Systems with *T. latifolia* also demonstrated a high capacity for NH_4^+ removal (77%), significantly outperforming systems with *H. latispatha*, which achieved efficiencies of 65%.

The removal of NO_3^--N (Figure 8) followed a similar trend to that observed for NH_4^+, with polyculture being the most effective treatment, with efficiencies of 91% and effluent concentrations of 2 mg/L. Systems with *T. latifolia* (78%) outperformed systems with *H. latispatha* (68%), highlighting the superiority of typical wetland plants in nitrate treatment.

As shown in Figure 9, in the case of NO_2^-, polycultures also showed the highest removal capacity (92%), with effluent concentrations of 0.57 mg/L.

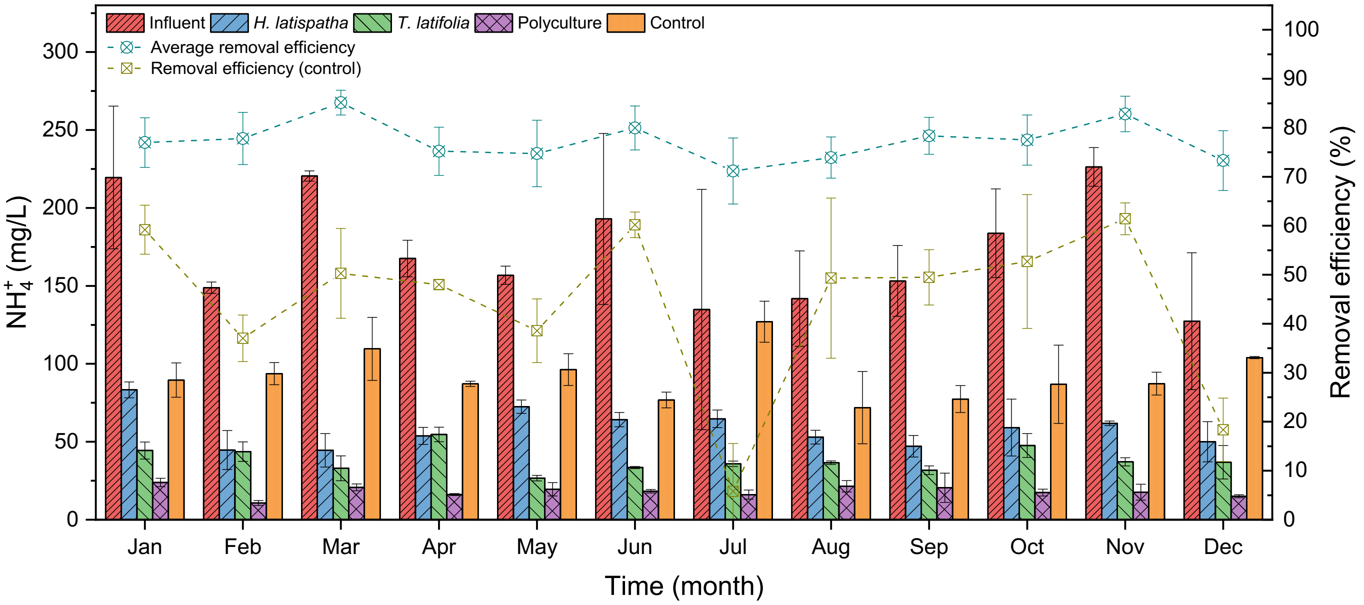


Figure 7. Inlet and outlet concentrations and removal efficiencies of NH_4^+ in CWs.

