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Use of Algae in Water Treatment Isolated from Some Regions of Baghdad

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ABSTRACT: In this work, wastewater was treated using a locally isolated microalga (Chlorella vulgaris Bejerinck) to reduce pollutant indices. By examining three factors, the effectiveness of Chlorella vulgaris in reducing COD, BOD5, and PO4 concentrations was investigated. At the wastewater treatment plant in Esala al-Sader, wastewater samples were taken from a main station. Three distinct wastewater dilutions—100% sludge, 75% sludge, and 25% sludge—were evaluated with distilled water.. During the early incubation period, During the incubation phase, there was a noticeable decrease in both the chemical and biological oxygen demands (BOD5 and COD). This is attributed to the efficient biodegradation of organic matter and the increased microbial activity driven by algal photosynthesisThe treatment process we carried out during the first seven days was effective and efficient. We observed a strong interaction between the algae and microorganisms, which enhanced oxygen production, nutrient absorption, and the decomposition of organic matter. However, we noted that the concentrations of biological oxygen demand (BOD5) and chemical oxygen demand (COD) and phosphorus increased during the incubation period in the treatment cycles. This was due to the autolysis of cells, the aging of the algal biomass, and the release of organic matter and phosphorus into the wastewater. All of this highlights the need to control the incubation period and maintain the biomass in a stable state to prevent pollution resulting from decomposition. The results also showed that sample C was more stable and performed more efficiently in removing pollutants. This was evident in the decrease in the levels of COD and phosphate in the later stages of the experiment. We also observed the effectiveness of the common alga (Chlorella vulgaris) in treating wastewater and removing organic pollutants and nutrients, especially when the operating environment or conditions are improved. However, the treatment performance is significantly affected by the incubation period, nutrient availability, biomass mobility, and interactions between the algae and microorganisms.. Therefore, this research recommends periodic harvesting of algal biomass, optimizing treatment duration, and minimizing the release of organic matter and nutrients to increase the efficiency of pollutant removal in treated wastewater.

INTRODUCTION

Water resources suitable for direct human use are in short supply due to the world's population and economy growing at an accelerated rate.. Consequently, global attention will inevitably shift towards water remediation as the primary focus (Ajay Kumar et al*.,* 2024).Water serves as a crucial resource and a key raw material in various sectors such as electronics pharmaceuticals, petrochemicals, food and beverage, agrochemicals, gas and oil, as well as for domestic usage (Wang et al.,2023).

