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Integrated Assessment Of Chlorella Vulgaris, Scenedesmus Obliquus, And Nostoc Muscorum For Efficient Algal-Based Sewage Remediation

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Abstract
The increasing demand for sustainable wastewater treatment solutions has led to growing interest in algal-based remediation systems. This study investigates the efficacy of three microalgal species—Chlorella vulgaris, Scenedesmus obliquus, and Nostoc muscorum—in treating municipal sewage collected from Dibrugarh, Assam. Laboratory-scale batch reactors were inoculated with algal cultures and monitored over 14 days for biomass accumulation, nutrient removal, and changes in physicochemical parameters. Results showed significant pollutant reductions in algal-treated systems, with C. vulgaris achieving the highest removal efficiencies for COD (85.2%), BOD (89.6%), total nitrogen (78.5%), and total phosphorus (82.0%). Dissolved oxygen levels increased markedly, particularly in C. vulgaris treatments (from 1.2 mg/L to 7.8 mg/L), while pH rose due to active photosynthesis. Algal biomass growth was positively correlated with pollutant removal efficiency, confirming the potential of these organisms as bioagents in sewage remediation. Compared to the control, which showed minimal improvement, algal treatments demonstrated clear advantages in pollutant removal and ecological restoration. This study proposes a novel, eco-friendly approach for decentralised sewage management, emphasising algae as multifunctional agents in wastewater bioremediation and resource recovery.
Keywords: *Algae, Sewage, Chlorella vulgaris, Biological oxygen demand*

1. INTRODUCTION

The increasing global population and rapid urbanization have led to a significant surge in the generation of domestic and industrial wastewater, placing tremendous pressure on existing sewage treatment infrastructure. Traditional wastewater treatment methods, such as activated sludge processes, trickling filters, and membrane bioreactors, although effective, are often energy-intensive, expensive, and generate large amounts of sludge that require further processing and disposal (Talaiekhozani *et al.*, 2021). In this context, algal-based wastewater treatment systems have garnered considerable attention due to their potential to offer a low-cost, sustainable, and environmentally friendly alternative.

Microalgae, which are photosynthetic organisms capable of thriving in nutrient-rich environments, can assimilate significant quantities of nitrogen and phosphorus—the primary pollutants in municipal sewage—thereby mitigating eutrophication when effluents are discharged into aquatic systems (Koutra *et al.*, 2021). Unlike conventional treatment systems, algae can simultaneously perform carbon dioxide sequestration and oxygen generation through photosynthesis, supporting aerobic microbial activity that aids in the decomposition of organic matter (Jebali *et al.*, 2022). Furthermore, algal systems not only remove pollutants but also generate valuable biomass that can be used for biofuel production, animal feed, biofertilizers, and other bioproducts (Al-Harahsheh *et al.*, 2022).

Recent advances in biotechnology and photobioreactor design have significantly enhanced the efficiency and applicability of algae in wastewater treatment. Studies have demonstrated the feasibility of coupling algal cultivation with wastewater treatment in high-rate algal ponds, photobioreactors, and hybrid systems, which have shown promising results in terms of pollutant removal, cost-effectiveness, and biomass yield (Chen *et al.*, 2020; Rawat *et al.*, 2021). Additionally, co-cultivation of algae with bacteria (algal-bacterial consortia) has proven to be more effective in nutrient removal and system stability compared to monocultures, highlighting the importance of microbial synergy in sewage bioremediation (de Godos *et al.*, 2021).

This study aims to evaluate the potential of selected microalgal species—*Chlorella vulgaris, Scenedesmus obliquus,* and *Nostoc muscorum*—for the treatment of municipal sewage under controlled laboratory conditions. The study sought to assess the pollutant removal efficiency (COD, BOD, total nitrogen, total phosphorus, and heavy metals), monitor algal biomass growth, and investigate the influence of algal activity on key physicochemical parameters such as dissolved oxygen and pH.

2. METHODOLOGY

2.1 Study Design

This study was designed to assess the efficiency of microalgae in treating municipal sewage through a controlled laboratory-scale experiment. The methodology involved isolation and cultivation of microalgal species, treatment of sewage samples using algal cultures, and analytical assessment of water quality parameters before and after treatment. Comparative evaluation with a control (untreated sewage) was also conducted.

2.2 Site and Sample Collection

Raw sewage samples were collected from the sewages of Dibrugarh, Assam. Samples were collected in sterile 10-liter containers and transported to the laboratory within 4 hours under cooled conditions to prevent degradation of organic material.

