PHYTOREMEDIATION OF MUNICIPAL WASTEWATER USING SUBMERGED MACROPHYTE *Vallisneria Spiralis* Linn

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ABSTRACT

This study investigates the effectiveness of phytoremediation, a nature-based wastewater treatment method, using the submerged macrophyte Vallisneria spiralis to treat municipal wastewater. Conventional treatment methods are often expensive and energy-intensive, making sustainable alternatives crucial.

We evaluated the performance of V. spiralis by setting up five different treatment concentrations of municipal wastewater (20%, 40%, 60%, 80%, and 100%) in controlled laboratory conditions over a 15-day period. Physicochemical parameters such as Biochemical Oxygen Demand (BOD), Chemical Oxygen Demand (COD), Total Suspended Solids (TSS), Total Dissolved Solids (TDS), and key nutrients like nitrates (NO_3), phosphates (PO_4), and ammonia nitrogen (NH_3-N) were analyzed both before and after treatment.

The results showed significant reductions across all measured parameters, with removal efficiencies increasing as the initial wastewater concentration rose. The plant was particularly effective at removing organic pollutants, with BOD and COD levels in the 100% concentration set dropping from 220 mg/L to 32 mg/L and 340 mg/L to 54 mg/L, respectively. Nutrient removal was also highly successful; ammonia nitrogen was almost completely removed in most sets. The treatment also led to a substantial reduction in TSS and TDS, improving overall water clarity. The findings demonstrate that V. spiralis is a promising, efficient, and cost-effective solution for municipal wastewater treatment.

KEYWORDS: *Phytoremediation, submerged macrophytes, Municipal Wastewater, Vallisneria spiralis Linn.*

INTRODUCTION

Rapid urbanization and industrialization have led to an exponential increase in municipal wastewater generation, posing significant threats to water quality, aquatic ecosystems, and public health (Kumar et al., 2020). Conventional wastewater treatment methods, such as activated sludge and chemical treatments, are often costly, energy-intensive, and produce secondary pollution (Sharma & Bhattacharya, 2017). In this context, nature-based solutions such as phytoremediation have emerged as sustainable and cost-effective alternatives for wastewater treatment (Ali et al., 2013).

Phytoremediation is a process that utilizes plants to remove, degrade, or stabilize contaminants from water, soil, and sediments through biological, chemical, and physical mechanisms (Mishra et al., 2008). Aquatic macrophytes, in particular, play a pivotal role in wastewater treatment by enhancing nutrient uptake, promoting microbial activity in the rhizosphere, and improving water quality through sediment stabilization (Brix, 1997). Submerged macrophytes such as *Vallisneria spiralis* are particularly effective because of their high surface area, efficient nutrient absorption, and ability to oxygenate the water column (Chaudhary et al., 2018).

The *Vallisneria spiralis*, a perennial submerged plant, is widely distributed in freshwater ecosystems and has been reported to remove excess nutrients such as nitrogen and phosphorus, as well as heavy metals and organic pollutants, from wastewater (Sood et al., 2012). Its rapid growth, adaptability to varying environmental conditions, and low maintenance requirements make it a promising candidate for municipal wastewater treatment systems (Jena et al., 2021). Furthermore, the plant's role in enhancing dissolved oxygen levels facilitates microbial degradation of organic matter, improving overall treatment efficiency (Zhao et al., 2014).

Given the growing need for environmentally friendly and economically viable wastewater management solutions, evaluating the potential of *Vallisneria spiralis* for municipal wastewater phytoremediation could contribute significantly to sustainable urban water management strategies. This study aims to assess the efficiency of *Vallisneria spiralis* in removing nutrients and improving water quality parameters in municipal wastewater under controlled conditions.

Municipal wastewater originates mainly from domestic, commercial, and institutional sources and typically contains a complex mixture of organic matter, nutrients such as nitrogen and phosphorus, pathogens, heavy metals, and various emerging contaminants (Kumar et al., 2020). The growing pressures from urbanization and industrial expansion have significantly increased the volume of wastewater generated, thereby straining existing treatment infrastructures (Zhou et al., 2018). Traditional treatment methods like activated sludge, trickling filters, and chemical coagulation are often costly, require high energy inputs, and produce large amounts of sludge that necessitate further disposal (Sharma & Bhattacharya, 2017). Consequently, there is a global push toward sustainable, cost-effective, and environmentally sound alternatives, among which phytoremediation has emerged as a promising approach (Ali et al., 2013).

Phytoremediation is an environmentally friendly technique that employs plants and their associated microorganisms to remove, degrade, or immobilize pollutants from contaminated environments (Salt et al., 1998). In aquatic systems, certain plants known as macrophytes play a crucial role in pollutant removal through mechanisms such as direct uptake, adsorption to root surfaces, microbial activity in the rhizosphere, and stabilization of sediments (Brix, 1997). This form of treatment offers several advantages over conventional methods, including lower operational and energy costs, minimal secondary pollution, and the capability to treat water directly at the contamination site (Vymazal, 2010).

Among aquatic macrophytes, different groups such as emergent, floating, and submerged plants contribute to pollutant removal in unique ways (Sood et al., 2012). Submerged macrophytes are particularly effective due to their continuous contact with the water column, large surface area that supports microbial growth, and their ability to release oxygen into the surrounding water through photosynthesis (Chaudhary et al., 2018). These plants not only facilitate nutrient removal but also help reduce turbidity, enhance sedimentation, and improve the aesthetic and ecological quality of treated water (Wu et al., 2014).

The *Vallisneria spiralis*, also known as tape grass or eelgrass, is a submerged, perennial aquatic plant commonly found in freshwater environments across tropical and subtropical regions (Liu et al., 2016). Its morphology—characterized by long, ribbon-like leaves and an extensive root system—combined with its tolerance to diverse environmental conditions, makes it a strong candidate for wastewater treatment (Jena et al., 2021). Ecologically, *V. spiralis* contributes to nutrient cycling, sediment stabilization, and provides habitat for aquatic fauna (Sood et al., 2012). Its ability to release oxygen into the rhizosphere further supports microbial activity, which enhances the breakdown of organic pollutants and improves overall water quality (Zhao et al., 2014).

Numerous studies have confirmed the phytoremediation potential of *Vallisneria spiralis* in various wastewater treatment scenarios. For instance, it has demonstrated the ability to effectively remove nutrients such as ammonium, nitrate, and phosphate, as well as organic pollutants like chemical oxygen demand (COD) from municipal wastewater (Sood et al., 2012). Additionally, research by Mishra et al. (2008) highlights the species' high capacity for mercury uptake, suggesting its usefulness in heavy metal remediation. Chaudhary et al. (2018) further observed the plant's role in significantly reducing nitrogen and phosphorus levels, thus mitigating eutrophication. In constructed wetlands, *V. spiralis* has been shown to increase dissolved oxygen levels, promote microbial degradation of contaminants, and reduce suspended solids in the treated water (Zhao et al., 2014). Jena et al. (2021) reported removal efficiencies exceeding 70% for total nitrogen and phosphorus in municipal wastewater treatment setups utilizing this plant.

However, the effectiveness of *V. spiralis* in phytoremediation is influenced by a variety of environmental and operational factors. These include water depth, light availability, temperature, hydraulic retention time, and the concentrations of pollutants present (Vymazal, 2010). While high nutrient levels can initially enhance uptake, excessive concentrations may lead to physiological stress in the plant (Liu et al., 2016). Seasonal variations also play

a role, with greater remediation performance generally observed during warmer months due to increased metabolic and photosynthetic activity (Chaudhary et al., 2018).

Despite promising results in laboratory and small-scale field trials, the application of *Vallisneria spiralis* in large-scale municipal wastewater treatment remains limited. Further research is needed to refine treatment system designs, assess the long-term accumulation of pollutants in plant tissues, and develop effective strategies for biomass management following harvest (Ali et al., 2013). Additionally, integrating *V. spiralis* with other macrophyte species in hybrid systems could improve treatment efficacy and resilience under variable environmental conditions, paving the way for more robust and sustainable wastewater treatment solutions.

RESEARCH METHODOLOGY
A. Test Plant
The test plant used in this study was *Vallisneria spiralis* L., a submerged perennial aquatic macrophyte belonging to the family Hydrocharitaceae. Commonly known as eelgrass, tape grass, or coiled vallisneria, it is a rhizomatous species that produces rosettes of long, strap-like leaves ranging from a few centimeters to over 5 meters in length, depending on water depth and environmental conditions (Global Invasive Species Database \[GISD], 2025). The leaves arise from a basal rosette anchored in the sediment via stout rhizomes, with numerous roots—up to 40 cm in length—emerging from each node (Greater Wellington Regional Council, 2004).

The *Vallisneria spiralis* thrives in still or slow-flowing freshwater habitats, including lakes, rivers, and wetlands (GISD, 2025). The species is dioecious, producing separate male and female plants. Male flowers are released into the water column, while female flowers are borne on a long, spiral peduncle that unfurls to reach the surface (Warrington, 1994). The plant reproduces primarily through vegetative propagation, with new shoots arising from rhizome extensions.