Because of the numerous contaminants present, there is rising worry over the direct discharge of polluted water from these operations, which poses major threats to the ecosystem. A variety of substances can be found in wastewater (WW), some of which, at certain amounts, are extremely harmful to living things. Additionally, Both nutrients that are inorganic or organic are released into the atmosphere in considerable quantities., resulting in increased levels of both biological and chemical oxygen demand (BOD). Eutrophication of water bodies resulting from excessive phosphorus (P) and nitrogen (N) loadings leads to environmental issues such as the production of undesirable emissions and solid wastes into the atmosphere. (Chen, et al.,2020;. Wang, et al.,2022). It alike fosters the proliferation of harmful microorganisms that jeopardize various aquatic organisms and deteriorate drinking water's quality, resulting in common health issues in the vicinity of the discharge site (Hena et al., 2021). Wastewater poses a significant global challenge due to its harmful impacts on natural resources. It contains substantial amounts of both inorganic and biological materials, such as carbon content volatile components and alkaline salts , that interact with the environment's normal processes, leading to pollution. Consequently, It is crucial to treat sewage before releasing it into ecosystems (Abdel-Raouf, *et al.,* 2012 To diminish the levels of BOD and COD in effluents and enhance their quality, biological treatment employs the metabolic processes of microorganisms to decompose and convert wastewater pollutants into biomass and various gases (N2, CH4, CO2, and SO2). (Dalvi et al.,2021). Biodegradation procedures using a variety of microorganisms, including fungus, yeast ,bacteria, and microalgae, are included in biological therapies (Ali et al., 2021; Danso, et al.,2022). Because they are naturally occurring and contain useful compounds, microalgae are especially appealing because they can form or produce complex materials that are thought to be highly active against various pathogenic bacteria and fungi. In terms of the potential for breakdown yield, biological treatments are intended to create a system that facilitates the treatment of wastewater (WW) (Khan et.al 2018). Numerous chemical elements, including phenols, fatty acids, indoles, terpenes, acetogenins, and certain volatile halogenated hydrocarbons, have demonstrated action against pathogenic bacteria when isolated from microalgae (Sukla et.al 2019) . The goal of biological treatments is to provide a system that makes it easier to treat waste that has been disposed of based on the potential for breakdown yield. Because they use natural processes, biological wastewater treatment (WWT) seems straightforward at first, but it is actually complicated and still poorly understood at the nexus of biology and biochemistry (Pereliaeva E.V., *et al.,*2022). However, a number of [micro algae species] have long demonstrated their ability to thrive in wastewater environments, absorbing all of the available phosphorus [P], inorganic nitrogen [N], and organic carbon [C] from the wastes (Guldhe et al.,2017 ; Chen et al., 2018). Light is a fundamental environmental variable that critically influences the growth, metabolism, andphotosynthetic efficiency of microalgae, particularly in the context of large-scale biomass production Chowdury KH et al., 2020). Optimizing light conditions is essential for enhancing microalgal productivity, given that Chlorophyllaand bact as critical pigments involved in light harvesting and charge separation, which in turn dictate the overall photosynthetic performance and biomass accumulation(Koh HG, et al.,2019).

Microalgae with unique qualities and features have been used in the past to cure a variety of toxicants emitted from the home, industrial, and agricultural sectors. At the moment, several microalgae have been documented to detoxify domestic WW, including *Scenedesmus obliqus, Scenedesmus abundans, Scenedesmus sp., Chlorella sp., Chlorella sorokiniana, Chlorella vulgaris and Chlorella variabilis*(Tran et al.,2021;SundarRajan et al., 2020;de Souza et al., 2019).

As per earlier research, ordinary open ponds or even sophisticated High-quality algal pond are the most effective and productive techniques for cultivating microalgae from wastewater (Chen et al.,2018; Guldhe, et al.,2017). Wastewaters come in a variety of forms, including piggery , dairy, , urban, and poultry wastewaters. and also there are a variety of industrial effluents: from oil mill , chemical, paper mill, , sugar mill effluents etc.., (Plohn, et al.,2021). Recently, a great many of bioremediation oriented studies have been documenting attached growth of microalgae and their combinations with bacteria for effective treatment of the wastewaters using various closed cultivation systems such as photobioreactors (Petrini, et al 2020;. Rosli, et al.,2020)which appear to be quite promising in terms of efficiency and convenience of use in closed systems.. However, in the case of an industrial setup or a commercial setup, many of the stakeholders prefer the open cultivation methods purely because it is easier to maintain and economical than the other process which is closed cultivation. In addition, it is not appropriate to cultivate the attached growth in a large scale cultivation process. It would be very unstable since they would not be able to handle the wastewaters of these volumes, the attachment of the micro algal and their distribution would be very uneven and disproportionate, clogging and the bad odour. However, the suspended growth of the micro algal in the open cultivation would be manageable at quadratic volumes as well(Paddock 2019; Ho 2020). Microalgae bioremediation of municipal and sewage wastewaters has been the focus for decades. Wastewater purification, processed sewage recycling, municipal waste and the production of microalgae biomass for green energy have all been the subject of numerous studies (Robles 2020). One of the microalgae species that has been studied the most for wastewater treatment research is Chlorella vulgaris (Otondo et al .,2018). Because of its small cells, exceptional chlorophyll content, potential for rapid growth, and high tolerance to changing environmental conditions, C. vulgaris—which is typically found in freshwaters—is the best microalga for treating wastewater and producing bioenergy at the same time (Klassen, et al.,2017). This study aims to test the effectiveness of an algae in purifying the water isolated from the station from contaminants. It also aims to determine which of the three contaminants produces the best results.

