

Bioremediation of Wastewater – Effect of Algae in Bioremediation of Nitrate and Phosphate Content in Wastewater

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ABSTRACT: The progressive adoption of urban, industrial lifestyle deteriorated the quality of freshwater reservoirs and caused a huge challenge in the world in maintaining the demand and quality of water. Industrial, Agricultural and Domestic processes contaminate water with chemical and biological pollutants that cannot be released into the environment until treated to have less than specified limit of pollutants. Traditional Wastewater treatment plants are effective in remediating suspended solids and many harmful elements, but it is not effective in nitrate and phosphate content removal, causing these toxic elements discharged to the environment. This study aims to determine the effectivity of algae to bioremediate the nitrate and phosphate content from wastewater. Four different specimens of Algae were tested with wastewater in bioreactor setup for ten days over five trials, and the nitrate and phosphate contents were measured every day using HACH-DR-890 colorimeter to see how effectively the specimen remediate the nitrate and phosphate contents. The collected data analysis shows all the four specimens were able to effectively remediate 93-96% of nitrate and 73-86% of phosphate content. *Chlamydomonas reinhardtii* (95.66%) is most effective in nitrate remediation followed by *Chlorella vulgaris* (94.98%), *Arthrospira platensis* (94.57%) and *Scenedesmus quadricauda* (93.97%). In the phosphate remediation, *Scenedesmus quadricauda* (85.22%) is most effective followed by *Chlorella vulgaris* (82.46%) *Chlamydomonas reinhardtii* (78.12%) and *Arthrospira platensis* (73.36%). This project concludes that bioremediation using Algae is a viable technology in treating wastewater for nitrate and phosphate remediation in a natural and sustainable way in comparison to conventional treatment processes.

KEYWORDS: Environment pollution; Water treatment; Wastewater; Bioremediation; Bioreactor; Algae; Nitrate; Phosphate.

Introduction. Maintaining the quality of water is a huge challenge in the world. The growing population and progressive adoption of an urban, industrial lifestyle has deteriorated the quality of freshwater reservoirs around the world. Remediation of wastewater that is generated from domestic sewage, industrial and agricultural discharges have become a large concern for the developed and developing countries. Wastewater contains physical, chemical and biological pollutants, which include many harmful substances that cannot be released back into the environment until the wastewater is treated.

Traditional wastewater treatment plants are effective in the remediation of suspended solids and the physical and chemical pollutants, but toxic heavy metals (i.e. Cadmium (Cd), Chromium (Cr), Arsenic (As), Mercury (Hg), etc.) and nitrate and phosphate content removal processes are still not very effective and sustainable, causing these toxic elements to be discharged into ground water. Bioremediation is an effective and eco-friendly method in removing those elements. It uses naturally occurring microorganisms (yeast, fungi, algae, bacteria) to break down, or degrade, hazardous substances into less toxic or nontoxic substances. The objective of this study is to determine the effectivity of algae to bioremediate the Nitrate and Phosphate content from wastewater. Algae absorbs nitrogen in form of nitrate and phosphorus in form of phosphate from the wastewater and clean it by separating the nutrients as biomass. This algal biomass can also be used later as an energy source, turned into biofuel, which is renewable and has a small carbon footprint.

Materials and Methods. Collection of wastewater and algae specimens. Due to rules and regulations on handling wastewater, the wastewater was simulated by mixing fertilizer Ammonium Nitrate and Diammonium Phosphate with distilled water and filtering using Whatman No.1 filter paper to remove suspended solid particles.

There are many different species of algae available, such as *Chlorella*, *Ankistrodesmus*, *Scenedesmus*, *Euglena*, *Chlamydomonas*, *Arthrospira*, *Oscillatoria*, *Micractinium*, etc. For this study, the algae specimen collected were readily available, easy to grow/culture, does not have adverse effect on environment and human being and are known to be pollution tolerant and having a high absorption capacity, and thus effective in remediation of wastewater. Based on the abovementioned criteria, the algae specimens selected were a. *Chlamydomonas reinhardtii* b. *Arthrospira platensis* (*Spirulina*) c. *Scenedesmus quadricauda* and d. *Chlorella vulgaris*.