The species is known for its ecological role in aquatic systems, particularly in nutrient cycling and habitat stabilization. Its extensive root and rhizome network binds sediments, reducing turbidity and facilitating colonization by benthic microorganisms (Qian et al., 1999, as cited in Vajpayee et al., 2001). These properties make it an effective phytoremediation agent capable of removing excess nutrients, suspended solids, and certain heavy metals from contaminated waters (Sood et al., 2012). In one study, *V. spiralis* removed up to 95% of chromium from diluted tannery effluent, primarily via surface adsorption and bioaccumulation (Vajpayee et al., 2001).

In the present experiment, healthy *V. spiralis* plants were collected from a shallow freshwater body with minimal pollution. Plants were selected for uniform size and vigor, ensuring intact root systems to promote acclimatization and growth during the experiment. They were rinsed gently to remove debris and epiphytes, then acclimated in dechlorinated tap water for seven days under controlled laboratory conditions before being introduced into the treatment systems. This acclimatization period minimized transplant shock and allowed the plants to recover metabolic stability prior to exposure to municipal wastewater (Mishra & Tripathi, 2008).

B. Treatment Study
This study was conducted to evaluate the phytoremediation potential of *Vallisneria spiralis* for the treatment of municipal wastewater under controlled laboratory conditions. Municipal wastewater samples were collected from a local sewage inlet point following standard sampling procedures outlined by the American Public Health Association (APHA, 2017). The collected wastewater was stored in clean, high-density polyethylene containers and transported to the laboratory for further characterization.

Prior to the phytoremediation experiment, the wastewater was analyzed for key physicochemical parameters, including pH, electrical conductivity (EC), total suspended solids (TSS), total dissolved solids (TDS), total solids (TS), biochemical oxygen demand (BOD), chemical oxygen demand (COD), nitrates (NO_3^-), ammonia nitrogen (NH_3-N), total nitrogen, phosphates (PO_4^{3-}), sulphates (SO_4^{2-}), and chlorides (Cl^-), using standard methods (APHA, 2017; Trivedy & Goel, 1986). These parameters were selected because they are critical indicators of wastewater pollution and directly influence the growth and remediation efficiency of aquatic macrophytes (Sood et al., 2012).

For the phytoremediation trials, *Vallisneria spiralis* specimens were collected from a shallow, unpolluted freshwater body, ensuring that only healthy plants with intact roots and leaves were selected. Plants were acclimatized for seven days in tap water to minimize transplantation shock and to remove any adhered sediments or epiphytes (Jena et al., 2021).

Five experimental treatments were prepared by diluting the municipal wastewater with distilled water to obtain different concentrations: 20%, 40%, 60%, 80%, and 100% (undiluted). Each treatment was set up in triplicate in rectangular glass aquaria of equal volume (15 L), containing a uniform biomass of *Vallisneria spiralis* (fresh weight basis). A control set with distilled water and *Vallisneria spiralis* was also maintained to assess any changes in water quality parameters in the absence of wastewater contaminants (Chaudhary et al., 2018).

The aquaria were placed in a well-lit laboratory environment with natural photoperiod conditions, and no artificial aeration or nutrient supplementation was provided during the experiment. The water temperature and pH were monitored daily to ensure stability throughout the study period. The exposure duration was maintained for 15 days, during which plants were allowed to interact with the wastewater, facilitating the removal of pollutants via uptake, transformation, and sedimentation processes (Mishra et al., 2008).

At the end of the treatment period, the effluents from each set were collected and analyzed for all previously mentioned physicochemical parameters using the same methods as for initial characterization. The percentage removal efficiency for each parameter was calculated using the formula:

$$\text{Removal Efficiency (\%)} = [(C_i - C_f)/ C_i] \times 100$$

Where, C_i is the initial concentration (e.g., in mg/L) and C_f is the final concentration (e.g., in mg/L) after treatment (Ali et al., 2013).

RESULTS AND DISCUSSION

A. Pre-Treatment Status

The initial physicochemical characterization of the municipal wastewater and its diluted treatment sets (20%, 40%, 60%, 80%, and 100% concentration) indicated that most parameters increased proportionally with the concentration of sewage. The pH values ranged from 7.01 in the 20% dilution (Set I) to 7.21 in the undiluted sewage (Set V), indicating near-neutral conditions across all treatments (Table 1). This pH range is typical of municipal wastewater, which often falls between 6.5 and 8.0 due to domestic and organic inputs (Metcalf & Eddy, 2014). Electrical conductivity (EC) showed a gradual increase from 12 µS/cm in Set I to 23 µS/cm in Set V, reflecting the rising ionic content with increasing sewage concentration (APHA, 2017).

Suspended and dissolved solids exhibited a clear trend of increase with sewage concentration. Total suspended solids (TSS) ranged from 18 mg/L in Set I to 94 mg/L in Set V, while total dissolved solids (TDS) varied from 27 mg/L to 136 mg/L. Consequently, total solids (TS) increased from 45 mg/L in the most diluted set to 230 mg/L in undiluted sewage. High levels of TSS and TDS in raw sewage are often associated with domestic effluents containing food waste, detergents, and silt (Qadir et al., 2010).

Biochemical oxygen demand (BOD) and chemical oxygen demand (COD) also increased substantially with sewage concentration, with BOD ranging from 51 mg/L (Set I) to 220 mg/L (Set V), and COD ranging from 73 mg/L to 340 mg/L. These values are consistent with typical domestic sewage characteristics, where high organic loading contributes to elevated oxygen demand (Tchobanoglous et al., 2014). The observed BOD/COD ratios (approximately 0.7–0.65 across treatments) indicate a high proportion of biodegradable organic matter, which is favorable for biological treatment processes such as phytoremediation (Zhou et al., 2017).

Nutrient concentrations also followed an increasing trend with sewage concentration. Nitrate (NO_3^-) levels increased from 8 mg/L in Set I to 58 mg/L in Set V, while phosphate (PO_4^{3-}) rose from 3.8 mg/L to 20 mg/L. Similarly, sulfate (SO_4^{2-}) ranged from 6 to 33 mg/L, and chloride (Cl^-) from 83 to 418 mg/L. Elevated nutrient concentrations are a common feature of untreated municipal wastewater and contribute to eutrophication in receiving waters if discharged without treatment (Smith & Schindler, 2009). Ammonia nitrogen varied from 7 mg/L

to 32 mg/L, and total nitrogen from 35 mg/L to 182 mg/L, values that align with typical municipal wastewater profiles (Metcalf & Eddy, 2014).

Table 1 summarizes the initial physicochemical characteristics of municipal sewage influent and the corresponding treatment sets prepared by diluting the sewage with distilled water at concentrations ranging from 20% to 100%. The data reveal a gradual increase in contaminant levels with rising wastewater concentration across all measured parameters. For example, pH values remain near neutral, ranging slightly from 7.01 to 7.21, while indicators of pollution such as Electrical Conductivity (EC), Total Suspended Solids (TSS), Total Dissolved Solids (TDS), and Total Solids (TS) show a clear upward trend with increasing concentration. Similarly, organic load parameters like Biological Oxygen Demand (BOD) and Chemical Oxygen Demand (COD) increase significantly from the lowest to highest concentration sets, indicating higher pollutant loads. Nutrients such as nitrates (NO_3), phosphates (PO_4), sulfates (SO_4), chlorides (Cl^-), ammonia nitrogen, and total nitrogen also rise proportionally, reflecting the expected escalation in nutrient and ion content with higher sewage percentages. This baseline characterization is crucial for understanding the initial pollutant load and assessing the treatment efficiency in subsequent experimental sets.

Table 1
Initial Physicochemical characteristics of Municipal Sewage influent and Treatment sets prepared with distilled water for dilutions.

Treatment Parameter	Unit	Set I (Conc-20%)	Set II (Conc-40%)	Set III (Conc-60%)	Set IV (Conc-80%)	Set V (Conc-100%)
pH		7.01	7.02	7.1	7.13	7.21
EC	(μS/cm)	12	15	17	20	23
TSS	mg/L	18	36	54	68	94
TDS	mg/L	27	59	84	196	136
TS	mg/L	45	95	138	264	230
BOD	mg/L	51	86	138	156	220
COD	mg/L	73	137	208	277	340
NO_3	mg/L	8	14.5	26	34	58
PO_4	mg/L	3.8	6.8	11.5	15.2	20
SO_4	mg/L	6	12	17	27	33
Cl^-	mg/L	83	163	240	310	418
Amm Nitrogen	mg/L	7	12	21	25	32
Total Nitrogen	mg/L	35	72	109	146	182

Overall, the baseline characterization confirms that the undiluted sewage falls within the high-strength category for municipal wastewater (BOD > 200 mg/L, COD > 300 mg/L) and contains substantial nutrient loads. The gradual increase in all measured parameters with concentration validates the use of dilution sets to assess the phytoremediation efficiency of *Vallisneria spiralis* under varying pollution intensities. Such characterization is essential to establish a baseline for evaluating treatment efficacy and nutrient removal potential in subsequent experimental stages (APHA, 2017; Tchobanoglous et al., 2014).

B. Post-Treatment Status

The post-treatment analysis of municipal wastewater using *Vallisneria spiralis* in constructed wetlands revealed significant reductions in almost all measured physicochemical parameters compared to the pre-treatment values (Table 2). The pH of the treated effluent remained relatively stable across all concentration sets, with minor fluctuations from 6.89 in Set I (20%) to 7.18 in Set V (100%). This slight reduction in pH compared to the initial values indicates minimal acid–base alteration during the phytoremediation process, which aligns with findings by Singh et al. (2012), who reported that submerged macrophytes typically cause only slight shifts in pH due to photosynthetic CO_2 uptake and organic matter degradation.