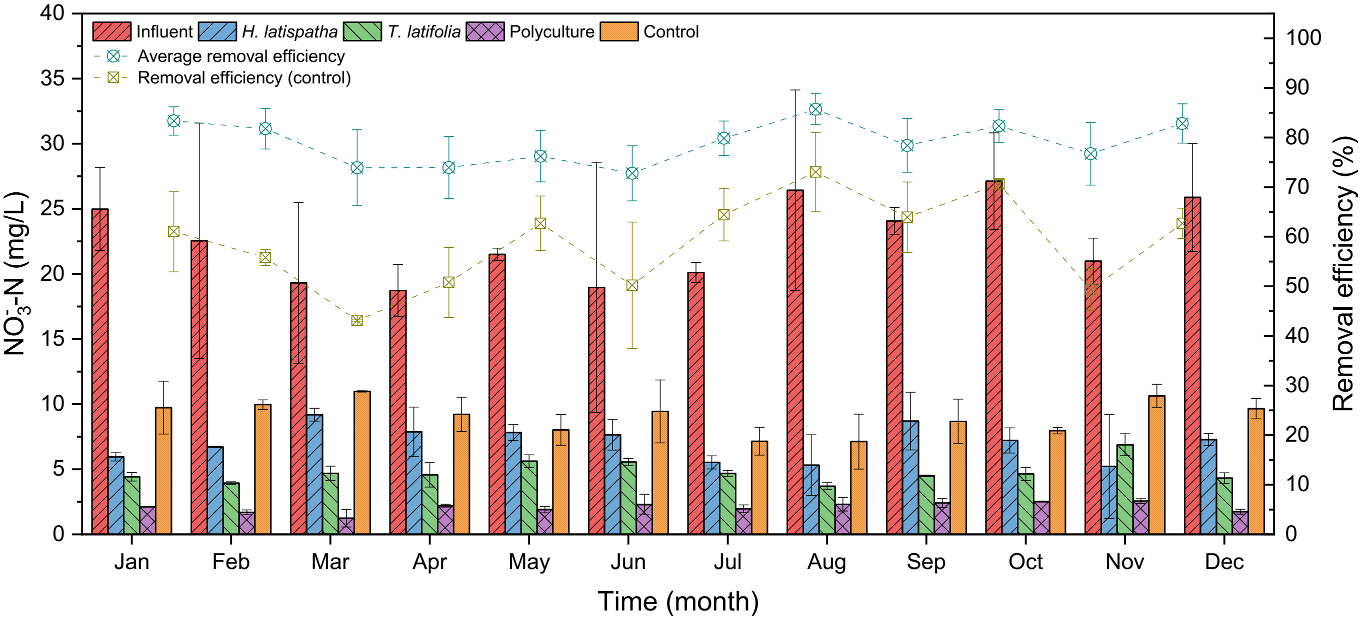


Figure 8. Inlet and outlet concentrations and removal efficiencies of NO_3^--N in CWs.

Systems with *T. latifolia* showed high removal efficiencies (82%), with effluent concentrations of 1.4 mg/L. Systems with *H. latispatha*, on the other hand, showed lower efficiencies (73%). No significant differences were observed across the study period for systems with the same treatment, indicating stability in the efficiency of constructed wetlands over time. However, significant differences were found between the different treatments in all cases, highlighting that polycultures were consistently superior in the removal of NH_4^+, NO_3^-−N, and NO_2^-−N.

The above results show that polycultures of *H. latispatha* + *T. latifolia* maximize the removal of nitrogen compounds in slaughterhouse wastewater, which could be attributed to the combination of plant absorption mechanisms and microbial activity facilitated by the roots. The superior capacity of *T. latifolia* compared to *H. latispatha* could be explained by its greater tolerance and adaptation to flooded and nutrient-rich environments.

When comparing the values in this study with those reported in other research on ornamental plants, it can be seen that the removal efficiencies of *H. latispatha* are lower than those of other ornamental species evaluated in effluent treatment systems. For example, in the study by Madeira *et al*. (2022) in Portugal, an NH_4^+ removal efficiency of 72% was reported, which is higher than that obtained in this study. Additionally, Michael *et al*. (2020) reported an NH_4^+ removal efficiency of 97.4% in a system that used *Cyperus papyrus*, demonstrating significantly higher efficiency in wetland plants compared to ornamental plants.