Experiment setup. To allow a healthy environment for the algae to grow and bioremediate nitrates and phosphates, A bioreactor system was designed using 500 ml Erlenmeyer flasks connected with an air pump to provide a source of carbon dioxide in the flasks. A fluorescent light was kept on for a fast photosynthesis of green algae. The bioreactor was used as an experimental prototype of the Algae based Wastewater Treatment System (AWTS). The filtered and untreated wastewater was used as control solution. The algae specimen solutions were made by adding 4 ml of each algae stream separately into 200 ml of filtered wastewater in bioreactor flasks. The test was con

ducted under controlled conditions (temperature $72 \pm 2^\circ\text{F}$) for a total duration of ten days, and also repeated in five different trials using the same control and specimen solutions. Using a portable Hach DR-890 colorimeter, measurements of the nitrate and phosphate concentration were taken every day for a total duration of ten days.

The nitrate concentration of the solution was measured by cadmium reduction method (Hach Method 8039). In this method, the cadmium metal reduces nitrates present in the sample to nitrite. This nitrite forms an amber-colored product by reacting in acidic medium with Sulfanilic acid. This amber color indicates the presence of nitrate and intensity is converted to (N-NO^3) mg/L or ppm.

Similarly, the phosphate concentration of the solution was measured by the molybdate-ascorbic acid method (Hach Method 8048). The orthophosphate present in the sample reacts with molybdate in the reagent in the acid medium to produce a phospho-molybdate complex. Ascorbic acid reduces this complex and forms a blue color. This blue color indicates the presence of phosphate and the intensity is converted to (P-PO^4) mg/L or ppm.

Results and Discussion. The purpose of this experiment was to find out how effectively different specimen of algae can bioremediate the Nitrate and Phosphate content from wastewater. Based on the collected data analysis, it is observed that all the four types of algae specimen were able to reduce Nitrate concentration from wastewater. The initial concentration of nitrate in the wastewater sample ranged from 178.25 to 188.50 mg/L (Mean: 183.28 mg/L). The 4 algal specimens were able to reduce it to the following: Chlamydomonas reinhardtii (7.96 mg/L), Arthrospira platensis (9.96 mg/L), Scenedesmus quadricauda (11.06 mg/L) and Chlorella vulgaris (9.20 mg/L). (Table 1).

Table 1. Nitrate concentration over 10 days (Average of 5 trials).

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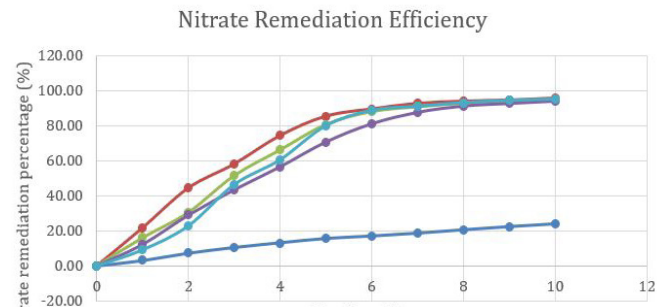
Days	Control Solution	Sp-1 (C. reinhardtii)	Sp-2 (A. platensis)	Sp-3 (S. quadricauda)	Sp-4 (C. vulgaris)
0	183.28	183.28	183.28	183.28	183.28
1	177.79	143.62	153.90	160.96	166.48
2	170.20	101.87	127.38	130.30	141.55
3	164.33	76.84	88.97	103.83	98.41
4	159.48	47.14	62.13	79.77	72.56
5	155.05	26.83	35.45	53.79	36.99
6	152.53	19.32	21.96	34.74	20.91
7	149.61	13.49	16.94	22.77	16.69
8	146.13	11.19	13.27	16.21	12.82
9	142.90	10.03	11.93	13.51	10.49
10	140.05	7.96	9.96	11.06	9.20

$$Et = \frac{(Co - Ci)}{Co} * 100$$

where C_0 and C_i were the concentrations of contaminant at the start of experiment (day 0) and day i , respectively.

The nitrate concentration of the solution was measured by cadmium reduction method (Hach Method 8039). In this method, the cadmium metal reduces nitrates present in the sample to nitrite. This nitrite forms an amber-colored product by reacting in acidic medium with Sulfanilic acid. This amber color indicates the presence of nitrate and intensity is converted to (N-NO^3) mg/L or ppm.