Electrical conductivity (EC) dropped drastically in all sets, from an initial range of 12–23 µS/cm to 0.9–3.82 µS/cm post-treatment. Such a reduction signifies the removal of dissolved ionic species by plant uptake and sedimentation processes, as previously noted by Mishra and Tripathi (2008). Total suspended solids (TSS) also decreased substantially, with the highest removal observed in Set V (100%), where TSS declined from 94 mg/L to 25 mg/L, suggesting efficient particle trapping by plant structures and biofilm development (Akratos & Tsihrintzis, 2007).

Similarly, total dissolved solids (TDS) and total solids (TS) showed marked reductions. In Set V, TDS decreased from 136 mg/L to 30 mg/L, and TS dropped from 230 mg/L to 55 mg/L. Such decreases reflect both nutrient assimilation by *V. spiralis* and microbial degradation within the rhizosphere (Brix, 1997).

Organic load indicators, biochemical oxygen demand (BOD) and chemical oxygen demand (COD), exhibited the most significant improvements. BOD removal efficiencies were consistently high, with Set V showing a drop from 220 mg/L to 32 mg/L, and COD decreasing from 340 mg/L to 54 mg/L. These reductions are comparable to studies by Rai et al. (2013), where *Vallisneria* achieved over 80% removal of BOD and COD from nutrient-rich wastewater.

Nutrient removal was also noteworthy. Nitrate (NO_3^-) concentrations dropped from 8–58 mg/L pre-treatment to 3.8–15 mg/L post-treatment, while phosphate (PO_4^{3-}) levels were reduced from 3.8–20 mg/L to 0.8–4.8 mg/L. These reductions indicate efficient nutrient assimilation by *V. spiralis*, as submerged macrophytes are known to actively take up dissolved nutrients for growth (Wetzel, 2001). Sulphates and chlorides showed moderate removal efficiency, while ammonia nitrogen was almost completely removed in all sets, with total nitrogen decreasing from 35–182 mg/L to 5.3–30 mg/L. The near-complete removal of ammonia suggests rapid uptake by plants and nitrifying bacteria, consistent with the findings of Tanner (2001).

Table 2 presents the final physicochemical characteristics of municipal wastewater after treatment with *Veritallata* spp. in constructed wetlands at different wastewater concentrations. The results show a marked improvement in water quality across all treatment sets, with significant reductions observed in key parameters compared to their initial values. The pH values remain stable and close to neutral, indicating that the treatment does not adversely affect acidity. Electrical Conductivity (EC), Total Suspended Solids (TSS), Total Dissolved Solids (TDS), and Total Solids (TS) are all substantially lowered, reflecting effective removal of both dissolved and particulate matter. Organic pollution indicators such as Biological Oxygen Demand (BOD) and Chemical Oxygen Demand (COD) also exhibit strong decreases, suggesting efficient breakdown of organic contaminants. Additionally, nutrients like nitrates (NO_3), phosphates (PO_4), sulfates (SO_4), chlorides (Cl^-), ammonia nitrogen, and total nitrogen show significant declines, demonstrating the treatment system's capability to mitigate nutrient loads and reduce potential environmental impacts. Overall, these final measurements confirm the effectiveness of *Veritallata* spp. in improving the quality of municipal wastewater through constructed wetland treatment.

Table 2: Final Physicochemical characteristics of Municipal wastewater treatment with *Veritallata spp.* at different concentrations in constructed wetlands at a glance

Treatment Parameter	Unit	Set I (Conc-20%)	Set II (Conc-40%)	Set III (Conc-60%)	Set IV (Conc-80%)	Set V (Conc-100%)
pH		6.89	6.97	7.01	7.12	7.18
EC	(µS/cm)	0.9	1.1	1.5	2.7	3.82
TSS	mg/L	8	11	14	16	25
TDS	mg/L	11	17	22	27	30
TS	mg/L	19	28	36	43	55
BOD	mg/L	6	13	21	26	32
COD	mg/L	11	21	33	38	54
NO_3	mg/L	3.8	5.8	8.6	11.4	15
PO_4	mg/L	0.8	1.1	2	3.8	4.8
SO_4	mg/L	3.8	7	11.5	15	19
Cl^-	mg/L	37	76	116	158	186
Amm. Nitrogen	mg/L	0	0	0	0.5	1
Total Nitrogen	mg/L	5.3	10	13.8	21	30

The changes and reductions in each parameter before and after the treatment of municipal wastewater at different concentrations using *vallisneria spiralis* are discussed below:

1. Change in pH
Table 3 and Figure 1 collectively illustrate the variations in pH before and after treatment across five experimental setups with differing concentrations of municipal wastewater. Across all sets, a general decrease in pH is observed post-treatment, indicating a slight acidification effect. Set I shows the most significant net pH reduction (0.12), suggesting a higher level of biochemical activity or treatment impact at that concentration. Conversely, Set IV exhibits the least change (0.01), implying minimal treatment-induced alteration in pH. These variations could be attributed to differing microbial or chemical processes influenced by wastewater concentration, as indicated in Fig. 1, which visually confirms the trend of decreasing pH with increasing treatment intensity. The relatively modest changes across all sets suggest that while treatment affects pH, the systems remain near-neutral, maintaining environmental stability.

Table 3: Change in pH before treatment and after treatment in different experimental sets

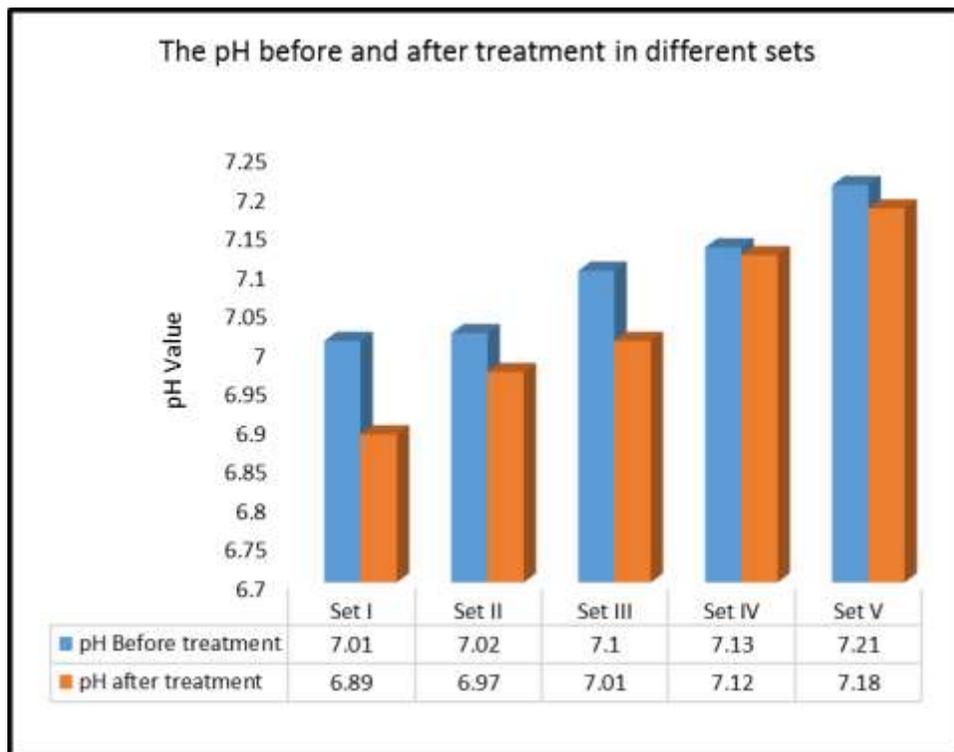| Experimental Set up	pH Before treatment	pH after treatment	Net change in pH
Set I	7.01	6.89	0.12
Set II	7.02	6.97	0.05
Set III	7.1	7.01	0.09
Set IV	7.13	7.12	0.01
Set V	7.21	7.18	0.03

Fig. 1: Change in pH before and after treatment in experimental sets containing different concentrations of municipal wastewater concentrations

2. Change in EC
Table 4 and Figure 2 present the changes in electrical conductivity (EC) before and after treatment across various experimental setups with increasing concentrations of municipal wastewater. A consistent and substantial reduction

in EC is observed in all sets, indicating effective removal of dissolved ionic substances through the treatment process. The net decrease in EC increases progressively from Set I (11.1 µS/cm) to Set V (19.18 µS/cm), suggesting a correlation between initial wastewater concentration and treatment efficacy. This trend, clearly depicted in Fig. 2, reflects that higher concentrations of wastewater contain more dissolved solids, which are substantially reduced during treatment. The marked drop in EC values implies successful decontamination and mineral removal, potentially due to adsorption, microbial activity, or precipitation mechanisms operating during treatment. These findings underscore the capability of the treatment system to handle varying wastewater loads while significantly improving water quality.