MATERIALS AND METHODS

Sample collection : Samples of sewage were collected at the nearby Esala al-Sader station's preliminary sedimentation, and the characteristics are displayed in Table (1). Samples autoclaved for 15 minutes at 121°C to destroy any microorganisms in the wastewater to demonstrate how *Chlorella vulgaris* impacts the treatment of wastewater.

Table 1 (1). The wastewater sample's properties (pH = 7.43)

samples	concentration (mg/L)	Parameters Average
Sample a 100%	100%	BOD_5, COD , PO_4
Sample b 75%	75%	BOD_5, COD , PO_4
Sample c 50%	50%	BOD_5, COD , PO_4

Pollutants Analysis: Wastewater samples were collected from Esala al-Sader station and analyzed for BOD_5, COD and PO_4 2- according to Anon. (2005) standardized techniques and thereafter placed in certain ponds for the development of Chlorella vulgaris.

Artificially synthesized medium: *Chlorella vulgaris* was gathered in the Baghdad, Iraq, Tigris River Al-Rasheed region. In this investigation, streaking on plate agar methods were employed for purification and algae isolation. Chu-10 algae culture medium is made from mineral nutrient salts that are separated into two categories: micronutrients and macronutrients. After adding 10 milliliters of the separated culture to a flask holding 100 milliliters of Chu-10 medium, the algal culture sample was cultured for 14 days at 30 degrees Celsius, with a photoperiod of light :dark 14:10, and then transferred to 1000 milliliters of sterilized and filtered wastewater sample(Karin, Larsdotter2006).

Species of *Chlorella vulgaris* have been extracted by centrifugation. After centrifuging the culture broth for 10 minutes at 500–600 rpm, a diaphanous liquid was obtained, and its concentration was determined using a turbidity meter (780 NTU) by diluting the liquid with water (Dewayanto et al., 2023).

Laboratory scale biological treatment design: The samples were examined in a lab using the standard procedure for water and waste water analysis (APHA, 1995). The following 3 water components were studied: $PO_4^{\ 2-}$, Chemical oxygen demand and Biochemical Oxygen Demand oncentration and a chlorella treatment unit was built to track BOD_5, COD, and reductions at each stage of the treatment plan.

Results

BOD, COD, and PO_4 removal

The conventional methods outlined by were used to measure the wastewater for PO_4, Chemical Oxygen Demand (referred to as COD), and Biochemical Oxygen Demand (referred to as BOD). [American Public Health Association2005].

Table 2: Measurements of BOD ,COD and PO_4

samples	Sample a 1 day	Sample b 1 day	Sample c 1 day
BOD (mg/l)	66	96	66
COD (mg/l)	148	84	59
PO_4(mg/l)	7.382	4.169	8.032
7 day			
BOD (mg/l)	36	12	12
COD (mg/l)	73	85	57
PO_4(mg/l)	9.830	16.485	9.486
14 day			
BOD (mg/l)	144	120	162
COD (mg/l)	113	81	46
PO4(mg/l)	12.316	12.087	7.611

Removal of BOD

As shown in Figure 3, the biological oxygen demand (BOD_5) values exhibited significant variations throughout the experiment across all studied samples. On the first day, we observed that the biological oxygen demand (BOD_5) concentrations were 66 mg/L for sample A, 96 mg/L for sample B, and 66 mg/L for sample C. After seven days of incubation, we observed a decrease in BOD values to 36 mg/L for sample A and 12 mg/L for samples B and C. However, on the fourteenth day, there was a significant increase in BOD concentrations, reaching 144 mg/L for sample A, 120 mg/L for sample B, and 162 mg/L for sample C.

