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# Nutrient Removal From Sewage – An Experimental Study At Laboratory Scale Using Microalgae

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**Abstract:** Microalgae have the potential to utilize the nutrients in the wastewater for its growth and serve as a primary feedstock for the production of value added products. A feasibility study has been conducted to determine the capability of the microalgae (*Chlorella sp*) to grow in sewage without sterilization in a laboratory scale batch process under the continuous illumination of light. In the present investigation, the algal growth and nutrients removal in the sewage has been studied and the experimental results revealed the growth period of microalgae as 9 days. The nutrients present in the sewage after algal growth was compared with the nutrients in the initial stage. The percentage removal of nutrients after the end of batch process is Phosphate-90.7%, Total solids -86%, Nitrates-Nitrogen-60%, Ammonia-Nitrogen-58.8%, Total dissolved solids -90%, and Total suspended solids -50%, respectively.

Keywords: Microalgae, Chlorella sp, Batch process, Nutrient removal.

# Introduction

Microalgae play the significant role in meeting the demand of energy and also serve as the primary feedstock for sustainable products. Eutrophication is the due to presence of nutrient level above the limit in marine and surface water due to human activities viz without proper treatment of waste water generated and mixed with surface water<sup>1</sup>. Microalgae have received a major attention as a source of alternate energy, to make it economical the process should undergo optimization in large scale which will meet the demand. An experimental study carried out to the compare the lipid productivity of microalgae in the stationery phase and exponential phase which consumes the domestic wastewater as a source of food<sup>2</sup>. Wastewater which is produced from various industries can also be a source of feed for the algae to grow and hence, play a dual role i.e., nutrient removal and biofuel production.

The current trends in the microalgal energy from wastewater has been reviewed and discussed by the previous researcher<sup>3</sup>. Also, a comparison was made on algal growth in the heterotrophic and autotrophic medium and it has been revealed that algae are well adapted to heterotrophic source and the biomass, lipid content is comparatively high in the heterotrophic medium<sup>4,5</sup>. The effluent from the secondary treatment unit of domestic wastewater treatment plant is used for large scale cultivation of microalgae for biofuel production<sup>6</sup>. Production of high microalgal biomass for high fuel production from wastewater and observed the inhibition in the growth of microalgae at the initial stage in the wastewater due to the presence of high nutrients<sup>7</sup> and, the other factors

that may inhibit the growth of microalgae are the bacteria and protozoa present in the municipal wastewater and also in their preliminary experiments the culture with raw effluent had a negative impact on the growth of microalgae therefore UV radiation & filtration treated sewage will help in betterment of microalgae growth<sup>3,6</sup>. The best suitable method for cultivation of algae in the wastewater is open pond way reactor for biofuel production<sup>8</sup>. The Bioremediation using microalgae has been considered for the removal of nutrients from the wastewater which can be reused to meet the demand of water and also to solve the issue of eutrophication. Recent studies reported that microalgae cultivation in artificial wastewater is found to be effective for nutrient removal<sup>9,10,11</sup>. However, when compared with raw sewage, artificial medium will not have toxin, biotic components and other solid materials<sup>3</sup>. Also the cultivation of *chlorella sp*. in artificial wastewater medium, under dark and light cycle revealed that nutrient removal efficiency is low<sup>11</sup>. With this in view, studies have been conducted to evaluate the feasibility of growth of microalgae (*Chlorella sp*) in domestic sewage without sterilization and under continuous illumination of light in a laboratory scale batch process observe the cell growth and its capability to remove the nutrients.

# **Materials And Methods**

### **Cultivation Of Microalgae**

The microalgae isolated and was identified as *Chlorella sp* under microscope which is shown in the Fig 1 and Fig 2, respectively .The microalgae was then cultivated in Modified Bolds Basal Medium<sup>12</sup> under sterile conditions. The stock solution was prepared for all the constituents in media with the following composition for 100ml:  $K_2$ HPO<sub>4</sub> 0.4g, CaCl<sub>2</sub>.2H2O 0.36g, MgSO<sub>4</sub>.7H<sub>2</sub>O 0.75g, NaNO<sub>3</sub> 15g, citric acid 0.06g, Na<sub>2</sub>EDTA.2H<sub>2</sub>O 0.01g, Sodium Carbonate 0.2g, Ammonium ferric citrate 0.06g, and A<sub>5</sub> trace solution(g/l) components as H<sub>3</sub>BO<sub>3</sub> 2.86, MnCl<sub>2</sub> 1.81, ZnSO<sub>4</sub>.7H<sub>2</sub>O 0.222, Na<sub>2</sub>MoO<sub>4</sub>.2H<sub>2</sub>O 0.390, CuSO<sub>4</sub>.5H<sub>2</sub>O 0.079, Co(NO<sub>3</sub>)<sub>2</sub>.6H<sub>2</sub>O 0.0494. From the stock the media has been prepared for 500ml with 5ml from each constituent and 0.5 ml of A<sub>5</sub> trace solution.