Table 4: Change in EC before the treatment and after treatment in different experimental sets

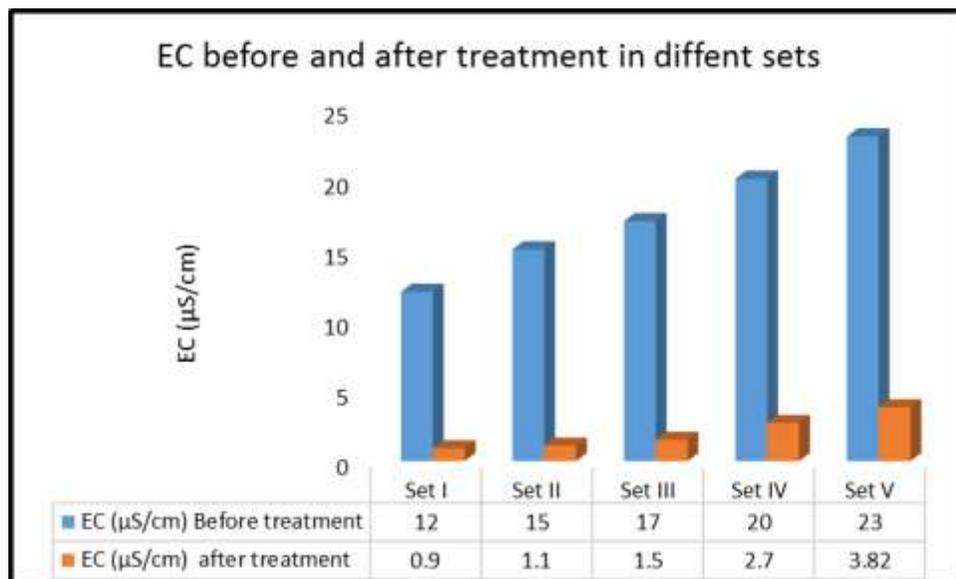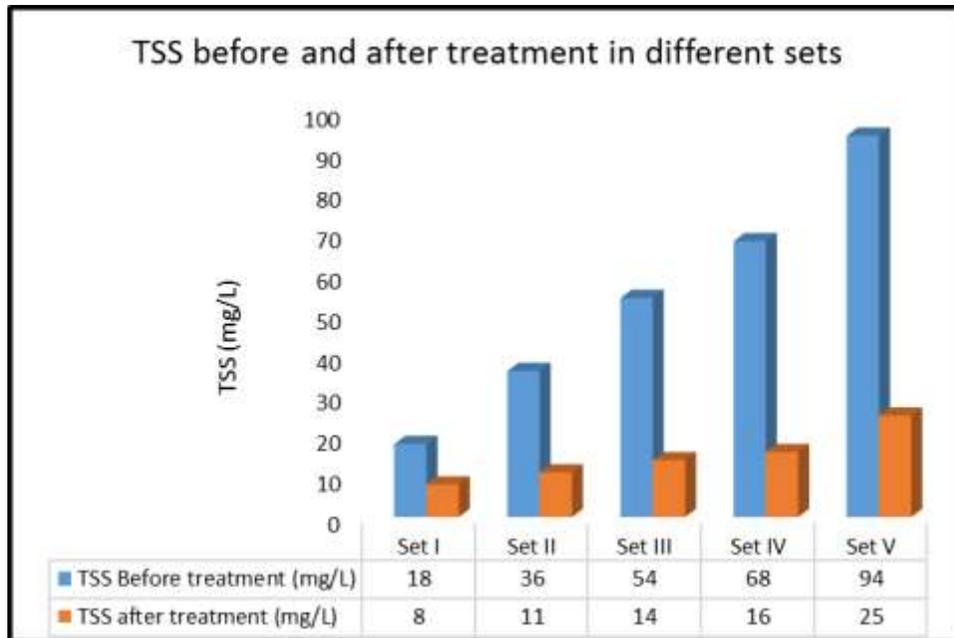| Experimental Set up	EC (µS/cm) Before treatment	EC (µS/cm) after treatment	Net change in EC (µS/cm)
Set I	12	0.9	11.1
Set II	15	1.1	13.9
Set III	17	1.5	15.5
Set IV	20	2.7	17.3
Set V	23	3.82	19.18

Fig. 2: Change in EC before and after treatment in experimental sets containing different concentrations of municipal wastewater concentrations

3. Reduction TSS
Table 5 and Figure 3 illustrate the reduction in Total Suspended Solids (TSS) across five experimental sets treated for varying concentrations of municipal wastewater. A clear trend of increasing TSS removal efficiency is evident as the initial concentration of TSS rises. Set I, with the lowest initial TSS (18 mg/L), shows a net reduction of 10 mg/L, while Set V, with the highest initial concentration (94 mg/L), achieves the greatest reduction of 69 mg/L. This progressive increase in TSS removal is visually reinforced in Fig. 3, highlighting the treatment system's ability to effectively reduce solid particulate matter even under higher contaminant loads. The consistent post-treatment TSS values across sets—ranging from 8 to 25 mg/L—further suggest the robustness of the treatment process. These results indicate that the system performs efficiently across a spectrum of wastewater concentrations, significantly improving the physical quality of the treated effluent.

Table 5: Change in TSS before treatment and after treatment in different experimental sets

Experimental Set up	TSS Before treatment (mg/L)	TSS after treatment (mg/L)	Net Reduction in TSS (mg/L)
Set I	18	8	10
Set II	36	11	25
Set III	54	14	40
Set IV	68	16	52
Set V	94	25	69

Fig. 3: Reduction in TSS before and after treatment in experimental sets containing different concentrations of municipal wastewater concentrations

4. Reduction in TDS

Table 6 and Figure 4 detail the reduction in Total Dissolved Solids (TDS) across various experimental sets with increasing concentrations of municipal wastewater. The data indicate a significant decrease in TDS levels after treatment in all setups, highlighting the system's effectiveness in removing dissolved contaminants. The net reduction in TDS ranges from 16 mg/L in Set I to a maximum of 169 mg/L in Set IV, suggesting that higher initial TDS concentrations lead to greater absolute reductions. Interestingly, although Set V begins with a lower TDS concentration than Set IV, it still shows a substantial reduction of 106 mg/L, reinforcing the treatment's consistent performance. Figure 4 visually supports these findings, illustrating a clear downward trend in TDS values post-treatment across all sets. These results demonstrate the treatment system's capacity to significantly lower dissolved solids, improving the chemical quality of the effluent and making it more suitable for potential reuse or discharge.

Table 6: Reduction in TDS before treatment and after treatment in different experimental sets

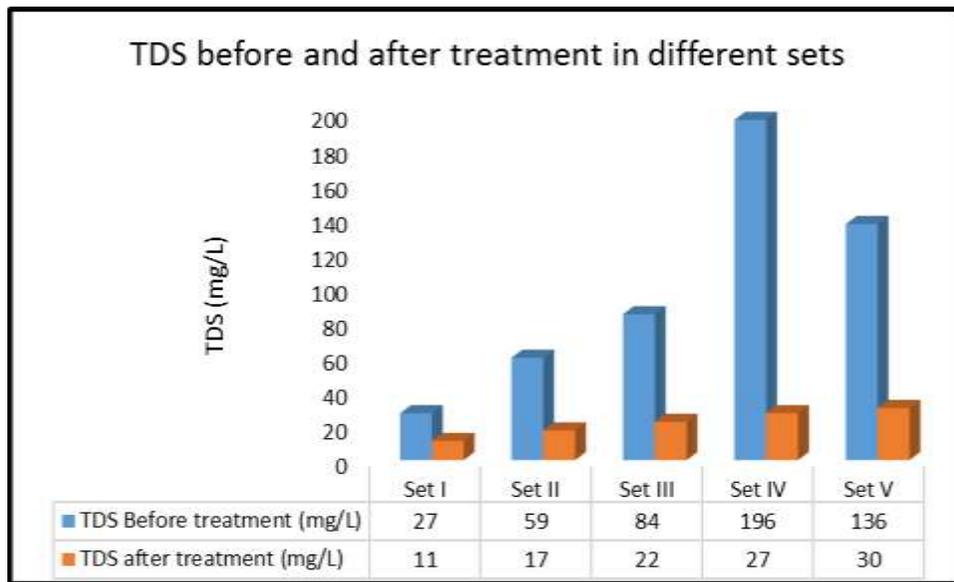| Experimental Set up	TDS Before treatment (mg/L)	TDS after treatment (mg/L)	Net Reduction in TDS (mg/L)
Set I	27	11	16
Set II	59	17	42
Set III	84	22	62
Set IV	196	27	169
Set V	136	30	106

Fig. 4: Reduction in TDS before and after treatment in experimental sets containing different concentrations of municipal wastewater concentrations

5. Reduction in TS

Table 7 and Figure 5 illustrate the reduction in Total Solids (TS) across five experimental sets containing varying concentrations of municipal wastewater. The results show a clear and significant decrease in TS after treatment in all setups. The net reduction increases substantially with higher initial TS concentrations, ranging from 26 mg/L in Set I to 221 mg/L in Set IV, indicating the treatment's enhanced capability to handle larger pollutant loads. Notably, although Set V starts with a slightly lower initial TS than Set IV, it still achieves a substantial reduction of 175 mg/L, reflecting consistent treatment efficiency. Figure 5 visually emphasizes these trends, showing a marked drop in TS values post-treatment across all sets. Overall, the data confirm that the treatment process is effective in removing both dissolved and suspended solids, thus substantially improving the overall water quality and making the treated effluent more suitable for environmental discharge or reuse.