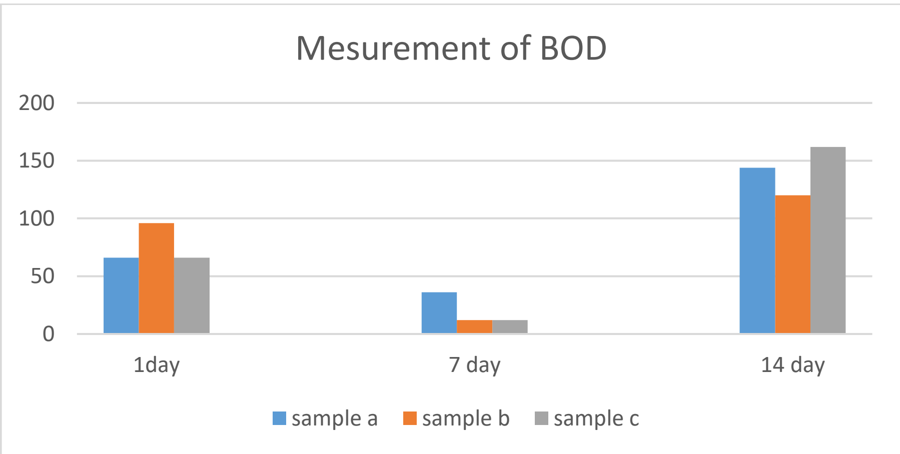

Figure 1 (1). BOD was temporarily removed for each of the three samples. (A= Wastewater at 100%). B is 75% wastewater. C=25% of the wastewater

Removal of COD

Figure 3 illustrates how the chemical oxygen demand (COD) values exhibited significant variation among the studied samples throughout the experiment. On the first day of the experiment,COD concentrations were 148 mg/L, 84 mg/L, and 59 mg/L for samples A, B, and C, respectively. After 7 days of incubation, COD values decreased to 73 mg/L in sample A and 57 mg/L in sample C, while sample B experienced a slight increase to 85 mg/L. By day 14, COD concentrations had changed more significantly, reaching 113 mg/L in sample A, 81 mg/L in sample B, and 46 mg/L in sample C.

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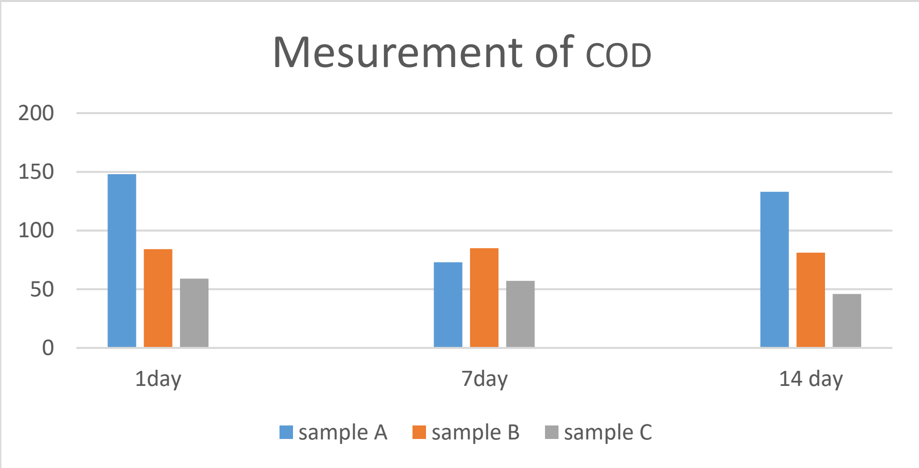

Figure (2). temporary elimination of COD in each of the three samples. (A= Wastewater at 100%). B is 75% wastewater. C=25% of the wastewater

PO_4 Removal

Figure 3 shows that phosphate concentrations fluctuated significantly during the experiment. On the first day, the concentrations were 7.382 mg/L, 4.169 mg/L, and 8.032 mg/L for samples A, B, and C, respectively. After seven days of incubation, these values changed, reaching 9.830 mg/L in sample A, 16.485 mg/L in sample B, and 9.486 mg/L in sample C. On the fourteenth day, the concentrations in samples A and B were high, at 12.316 mg/L and 12.087 mg/L, respectively, while the concentration in sample C decreased to 7.611 mg/L.