2.3 Selection and Cultivation of Algae

Three algal species *Chlorella vulgaris, Scenedesmus obliquus* and *Nostoc muscorum* were selected based on their known performance in nutrient uptake and wastewater tolerance. These species were obtained from National Collection of Industrial Microorganisms, Pune and cultured in Bold's Basal Medium (BBM) under controlled conditions

2.4 Experimental Setup

Batch experiments were conducted in 5-litre glass reactors containing 3 litres of raw sewage and an algal inoculum at a concentration of 0.5 g/L dry weight. Triplicate reactors were maintained for each algal species, and one set was left untreated as a control. All systems were incubated for 14 days under laboratory conditions. Daily stirring and periodic aeration ensured homogeneous mixing and optimal gas exchange.

2.5 Analytical Procedures

Water quality parameters were measured at Day 0 (before treatment), Day 7, and Day 14. The physicochemical parameters of sewage water were analysed following the Standard Methods for the Examination of Water and Wastewater (APHA, AWWA, WEF, 2017). The pH was measured using a calibrated digital pH meter (APHA Method 4500-H^+ B). Dissolved oxygen (DO) was determined using the Winkler titration method (Method 4500-O C) and cross-verified with a DO meter. Chemical oxygen demand (COD) was assessed using the closed reflux titrimetric method (Method 5220 C), while biological oxygen demand (BOD_5) was measured using the 5-day BOD test (Method 5210 B). Total nitrogen (TN) was analysed using the Kjeldahl method for total Kjeldahl nitrogen (Method 4500-Norg C), and total phosphorus (TP) was estimated by the ascorbic acid method following acid digestion (Method 4500-P E) Algal growth was quantitatively monitored throughout the 14-day treatment period by measuring biomass accumulation in terms of dry weight (g/L). Samples were collected at regular intervals (Day 0, Day 7, and Day 14) from each reactor. A known volume of algal culture was filtered using Whatman No. 1 filter paper, followed by washing with distilled water to remove any adhering impurities. The filter papers with algal biomass were then oven-dried at 60 °C for 24 hours to a constant weight. The dry weight was recorded and used as a direct measure of biomass productivity. This method provided a reliable and reproducible estimate of the cellular accumulation of organic matter, which served as an indicator of algal adaptation, nutrient uptake efficiency, and photosynthetic performance in sewage environments (Sutherland *et al.*, 2020; Razzak *et al.*, 2013).

2. 6 Statistical Analysis

Data were analyzed using ANOVA to determine the significance of nutrient removal between treated and control groups. A confidence interval of 95% ($p < 0.05$) was used. Correlation analysis was also performed between algal biomass and pollutant removal efficiency using Pearson's correlation coefficient in SPSS v25.

3. RESULTS

3.1 Algal Biomass Growth

All three algal species *Chlorella vulgaris*, *Scenedesmus obliquus*, and *Nostoc muscorum*—demonstrated significant growth over the 14-day sewage treatment period, indicating their adaptability and tolerance to the wastewater environment. Biomass accumulation was quantified in terms of dry weight (g/L), and the growth curve showed a continuous increase, particularly pronounced in *Chlorella vulgaris* (Figure 1)