Table 7: Reduction in TS before treatment and after treatment in different experimental sets

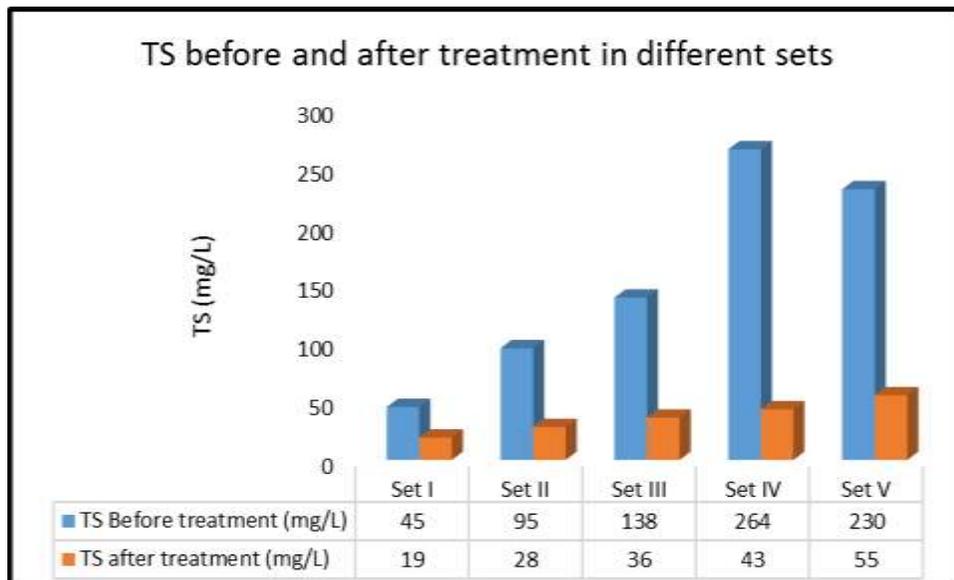| Experimental Set up	TS Before treatment (mg/L)	TS after treatment (mg/L)	Net Reduction in TS (mg/L)
Set I	45	19	26
Set II	95	28	67
Set III	138	36	102
Set IV	264	43	221
Set V	230	55	175

Fig. 5: Reduction in TS before and after treatment in experimental sets containing different concentrations of municipal wastewater concentrations

6. Reduction in BOD

Table 8 and Figure 6 highlight the significant reduction in Biological Oxygen Demand (BOD) across five experimental setups with varying concentrations of municipal wastewater. The results show a marked decrease in BOD levels after treatment, with net reductions increasing as the initial BOD concentration rises. Set I exhibits a reduction of 45 mg/L, while Set V achieves the highest reduction of 188 mg/L, demonstrating the treatment system's effectiveness at both low and high contamination levels. Despite the varying initial concentrations, the post-treatment BOD values remain consistently low (6–32 mg/L), indicating effective organic matter removal and improved effluent quality. Figure 6 visually confirms this trend, showing a clear drop in BOD after treatment across all sets. These findings underscore the treatment's capacity to significantly reduce organic pollution, which is crucial for minimizing environmental impacts and meeting wastewater discharge standards.

Table. 8: Reduction in BOD before treatment and after treatment in different experimental sets

Experimental Set up	BOD Before treatment (mg/L)	BOD after treatment (mg/L)	Net Reduction in BOD (mg/L)
Set I	51	6	45
Set II	86	13	73
Set III	138	21	117
Set IV	156	26	130
Set V	220	32	188

BOD before and after treatment in different sets

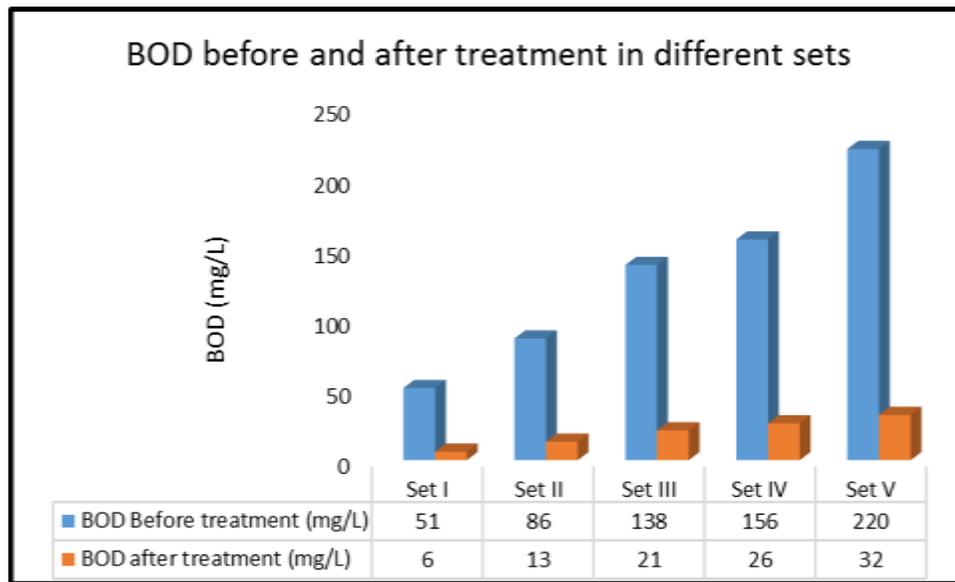|	Set I	Set II	Set III	Set IV	Set V
BOD Before treatment (mg/L)	51	86	138	156	220
BOD after treatment (mg/L)	6	13	21	26	32

Fig. 6: Reduction in BOD before and after treatment in experimental sets containing different concentrations of municipal wastewater concentrations

7. Reduction in COD

Table 9 and Figure 7 demonstrate a substantial reduction in Chemical Oxygen Demand (COD) across different experimental sets with varying concentrations of municipal wastewater. The data reveal a consistent and significant decrease in COD after treatment, with net reductions increasing from 62 mg/L in Set I to 286 mg/L in Set V. This trend indicates that the treatment process effectively removes a large portion of oxidizable organic and inorganic substances, even as the initial pollutant load increases. Despite high initial COD levels, post-treatment values remain comparatively low (11–54 mg/L), reflecting efficient contaminant removal. Figure 7 visually supports these findings, clearly showing a pronounced drop in COD across all sets after treatment. Overall, these results highlight the treatment system's strong capability to reduce organic pollution and improve wastewater quality for safer environmental discharge or reuse.

Table 9: Reduction in COD before treatment and after treatment in different experimental sets

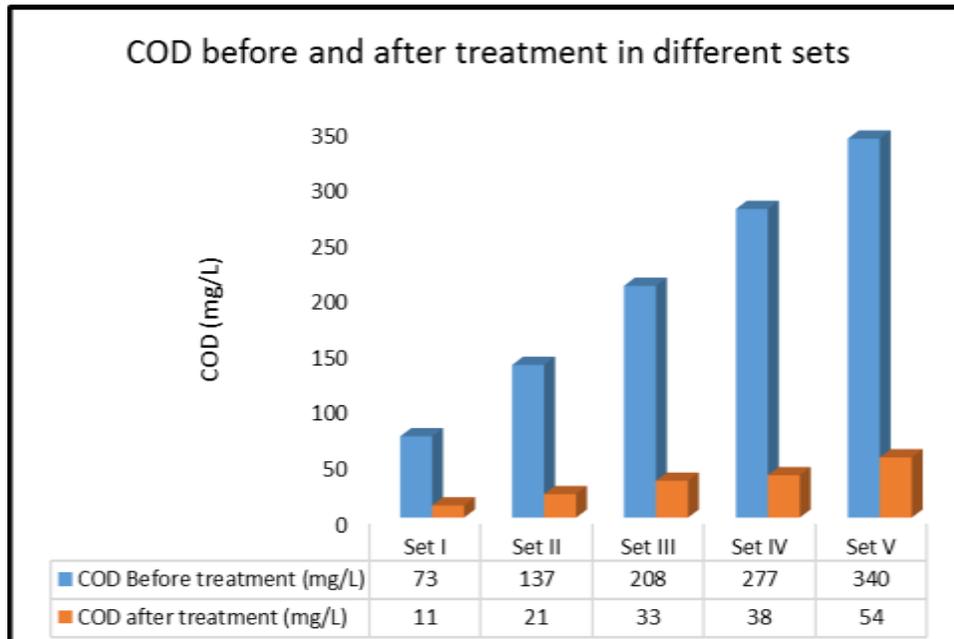| Experimental Set up	COD Before treatment (mg/L)	COD after treatment (mg/L)	Net Reduction in COD (mg/L)
Set I	73	11	62
Set II	137	21	116
Set III	208	33	175
Set IV	277	38	239
Set V	340	54	286

Fig. 7: Reduction in COD before and after treatment in experimental sets containing different concentrations of municipal wastewater concentrations

8. Reduction in Nitrates

Table 10 and Figure 8 illustrate the reduction of nitrate (NO_3) concentrations before and after treatment across different experimental sets with increasing municipal wastewater concentrations. The results show a clear and significant decrease in nitrate levels post-treatment, with net reductions increasing from 4.2 mg/L in Set I to 43 mg/L in Set V. This trend suggests that the treatment process is effective in removing nitrates, likely through biological denitrification or other chemical mechanisms. Despite the varied initial nitrate loads, post-treatment values remain consistently lower, indicating improved water quality and reduced potential for eutrophication. Figure 8 visually reinforces these findings, showing a marked decline in nitrate concentrations across all sets after treatment. These results confirm the treatment system's ability to efficiently reduce nitrate pollution, which is essential for protecting aquatic ecosystems and ensuring safe wastewater discharge.