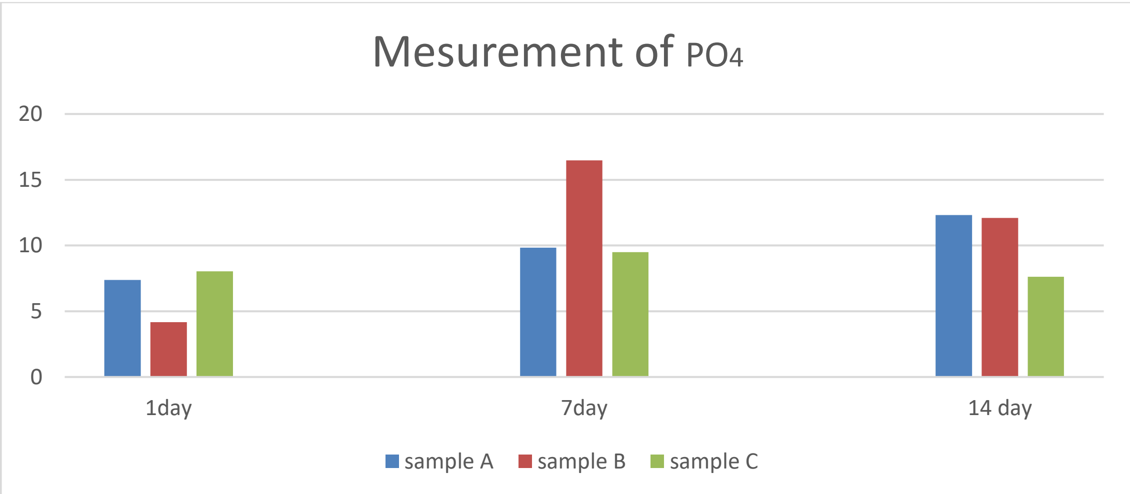

Fig. (3). temporary elimination of PO_4 in three samples. (Where A = 37.5 mg/L, B = 75% mg/L, and C = 25% mg/L of PO_4)

DISCUSSION

The initial decrease in BOD during the first seven days can be attributed to the biological decomposition of organic matter by microorganisms and algae present in the wastewater. During this time, bacterial respiration and metabolism were carried out using dissolved organic compounds, which led to a decrease in oxygen demand. It was also observed that microalgae have an indirect effect on wastewater purification through oxygen production during photosynthesis, which led to improved aerobic microbial decomposition as the BOD values decreased. Oxygen levels improved wastewater quality and increased the efficiency of the algal treatment system during the early incubation phase. This was demonstrated by the results of Mohammed (Dr. Alaa Karim et al., 2015), where the study showed that Chlorella vulgaris* reduced the biological oxygen demand (BOD_5) in water samples isolated from the Tigris River. The increase in BOD on the fourteenth day of our experiment was due to the accumulation of organic matter that could decompose within the treatment system, as well as the decomposition of microbial cells, algal biomass, and the presence of organic compounds in the wastewater.

Prolonged incubation periods are accompanied by physiological stress and nutrient depletion, leading to aging and decomposition of algal cells. This results in an increase in carbohydrates, proteins, amino acids, and dissolved organic matter, which strengthens bacterial activity and increases oxygen consumption. Therefore, we observed an increase in BOD values. Our experiment showed that sample C recorded the highest values at 162 mg/L, indicating decomposition of algal biomass and a greater accumulation of organic matter. Samples A and B recorded higher BOD values on the fourteenth day of the experiment because the organic matter was broken down more by the algae.. This interpretation aligns with the findings of Shen et al. (2010), who reported that prolonged microalgae cultivation can lead to the release of dissolved organic compounds following biomass decomposition. Furthermore, Mohammadi Khalafabadam et al. (2016) demonstrated that algal biomass decomposition significantly contributes to increased organic load and oxygen demand during later stages of wastewater treatment processes.