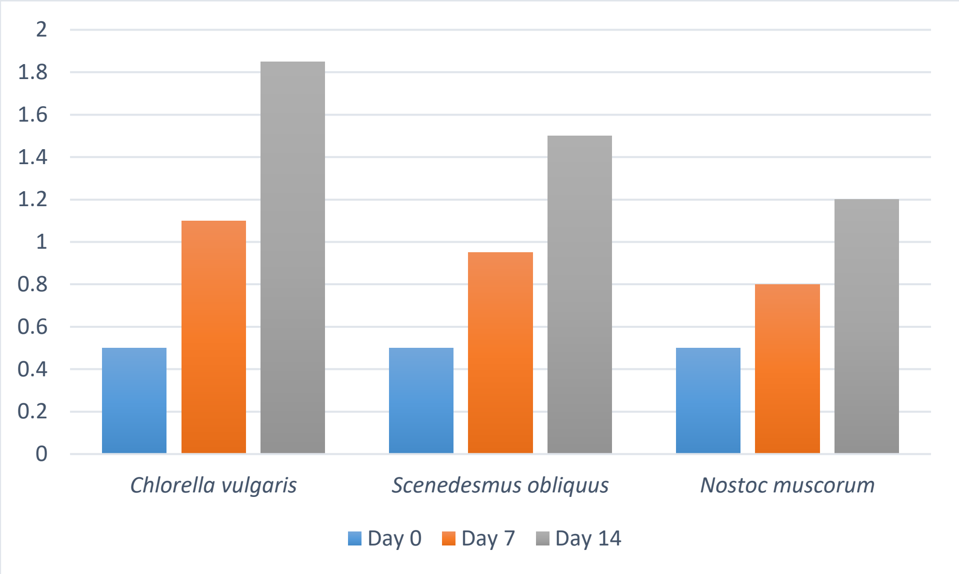

Figure 1: Algal Biomass (g/L dry weight) over 14 Days

3.2 Pollutant Removal Efficiency

The removal efficiency of pollutants (COD, BOD, Total Nitrogen, Total Phosphorus) was highest in reactors inoculated with *Chlorella vulgaris*, followed by *Scenedesmus obliquus* and *Nostoc muscorum.*

Table 1. Pollutant Removal Efficiency (%) After 14 Days

Parameter	Control	*Chlorella vulgaris*	*Scenedesmus obliquus*	*Nostoc muscorum*
COD	10.5%	85.2%	79.4%	68.1%
BOD	12.8%	89.6%	81.0%	72.3%
Total Nitrogen	5.3%	78.5%	71.2%	62.7%
Total phosphorus	7.2%	82.0%	74.5%	57.9%

Among the species tested, *Chlorella vulgaris* demonstrated the highest pollutant removal efficiencies across all parameters. Specifically, it achieved a BOD reduction of 89.6%**,** COD reduction of 85.2%, and total nitrogen of 78.5%. These results highlight *Chlorella's* strong metabolic capacity for nutrient uptake and organic matter degradation. *Scenedesmus obliquus* also performed well, with pollutant reductions ranging from 71.2% to 81.0%**,** while *Nostoc muscorum* exhibited comparatively lower efficiency, particularly for phosphorus (57.9%) and nitrogen (62.7%). This trend aligns with the differences in biomass accumulation observed among the species. The data confirm that algal-based treatment systems can significantly reduce organic load and nutrient concentrations in sewage, with effectiveness varying by species.

3.3 Variation in Dissolved Oxygen and pH

The dynamics of dissolved oxygen (DO) and pH were monitored throughout the 14-day treatment period to assess the photosynthetic activity and metabolic influence of the selected algal species (*Chlorella vulgaris*, *Scenedesmus obliquus*, and *Nostoc muscorum*) on sewage water.

Table 2. DO and pH Variation During the Treatment Period

Algal Species	DO Initial (mg/L)	DO Final (mg/L)	Change in DO (mg/L)	pH Initial	pH Final	Change pH

Chlorella vulgaris	1.2	7.8	+6.6	6.7	8.6	+1.9
Scenedesmus obliquus	1.3	6.9	+5.6	6.8	8.3	+1.5
Nostoc muscorum	1.1	5.6	+4.5	6.6	7.9	+1.3
Control (No Algae)	1.2	1.6	+0.4	6.7	6.9	+0.2

A progressive increase in DO was observed in all algal-treated reactors, with the most pronounced elevation in the *Chlorella vulgaris* treatment group, where DO levels rose from 1.2 mg/L to 7.8 mg/L by day 14. This enhancement is attributed to high photosynthetic oxygen production, which also supports aerobic microbial degradation of organic matter. The DO increase in *Scenedesmus* and *Nostoc* treatments was slightly lower but still substantial, indicating robust photosynthetic performance. In contrast, the control group, lacking photosynthetic biomass, exhibited only a marginal DO rise, likely due to limited natural aeration. Similarly, pH levels increased significantly in algal treatments, especially in the *Chlorella* culture, where it rose from 6.7 to 8.6. This shift toward alkalinity results from CO_2 uptake during photosynthesis, which reduces carbonic acid concentration in the medium.

4. DISCUSSION

The results of the present study demonstrate that microalgae can effectively reduce key pollutants in sewage, including organic matter (as measured by BOD and COD) and nutrients (nitrogen and phosphorus). The present study evaluated the bioremediation efficiency of three microalgal species—*Chlorella vulgaris*, *Scenedesmus obliquus*, and *Nostoc muscorum*—in sewage treatment over a 14 days period. The results demonstrated notable species-specific differences in biomass accumulation, pollutant removal, and physicochemical modifications of the sewage environment, affirming the potential of algal-based systems in decentralised and sustainable wastewater treatment applications.