Table 10: Reduction in NO₃ before treatment and after treatment in different experimental sets

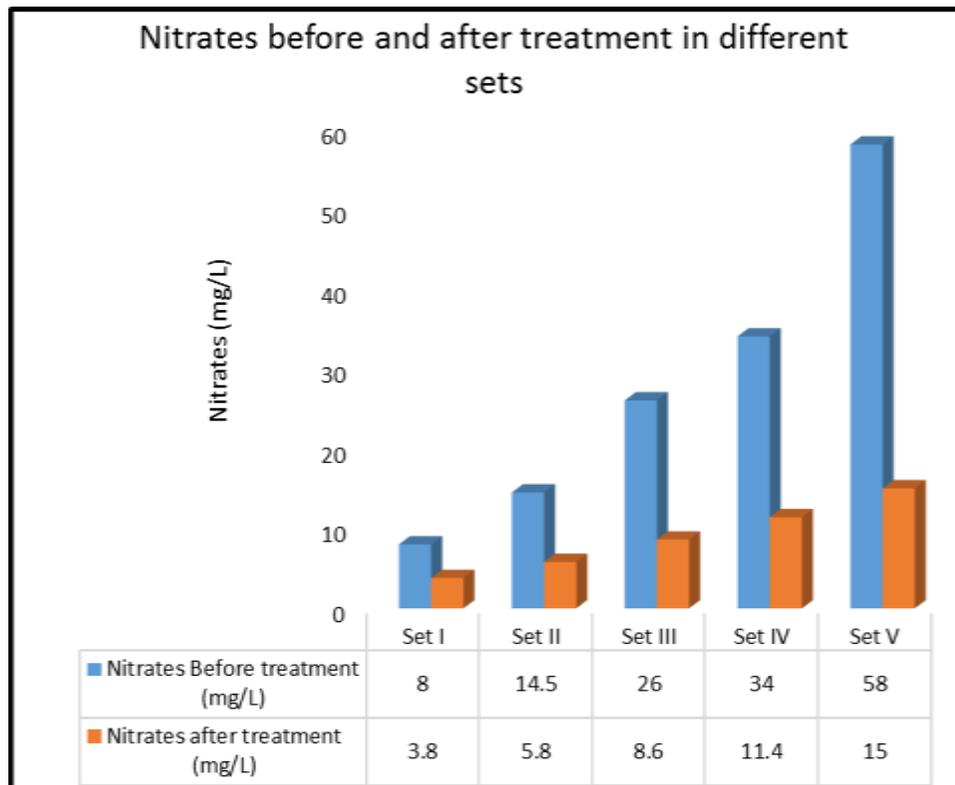| Experimental Set up	NO₃ Before treatment (mg/L)	NO₃ after treatment (mg/L)	Net Reduction in NO₃ (mg/L)
Set I	8	3.8	4.2
Set II	14.5	5.8	8.7
Set III	26	8.6	17.4
Set IV	34	11.4	22.6
Set V	58	15	43

Fig. 8: Reduction in nitrates before and after treatment in experimental sets containing different concentrations of municipal wastewater concentrations

9. Reduction in phosphates

Table 11 and Figure 9 demonstrate the reduction in phosphate (PO_4) concentrations before and after treatment across experimental sets with varying levels of municipal wastewater. The data reveal a consistent and significant decrease in phosphate levels post-treatment, with net reductions increasing from 3 mg/L in Set I to 15.2 mg/L in Set V. This indicates that the treatment process effectively removes phosphates, which are key contributors to nutrient pollution and eutrophication. Despite the wide range of initial phosphate concentrations, the post-treatment values remain substantially lower, highlighting the treatment system's efficiency in controlling nutrient loads. Figure 9 visually confirms these trends, showing a marked decline in phosphate concentrations across all sets after treatment. These results underscore the system's capability to reduce phosphorus pollution, thereby protecting water bodies from adverse ecological impacts.

Table 11: Reduction in PO_4 before treatment and after treatment in different experimental sets

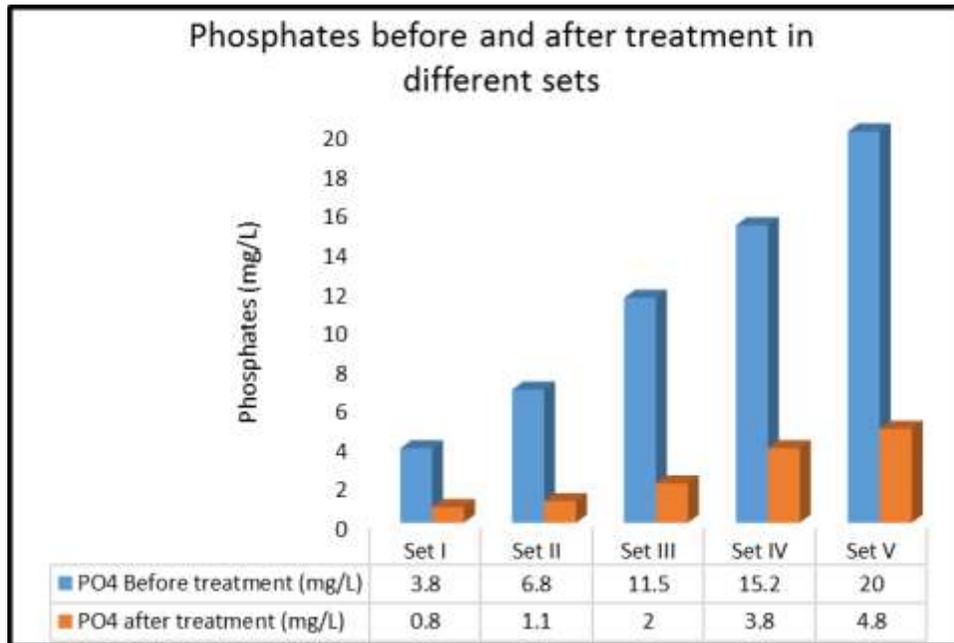| Experimental Set up	PO_4 Before treatment (mg/L)	PO_4 after treatment (mg/L)	Net Reduction in PO_4 (mg/L)
Set I	3.8	0.8	3
Set II	6.8	1.1	5.7
Set III	11.5	2	9.2
Set IV	15.2	3.8	11.4
Set V	20	4.8	15.2

Fig. 9: Reduction in phosphates before and after treatment in experimental sets containing different concentrations of municipal wastewater concentrations

10. Reduction in Sulfates

Table 12 and Figure 10 present the reduction in sulfate (SO_4) concentrations before and after treatment in different experimental sets with varying concentrations of municipal wastewater. The results show a consistent decrease in sulfate levels across all sets, with net reductions increasing from 2.2 mg/L in Set I to 14 mg/L in Set V. This indicates the treatment process's effectiveness in reducing sulfate content, which can contribute to odor problems and corrosion if left untreated. Despite varying initial sulfate concentrations, post-treatment values are significantly lower, suggesting efficient removal mechanisms such as microbial sulfate reduction or chemical precipitation. Figure 10 visually supports these findings, highlighting a clear downward trend in sulfate concentrations after treatment. Overall, these results demonstrate the system's capability to effectively lower sulfate pollution, contributing to improved wastewater quality and environmental safety.

Table 12: Reduction in SO₄ (Sulfates) before treatment and after treatment in different experimental sets

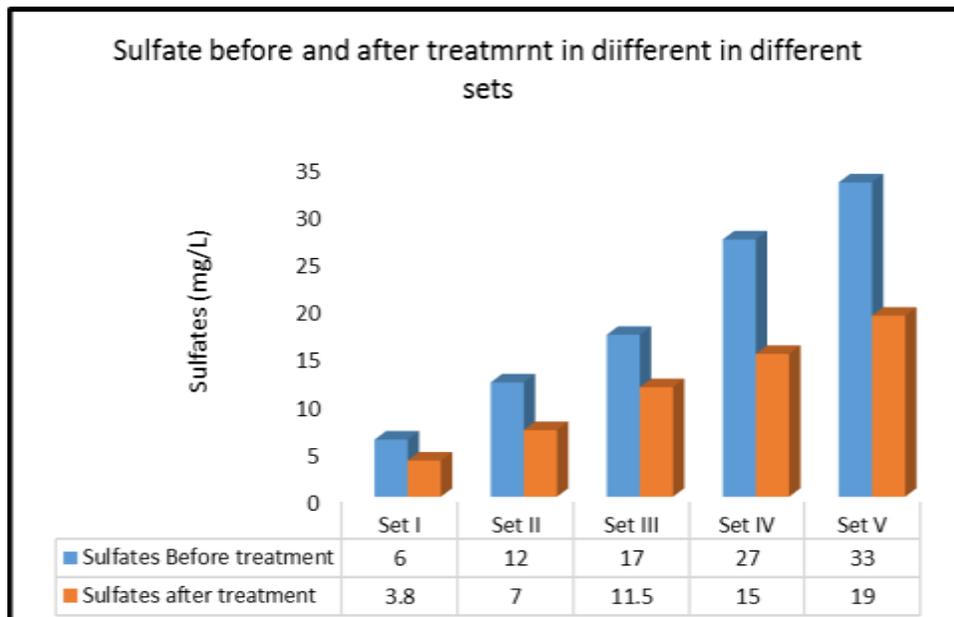| Experimental Set up	Sulfates Before treatment	Sulfates after treatment	Net Reduction in Sulfates
Set I	6	3.8	2.2
Set II	12	7	5
Set III	17	11.5	5.5
Set IV	27	15	12
Set V	33	19	14