Overall, the results during the early treatment phase show that the algal and microbial treatment system effectively reduced biodegradable organic matter. However, the extended incubation period promoted the re-release of organic compounds due to biomass decomposition, resulting in increased BOD concentrations during the later phase of the experiment

The initial decrease in COD during the first seven days within the wastewater treatment system is attributed to the efficient decomposition and the elimination of organic contaminants by algae and microbial activity The sources of metabolic processes and biomass production of carbon and energy are dissolved and suspended organic compounds that can be utilized by microorganisms and algae. This reduces the amount of chemically oxidized organic matter in wastewater. A decrease in the chemical oxygen demand (COD) improves wastewater quality and increases treatment efficiency. This was demonstrated in a study by Wang et al. (2010), who showed that algae such as Chlorophytum significantly reduce COD values in wastewater because they absorb nutrients and biologically decompose organic pollutants. Furthermore, the interaction between algae and bacteria increases wastewater treatment efficiency due to improved organic matter decomposition. However, the COD value of the sample increased on the fourteenth day of our experiment due to the release of organic matter resulting from the decomposition of algal biomass into the surrounding environment. This release is attributed to the prolonged incubation period, leading to cell aging and autolysis, and consequently, the diffusion of internal organic compounds such as lipids, carbohydrates, and proteins, in addition to external polymers. These compounds led to an increase in the COD value due to the increased concentration of chemically oxidized substances. This explanation was supported by the results of the study by Shen et al. (2010), who showed that long-term algal cultivation leads to a wider dispersal of organic matter due to biomass decomposition. Similarly, the study by Mohammadi Khalafabad et al. (2016) demonstrated that the accumulation of dissolved organic carbon and organic matter resulting from the decomposition of algal biomass increases COD values during subsequent treatment stages. However, Sample C showed a decrease in COD values in the experiment, with its lowest value being 46 mg/L on the fourteenth day of the experiment. This result indicates that the system was efficient and stable in removing organic matter. This suggests that the more stable algal activity, improved microbial adaptation, or biomass decomposition led to improved performance of Sample C compared to treatments in other studies. Chlorella algae were cultivated in wastewater from a textile industrial area, and the efficiency of COD removal was shown to increase from 38.3% to According to the study by Lim et al. (45), the COD (carbon dioxide) in the studied wastewater decreased by more than half due to the presence of Chlorella algae. In sample B, the COD values were relatively stable due to a balance between organic decomposition and the diffusion of organic compounds from algae or other microorganisms. Therefore, we can conclude that fluctuations in treatment systems under prolonged incubation conditions are due to the significant ability of algae to reduce COD and remove organic pollutants from wastewater. Furthermore, the treatment system functions efficiently when the incubation period and algal biomass are stable, and the decomposition of both organic matter and biomass is balanced.

The increase in phosphate concentrations is attributed to the decomposition of algae and organic matter, a consequence of the long incubation period. During this time, algal cells decompose and may undergo aging, leading to the diffusion of phosphorus compounds into the environment. This explains the increased concentration of dissolved phosphates, which exceeded the phosphorus uptake rates during the incubation period. Initially, phosphorus was the primary nutrient for the growth of algal and microorganism cells and the synthesis of adenosine triphosphate (ATP). It is also essential for metabolic activity. However, increasing the incubation period led to its depletion and the decomposition of the biomass, thus reducing adsorption efficiency and increasing phosphorus diffusion in wastewater. This explains the increased phosphate levels in sample B on days 7 and 14. Furthermore, a study by Lavrinovich et al. (2021) demonstrated that the common alga Chlorella effectively removes phosphate during its growth period. However, the kinetic states of phosphorus are affected by several factors, including nutrient balance, physiological stress, and biomass status. Additionally, a study by Larsdottir et al. (2007) showed that photosynthesis and its associated biosorption and precipitation processes are crucial for phosphorus removal when using algae in wastewater treatment. Moreover, a study by Shen et al. (2010) indicated that increasing the incubation period leads to increased phosphate concentration because the decomposition of algal mass and cells in general results in the diffusion of previously absorbed phosphorus into the surrounding environment. Wang et al. (2015) demonstrated that increased phosphate concentrations result from autolysis and decomposition of algal biomass due to intracellular mineralization of phosphorus compounds. Interestingly, in our experiment, phosphate concentrations were low for sample C, particularly on day 14 compared to day 7, because the remaining organisms continued to absorb phosphorus or there was partial phosphorus precipitation.. This behavior may suggest better stability and higher nutrient utilization efficiency in this treatment compared to samples (A) and (B).