4.1 Algal Biomass Growth and Adaptation

All three algal species exhibited significant biomass growth in raw sewage, with *C. vulgaris* showing the highest biomass accumulation from 0.50 g/L to 1.85 g/L in 14 days. This indicates not only its robustness in wastewater conditions but also its high adaptability to nutrient-rich environments. The continuous increase in algal biomass across all species confirms their effective acclimatisation to the organic and inorganic load of sewage. These findings corroborate earlier studies by Abdel-Raouf *et al.* (2020) and Zhao *et al.* (2021), who highlighted the capability of *Chlorella* and *Scenedesmus* to flourish in untreated or partially treated wastewater due to their efficient nutrient uptake and tolerance to pollutant stress.

The lower biomass production of *N. muscorum* may be attributed to its slower growth kinetics and sensitivity to high organic loads typically present in municipal sewage (Bhateria & Jain, 2021). Since biomass production is directly associated with nutrient uptake, the observed differences in pollutant removal efficiency may also be partially explained by these variations in growth performance.

4.2 Pollutant Removal Efficiency

Chlorella vulgaris demonstrated the highest removal efficiencies across all tested parameters—COD (85.2%), BOD (89.6%), total nitrogen (78.5%), and total phosphorus (82.0%)—emphasizing its superior metabolic versatility and pollutant assimilation capacity. These findings are consistent with reports by Mohsenpour *et al.* (2021) and Ramanan *et al.* (2020), which described Chlorella's exceptional ability to simultaneously uptake nitrogen, phosphorus, and organic carbon due to its high photosynthetic efficiency and production of extracellular polymeric substances (EPS), which facilitate nutrient adsorption.

Scenedesmus obliquus also achieved significant pollutant reductions (e.g., 81.0% BOD and 74.5% TP), aligning with prior findings by Wang *et al.* (2020) that documented its strong performance in high-rate algal ponds (HRAPs). In contrast, *Nostoc muscorum* exhibited comparatively lower removal efficiency, particularly for phosphorus (57.9%) and nitrogen (62.7%). This may be attributed to its cyanobacterial physiology, which favours slow nutrient uptake and limited adaptability to high nutrient concentrations unless supported by symbiotic bacterial interactions (Singh *et al.*, 2020).

The poor performance of the control reactor (e.g., COD removal of only 10.5%) emphasizes the central role of algae in pollutant abatement. This supports the ecological function of algae as primary producers that not only contribute to oxygenation but also directly assimilate dissolved pollutants for biomass synthesis (El-Sheekh *et al.,* 2022).

4.3 Influence on DO and pH

The dynamics of dissolved oxygen and pH during the treatment period further reflect the metabolic activity of the algae. The C. *vulgaris* reactor showed a substantial increase in DO (from 1.2 mg/L to 7.8 mg/L), indicating intense photosynthetic oxygen production. Elevated DO levels enhance aerobic microbial degradation of organic pollutants, thereby synergising the bioremediation process. This is in agreement with Barros *et al.* (2021), who reported DO levels above 6 mg/L in High Rate Algal Ponds (HRAPs) dominated by *Chlorella*, leading to accelerated organic carbon breakdown.

Similarly, a rise in pH from 6.7 to 8.6 in the *Chlorella* treatment reflects the biological uptake of CO_2 during photosynthesis, which shifts the carbonate equilibrium toward alkalinity. The pH shift also contributes to phosphorus precipitation and ammonia volatilization, thereby aiding in nutrient removal (Kumar *et al.*, 2022). The relatively lower changes in DO and pH in the *Scenedesmus* and *Nostoc* treatments reflect species-specific photosynthetic rates, while the control reactor showed only a marginal increase, demonstrating the minimal role of natural aeration or microbial respiration in the absence of algal biomass.

5. CONCLUSION

This study highlights the remarkable potential of microalgae—*Chlorella vulgaris*, *Scenedesmus obliquus*, and *Nostoc muscorum*—as sustainable agents for the bioremediation of sewage. The findings demonstrate that algae not only thrive in wastewater environments, exhibiting significant biomass growth, but also effectively remove organic pollutants (COD and BOD) and nutrients (nitrogen and phosphorus, while simultaneously enhancing dissolved oxygen levels and stabilizing pH. Among the species tested, *Chlorella vulgaris* consistently outperformed others across all parameters, indicating its superior adaptability, metabolic activity, and biosorption capacity.

Future scale-up and field application of this integrated algal system could revolutionise decentralised sewage treatment, particularly in developing regions, aligning with circular bioeconomy principles and sustainable wastewater reuse strategies.

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