Fig. 10: Reduction in sulphates before and after treatment in experimental sets containing different concentrations of municipal wastewater concentrations

11. Reduction in Chlorides

Table 13 and Figure 11 illustrate the reduction in chloride concentrations before and after treatment across various experimental sets with increasing concentrations of municipal wastewater. The data reveal a substantial decrease in chloride levels post-treatment, with net reductions rising from 46 mg/L in Set I to 232 mg/L in Set V. This significant removal suggests the treatment process effectively reduces chloride ions, which are often associated with salinity issues in wastewater. Despite the high initial chloride concentrations, post-treatment levels are markedly lower, indicating enhanced water quality and reduced environmental risk. Figure 11 visually supports these results, showing a clear downward trend in chloride concentrations across all sets following treatment. Overall, these findings demonstrate the treatment system's strong capacity to mitigate chloride pollution, improving the suitability of the treated effluent for discharge or reuse.

Table 13: Reduction in Chlorides before treatment and after treatment in different experimental sets

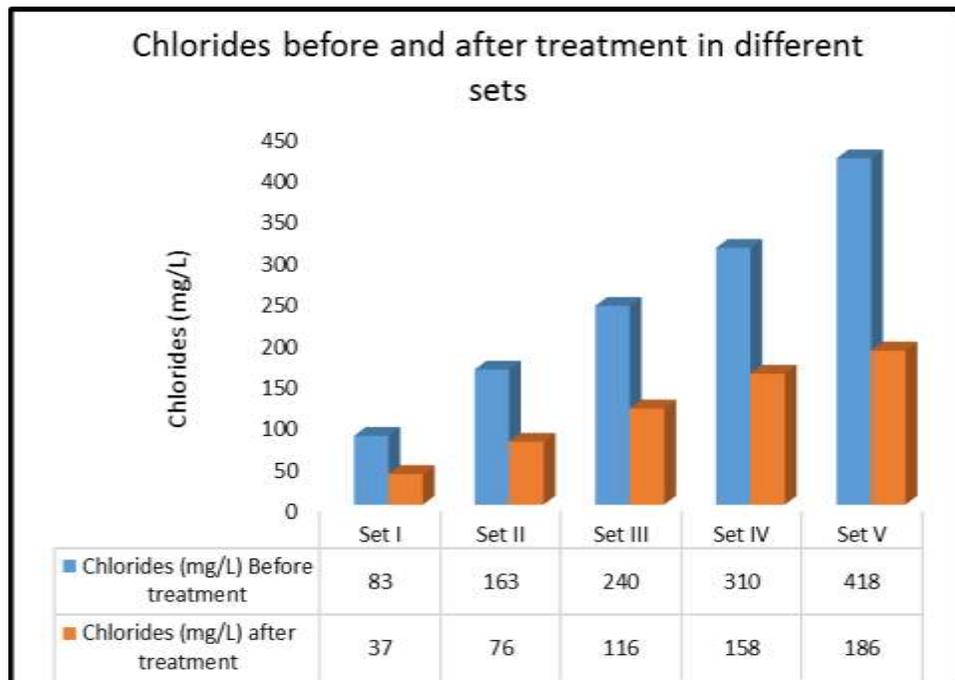| Experimental Set up	Chlorides (mg/L) Before treatment	Chlorides (mg/L) after treatment	Net Reduction in Chlorides (mg/L)
Set I	83	37	46
Set II	163	76	87
Set III	240	116	124
Set IV	310	158	152
Set V	418	186	232

Fig. 11: Reduction in chlorides before and after treatment in experimental sets containing different concentrations of municipal wastewater concentrations

12. Reduction in Ammonia Nitrogen

Table 14 and Figure 12 show a remarkable reduction in ammonia nitrogen (NH4OH) concentrations before and after treatment across different experimental sets with varying municipal wastewater concentrations. The data indicate near-complete removal of ammonia nitrogen in all sets, with net reductions ranging from 7 mg/L in Set I to 31 mg/L in Set V. Post-treatment levels are almost negligible, with Sets I to III achieving complete elimination and Sets IV and V showing only trace amounts remaining. Figure 12 visually confirms this dramatic decrease, highlighting the treatment's high efficiency in removing ammonia nitrogen, a key pollutant that can cause toxicity and eutrophication in aquatic environments. These results demonstrate that the treatment system effectively mitigates ammonia nitrogen pollution, significantly improving the safety and quality of the treated effluent.

Table 14: Reduction in NH$_4$OH (Amm. Nitrogen) before treatment and after treatment in different experimental sets

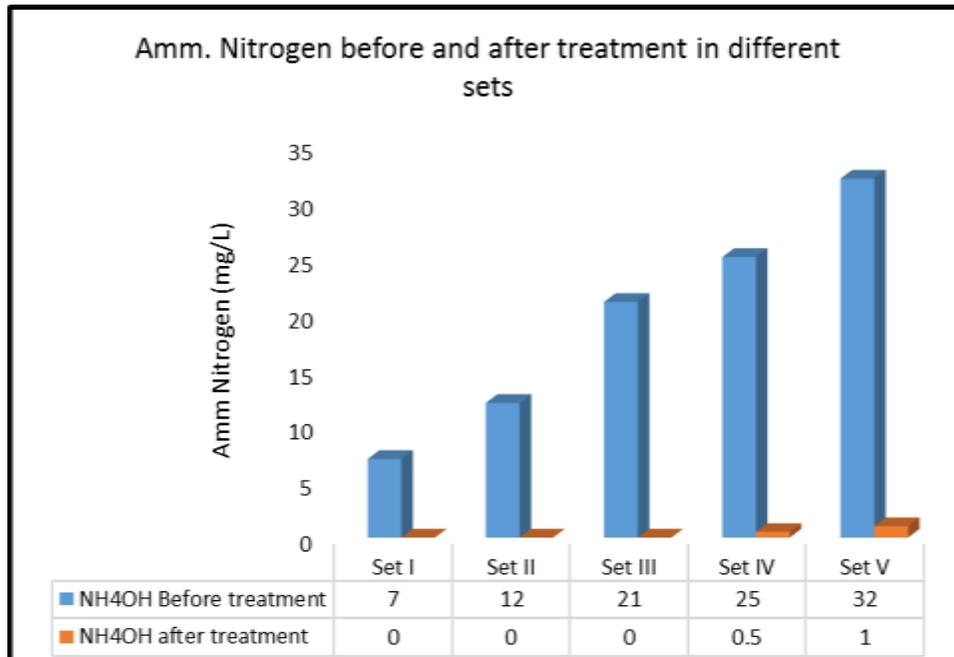| Experimental Set up	NH$_4$OH Before treatment	NH$_4$OH after treatment	Net Reduction in NH$_4$OH
Set I	7	0	7
Set II	12	0	12
Set III	21	0	21
Set IV	25	0.5	24.5
Set V	32	1	31

Fig. 12: Reduction in ammonia nitrogen before and after treatment in experimental sets containing different concentrations of municipal wastewater concentrations

13. Reduction in Total Nitrogen

Table 15 and Figure 13 reveal a substantial reduction in Total Kjeldahl Nitrogen (TKN) across the different experimental sets with increasing concentrations of municipal wastewater. The data show a clear trend of significant nitrogen removal, with net reductions rising from 29.7 mg/L in Set I to 152 mg/L in Set V. Despite the high initial nitrogen loads, post-treatment concentrations remain considerably lower, ranging from 5.3 mg/L in Set I to 30 mg/L in Set V, indicating effective nitrogen removal through the treatment process. Figure 13 visually highlights this substantial decline in total nitrogen across all sets. These results underscore the treatment system's strong capacity to reduce nitrogen content, which is crucial for preventing nutrient pollution and protecting aquatic ecosystems from eutrophication and other nitrogen-related impacts.