Figure 1. Nitrate remediation efficiency (%) of the algae specimens.



Hence it can be concluded that the Chlamydomonas reinhardtii specimen is able to achieve the most nitrate removal with 95.66%, and all the specimens were able to remediate nitrate to the range of 93-96%. The EPA prescribed limit for nitrate concentration in water is maximum 10 mg/L. Three of the four algal specimens were able to bring the nitrate concentration to that level over a period of ten days.

For the Phosphate remediation, all four of the algal species efficiently removed phosphorus from the wastewater. The phosphate concentration remediation from the wastewater by each of the four algae specimen is presented in Table 2. Prior to treatment with algae, the initial phosphorus content was on average 24.77 mg/L. After the ten days of experimentation, the phosphorus concentration was reduced to a maximum of 3.66 mg/L.

Table 2. Phosphate concentration over 10 days (Average of 5 trials).trials).

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Days	Control Solution	Sp-1 (c. reinhardtii)	Sp-2 (A. platensis)	Sp-3 (S. quadricauda)	Sp-4 (C. vulgaris)
0	24.77	24.77	24.77	24.77	24.77
1	24.08	20.55	22.32	21.72	20.74
2	23.29	18.44	19.80	19.11	17.31
3	22.39	16.31	16.77	17.26	14.70
4	21.83	12.08	14.71	14.29	12.05
5	21.32	10.55	12.48	11.63	10.73
6	20.23	9.02	10.46	9.86	9.10
7	19.48	7.87	8.46	7.90	7.17
8	18.93	6.86	7.71	6.47	5.94
9	18.23	6.18	7.03	5.21	4.95
10	17.14	5.42	6.60	3.66	4.34

Based on the formula

$$PE = \frac{(Q_0 - Q_i)}{Q_0} \times 100$$

where Q_0 and Q_i were the concentrations of contaminant at the start of experiment (day 0) and day i respectively, the phosphate remediation efficiency by *Scenedesmus quadricauda* was more than other species during the experimental period. 85.22% of the initial phosphate content was consumed by *Scenedesmus quadricauda*. It was then followed by *Chlorella vulgaris* (82.46%) *Chlamydomonas reinhardtii* (78.12%) and *Arthrospira platensis* (73.36%). (Figure 2) Regardless of the algal specie, by the end of the ten days period, a range of 73–85 % of the initial wastewater phosphate content was removed by the algal species in each of the reactors.

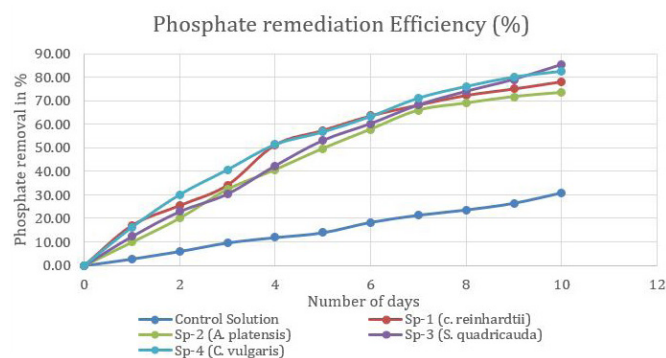


Figure 2. Phosphate remediation efficiency (%) of the algae specimens.

Conclusion. Based on the observations and test results mentioned above, it is concluded that all the algae specimens would be able to remediate nitrate and phosphate content from wastewater. For the Nitrate removal, *Chlamydomonas reinhardtii* is proven to be most effective, remediating upto 96% of the Nitrate concentration and making the water allowable to be discharged to environment again. For the Phosphate removal, while all the four specimens were effective, *Scenedesmus quadricauda* is proven to be the most effective algae specimen in the phosphate concentration remediation from wastewater, remediating upto 85.22% of the Phosphate concentration and making the water allowable to be discharged to environment again. Therefore, this experiment concludes that bioremediation of wastewater using algae provides an effective and environmentally acceptable option for wastewater treatment.

This study was focused on nitrate and phosphate removal aspect of bioremediation of wastewater. There are many other characteristics of wastewater treatment such as BOD and COD remediation, Removal of toxic Heavy metals etc. which can be studied further as future goals of this project.

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