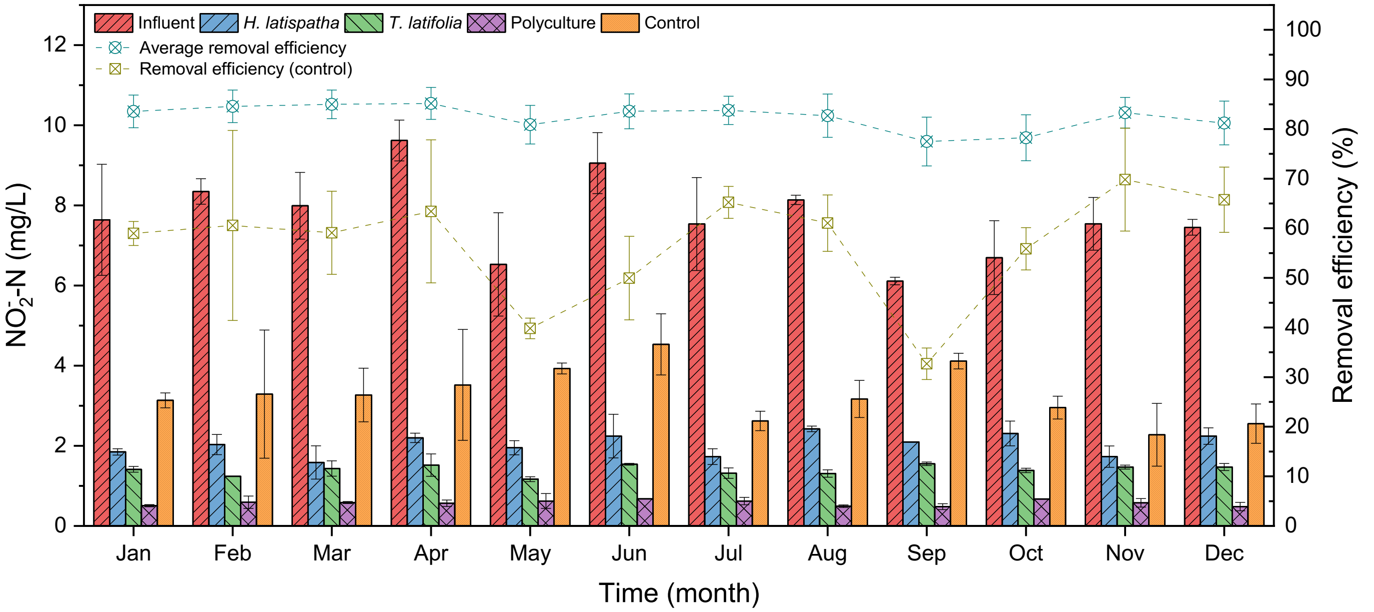


Figure 9. Concentrations at inlet, outlet, and removal efficiencies of NO_2^--N in CWs.

Regarding wetland plants, the removal efficiencies of *T. latifolia* in this study are close to the values reported by Galindo Montero *et al*. (2024) in Colombia, who documented NH_4^+ removal efficiencies of 89% in a horizontal subsurface wetland system with *Typha domingensis*. This suggests that wetland plants, such as *T. latifolia*, are more effective than ornamental plants in treating effluents. However, other studies, such as those by Suwerda *et al*. (2022), reported NH_4^+ removal efficiencies of 97.4%, indicating that some wetland species may be even more effective at removing these compounds.

Badejo *et al*. (2020) in Nigeria and Michael *et al*. (2020) in Tanzania also reported NO_3^--N removal efficiencies higher than those observed in this study. In particular, Michael *et al*. (2020) documented a NO_3^--N removal efficiency of 97.6%, higher than the values obtained for T. latifolia in this study (78%). This suggests that certain wetland species, such as *Cyperus papyrus*, perform better at removing NO_3^--N than *T. latifolia*.

The results obtained were compared with those reported in the literature for technologies that do not use constructed wetlands, with the aim of highlighting the advantages and limitations of wetlands in the treatment of this type of effluent. When comparing these results with those reported for technologies that do not use constructed wetlands, significant differences in pollutant removal capacity can be observed, although the efficiency of slaughterhouse wastewater treatment tends to be lower in systems without wetlands.

For example, Tabelini *et al*. (2023) in Brazil, using activated sludge systems and anaerobic lagoons, reported removal efficiencies of 70% for NH_4^+ and 64% for NO_3^-−N, values that are lower than those obtained in this study for *T. latifolia* (81% for NH_4^+ and 77% for NO_3^-−N). This indicates that the use of constructed wetlands may be more efficient than traditional biological systems, such as activated sludge systems, in the treatment of NH_4^+ and NO_3^-−N in slaughterhouse wastewater.

In addition, Michael *et al*. (2020) in Tanzania used a biodigester combined with a constructed wetland, reporting removal efficiencies of 97.4% for NH_4^+, 97.6% for NO_3^-−N, and 99.1% for NO_2^-−N. Although this combined system is highly efficient, the reported values are considerably higher than those obtained in this study. However, it is important to consider that the system evaluated by Michael *et al*. (2020) includes a biodigestion stage, which improves the removal of NH_4^+ and other pollutants, which may explain the differences in the reported efficiencies. In the case of slaughterhouse wastewater, a system based solely on constructed wetlands, such as the one evaluated in this study, has a considerably lower efficiency but still offers advantages compared to technologies that do not incorporate this component.