Overall, the results demonstrate that phosphate dynamics in algae-based wastewater treatment systems are governed by a balance between phosphorus uptake during active microbial growth and its release during biomass decomposition and cellular degradation.

These findings agree with those of McGriff [Monica, and Rothermel,2011), They found that adding algae to wastewater treatment might increase the effectiveness of COD removal to 70–80%. According to Azeez (Azeez. 2010), chlorella vulgaris may eliminate COD with an efficiency of up to 83%. Using citric acid with diluted ethanol synthesis from industry wastewater, Hammouda (Hammouda,et al 1995) was able to boost the micro algae Chlorella vulgaris's 61% COD removal efficiency. Furthermore, Zhang's research(Zhang et al .,2008) demonstrates that inorganic nutrients may be effectively extracted from residential effluents

CONCLUSION

The results of this research showed that the use of common algae (Chlorella vulgaris) as a type of microalgae played an important role in increasing and improving the quality of wastewater by reducing the concentrations of nutrients and organic pollutants, especially during the primary treatment period. There was a clear decrease in both the chemical oxygen demand (COD) and the biological oxygen demand (BOD) in most treatments. This led to increased efficiency in the biodegradation of organic matter within the algal-microbial system. The interaction between algae and microorganisms was strengthened by the increased oxygen presence and enhanced decomposition of compounds, especially biogenic ones, resulting in improved treatment efficiency. The results also showed that the incubation period and wastewater ccentration affected treatment efficiency. Sample C exhibited the best stability and a greater decrease in COD and phosphate levels during the later stages of incubation, as this sample contained the lowest wastewater concentration. Conversely, increased COD, BOD, and phosphate concentrations were observed in some treatments due to the extended incubation period. This increase resulted from increased decomposition of algal biomass, autolysis of cells, and the release of organic matter and phosphorus from within the cells into the wastewater. It should be noted that although treatment using algae was effective in removing pollutants and environmentally friendly, the efficiency of the treatment system was critically affected. Treatment duration and biomass management are crucial, as long incubation periods lead to reduced treatment performance due to the accumulation of dissolved organic matter resulting from biomass decomposition. Overall, the study indicates that using microalgae for wastewater treatment represents a promising and sustainable biological approach to reducing organic and nutrient pollution in wastewater. To increase treatment efficiency and minimize secondary pollution, operating conditions must be improved, particularly incubation duration and periodic biomass harvesting. This study demonstrates that the use of microalgae, in particular, plays a significant role in improving wastewater quality by reducing concentrations of organic pollutants and nutrients. During the primary treatment phase, a noticeable reduction in these concentrations was observed in most treatments.

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Contributions of Authors

N.R.G.: The article's principal investigator, fieldwork, conceptualization, writing, and draft review

S.N.L.; Draft review; data gathering methods in the lab.

M.S.A.: Essay proposal, draft evaluation, and feature definition.

R.M.I.: Article suggestion, drafts review.

Conflicts of interest

There is no conflict of interest disclosed by the writers.

Ethical approval

This report complies with all fieldwork ethical norms.

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