Table 15: Reduction in Total Nitrogen (TKN) before treatment and after treatment in different experimental sets

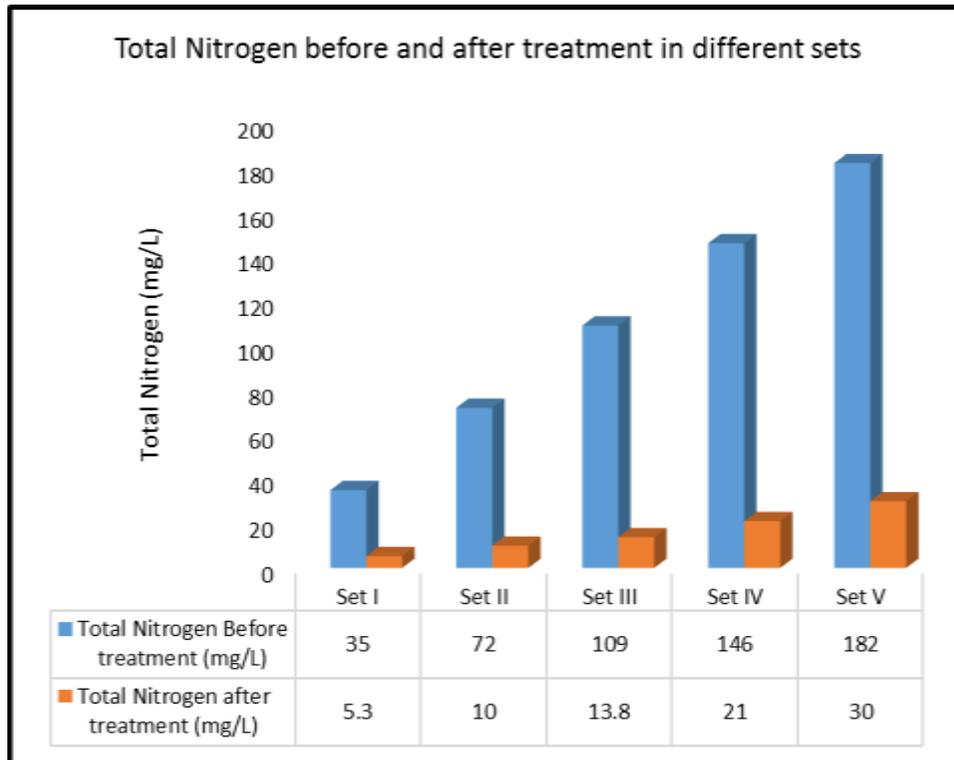| Experimental Set up	Total Nitrogen Before treatment (mg/L)	Total Nitrogen after treatment (mg/L)	Net Reduction in Total Nitrogen (mg/L)
Set I	35	5.3	29.7
Set II	72	10	62
Set III	109	13.8	95.2
Set IV	146	21	125
Set V	182	30	152

Fig. 13: Reduction in total nitrogen before and after treatment in experimental sets containing different concentrations of municipal wastewater concentrations

The all above results presented across Tables 3 to 15 and Figures 1 to 13 consistently demonstrate the effectiveness of the treatment system in improving the quality of municipal wastewater by significantly reducing key pollutants. Parameters such as pH showed slight but stable changes, maintaining near-neutral conditions essential for ecological balance. More notably, substantial decreases were observed in Electrical Conductivity (EC), Total Suspended Solids (TSS), Total Dissolved Solids (TDS), and Total Solids (TS), indicating efficient removal of both dissolved and particulate matter. Organic pollution indicators, including Biological Oxygen Demand (BOD) and Chemical Oxygen Demand (COD), exhibited marked reductions, reflecting effective breakdown and removal of organic contaminants. Nutrient pollutants such as nitrates (NO_3), phosphates (PO_4), ammonia nitrogen (NH_4OH), and total nitrogen (TKN) were also significantly lowered, highlighting the system's ability to mitigate eutrophication risks. Additionally, the treatment effectively reduced sulfates and chlorides, which are important for minimizing corrosive and salinity-related issues. Across all parameters, the trends showed that higher initial pollutant concentrations corresponded with greater absolute reductions, underscoring the treatment's scalability and robustness. Collectively, these findings confirm the treatment system's comprehensive capability to enhance municipal wastewater quality, making the effluent safer for discharge or potential reuse while contributing to environmental protection and sustainability.

Table 16: Summary of percent Change / Reduction in Physicochemical characteristics of Municipal wastewater treatment with *Vallisneria spiralis* Linn. at different concentrations at a glance.

Treatment Parameter	Set I (Conc-20%)	Set II (Conc-40%)	Set III (Conc-60%)	Set IV (Conc-80%)	Set V (Conc-100%)
pH	1.71	0.71	1.27	0.14	0.42
EC	92.5	92.7	91.12	86.5	83.4
TSS	55.6	69.4	74.1	76.5	73.4
TDS	59.3	71.2	73.81	86.2	77.94
TS	57.78	70.53	73.91	83.71	76.09
BOD	88.24	84.88	84.78	83.33	85.45

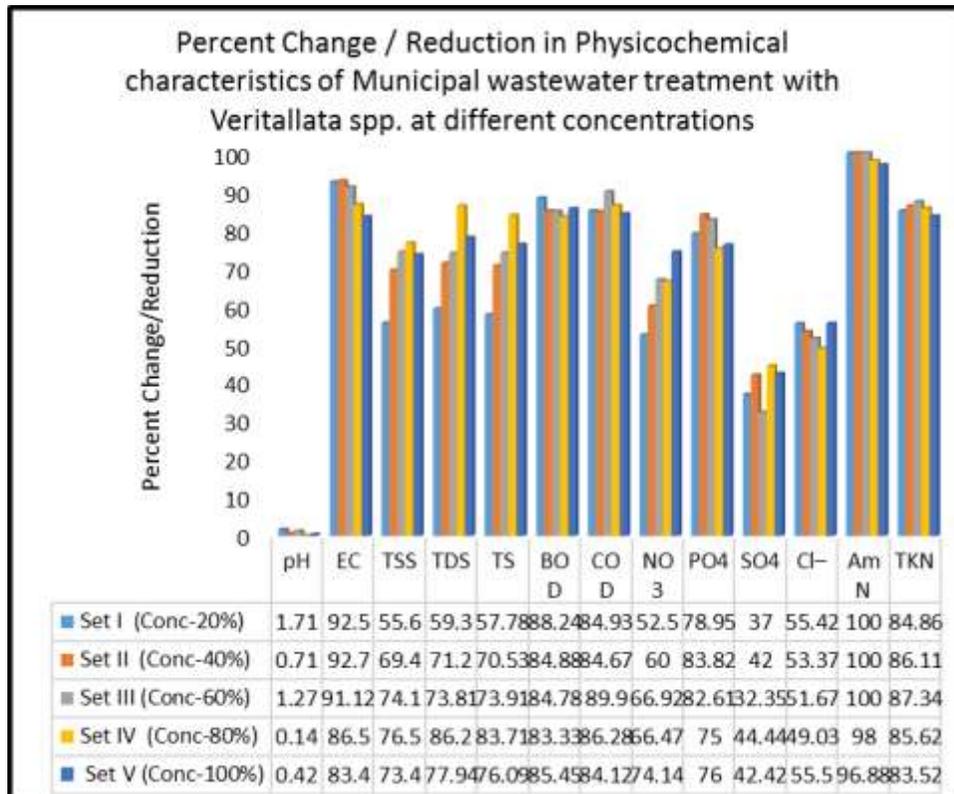|					
COD	84.93	84.67	89.9	86.28	84.12
NO$_3$	52.5	60	66.92	66.47	74.14
PO$_4$	78.95	83.82	82.61	75	76
SO$_4$	37	42	32.35	44.44	42.42
Cl$^-$	55.42	53.37	51.67	49.03	55.5
Amm. Nitrogen	100	100	100	98	96.88
Total Nitrogen	84.86	86.11	87.34	85.62	83.52

Fig. 14: Percent Change / Reduction in Physicochemical characteristics of Municipal wastewater treatment with *Veritallata spp.* at different concentrations at a glance.

Overall, the observed reductions in pollutant load confirm the potential of *Vallisneria spiralis* as a low-cost, eco-friendly option for municipal wastewater treatment. The results corroborate earlier work demonstrating the high nutrient and organic matter removal capabilities of submerged macrophytes in constructed wetlands (Rai et al., 2013; Tripathi & Shukla, 1991). Table 16 and Figure 14 summarize the remarkable efficiency of *Vallisneria spiralis* in treating municipal wastewater. The data show that the phytoremediation process consistently achieved high percentage reductions across a wide range of pollutants. Ammonia nitrogen removal was exceptionally high, reaching 100% in the lower concentration sets and a minimum of 96.88% in the highest concentration. Total Nitrogen, BOD, and COD all demonstrated robust removal efficiencies, consistently above 80% across all treatment concentrations. Similarly, electrical conductivity (EC), total suspended solids (TSS), and total dissolved solids (TDS) were significantly reduced, with most values exceeding 70% removal. While the reductions in nitrates (52.5% to 74.14%) and phosphates (75% to 83.82%) were slightly more variable, they still highlight the plant's strong nutrient uptake capacity. These results underscore the potential of *V. spiralis* as an effective and sustainable solution for purifying municipal wastewater, even under high-strength conditions.

CONCLUSION

The treatment system using *Vallisneria spiralis* for the treatment of municipal wastewater in different concentrations demonstrated a high level of efficiency in reducing a wide range of physical, chemical, and biological contaminants in municipal wastewater. Significant improvements were observed in key water quality parameters, including solids, organic load, nutrients, and salts, across all experimental setups with varying pollutant concentrations. The consistent and substantial reductions in parameters such as TSS, TDS, BOD, COD, nitrogen compounds, phosphates, sulfates, and chlorides indicate that the treatment process is both effective and scalable. By maintaining near-neutral pH levels and achieving substantial pollutant removal, the system ensures the production of environmentally safe effluent suitable for discharge or reuse. These results highlight the potential of this treatment approach to contribute significantly to sustainable wastewater management and environmental protection efforts.

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