On the other hand, in the study by Madeira *et al*. (2022) in Portugal, which used phytoremediation through vertical flow wetlands, an NH_4^+ removal efficiency of 72% was reported. This value is higher than that obtained in this study for *H. latispatha* (67%) and closer to that of *T. latifolia* (81%), suggesting that the type of wetland and the species used play a significant role in NH_4^+ removal efficiency. Furthermore, as this is a vertical flow wetland system, differences in performance may occur depending on the configuration and operating conditions of the systems.

Another key aspect to consider is the type of wastewater being treated. In this study, slaughterhouse wastewater is particularly complex, as it contains high concentrations of organic matter, suspended solids, and nutrients such as NH_4^+, which makes its treatment more challenging. Wetland plants such as *T. latifolia* are more effective in this type of water due to their ability to tolerate aquatic environments with high organic loads and their specialized nutrient absorption mechanisms. In contrast, technologies such as electrocoagulation and electro flotation, used by Akarsu *et al*. (2021) in Turkey, are effective for removing suspended solids and certain pollutants, but do not have the same capacity to efficiently remove nutrients such as NH_4^+ and NO_3^-−N, especially in slaughterhouse effluents. Electrocoagulation, for example, can remove up to 94% of COD; however, its performance in nutrient removal is limited, demonstrating a clear advantage of constructed wetland-based systems.

The results obtained in this study for the removal of NH_4^+, NO_3^--N, and NO_2^- using constructed wetlands with *H. latispatha* and *T. latifolia* are competitive with those reported for other technologies, especially in the treatment of slaughterhouse wastewater. Although activated sludge systems or combined biodigesters can achieve higher efficiencies in certain circumstances, constructed wetlands, as observed in this study, remain a viable and efficient option for the treatment of slaughterhouse effluents, with the added advantage of being natural and sustainable systems. Furthermore, the use of ornamental plants in combination with wetland plants in polyculture shows optimized performance in contaminant removal, which could be a key strategy for improving the efficiency of slaughterhouse wastewater treatment systems.

Conclusions

This study provides robust evidence on the effectiveness of constructed wetlands (CWs) as a sustainable and economical solution for treating slaughterhouse wastewater, with a special focus on the removal of nitrogenous pollutants such as TN, NH_4^+, NO_3^-, and NO_2^-. Through a rigorous experimental design, it was demonstrated that CWs, particularly those planted with *Typha latifolia* and *Heliconia latispatha*, are capable of achieving competitive removal efficiencies comparable to those of more expensive conventional technologies, such as activated sludge and electrocoagulation systems.

The research revealed that polyculture of *T. latifolia* and *H. latispatha* offered the highest efficiency in removing TN (64–65%) and NH_4^+ (89%), highlighting the synergy between the two species, which not only favored nutrient absorption but also supported key microbiological processes in the rhizosphere. Furthermore, comparative analysis with other studies and technologies showed that, although CWs do not achieve the removal levels of other advanced treatment technologies, such as nanofiltration membranes or combined biodigesters, their low cost, operational simplicity, and sustainability make CWsa viable option, especially in regions with limited resources.

The superior performance of *T. latifolia* in nitrogen removal, together with the good results obtained with *H. latispatha* in combination, underscores the importance of selecting plant species based on effluent characteristics and treatment objectives. These findings suggest that constructed wetlands can be an effective solution for the bioremediation of slaughterhouse wastewater, helping to mitigate the environmental impact of these effluents in an ecological and economical manner.

Finally, the results also highlight the importance of considering local climatic and environmental conditions in the design of CW systems, as factors such as temperature, solar radiation, and relative humidity significantly influence the growth and performance of plant species. This study provides a useful foundation for future research that can further optimize CWsystems, expand their applicability to other industries and regions, and promote the use of nature-based solutions for wastewater treatment globally.

Hybrid constructed wetlands (horizontal and vertical flow) are recommended for treating slaughterhouse wastewater due to their greater capacity to promote nitrification and denitrification processes. It is also suggested to combine CWs with complementary technologies, such as biodigesters or anaerobic bioreactors, especially when dealing with wastewater with high organic load and high nitrogen concentrations, such as effluents from slaughterhouses. This combination can improve the overall efficiency of the system by addressing different types of pollutants and offering solutions for situations where CWs alone are not sufficient to comply with environmental regulations and standards.

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