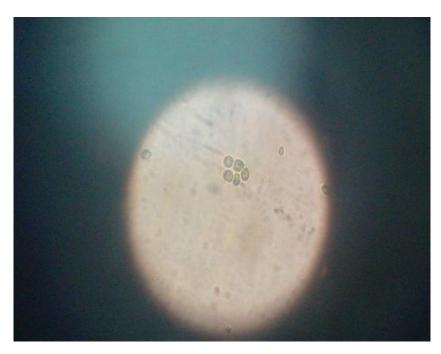
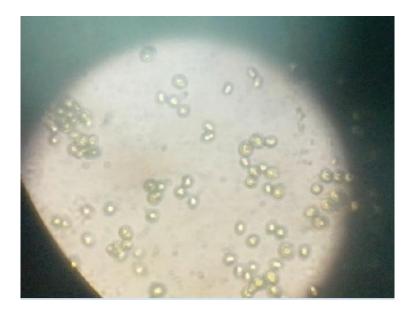


Fig .1 Image of chlorella species under 45x



#### Fig. 2 Image of chlorella species under 10x magnification

#### **Experimental Design**

#### Sample Collection

The wastewater has been collected from the collection tank of domestic sewage treatment plant before it undergoes any pre treatment.

#### **Feasibility Studies**

The experiments have been designed to check the feasibility of growth of microalgae in the raw sewage. In order to determine the growth potential, the raw sewage sample has been collected and various dilutions were made viz., 10%, 20%, 30%, and 40% without sterilization. A 10% inoculum was added to various sample dilutions and raw sample (100%) for nutrient removal. All the experiments were carried in 500ml conical flasks with working volume of 300ml and the flasks were covered with cotton plug for gaseous exchange system under the continuous illumination of light (TL5 lamp). The culture was subjected to continuous illumination with 1500 Lux measured using TES light meter (TES CORP). The nutrients present in the sewage have been analyzed as per standard procedure of American Public Health Association <sup>13</sup>

#### **Growth Curve And Nutrient Analysis**

The spectral analysis was carried out at 680 nm<sup>7</sup> for monitoring the cell growth using UV Spectrophotometer (make: CYBERLAB, USA). The sample was centrifuged at 5,000 rpm for 15 minutes and the supernatant was taken for nutrient analysis.

#### **Results And Discussion**

#### Growth Of Algae In Wastewater

The raw sewage (100%) has been diluted to various concentrations viz., 10%, 20%, 30% and 40% to study the algal growth potential. The growth patterns of 10%, 20%, 30% and 40% diluted samples are depicted in Fig. 3. It can be noticed from the figure that the growth of microalgae is viable in sewage without sterilization and hence, the inoculum was added to the raw sewage (100%) and the growth was observed, as depicted in Fig. 4. It can be noticed from the figure that growth curve has be obtained as the microalgal cells were in the log phase of during the period of  $2^{nd}$  to  $4^{th}$  day, the stationery phase has been achieved at  $4^{th}$  day and remains till  $6^{th}$  day. The microalgae growth has been observed up to 9 days. From the figure, it is evident that the microalgae (*Chlorella*)

*sp*) can grow in the domestic sewage under the continuous illumination of light. Further, after the growth period the sample has been taken for nutrient analysis.

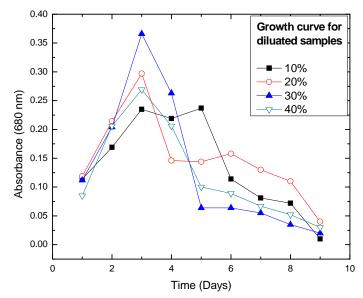


Fig.3 Growth Curve of Microalgae in diluted samples

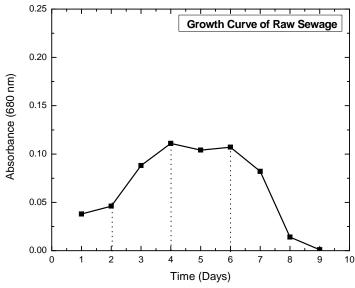


Fig. 4 Growth curve of Microalgae in raw sewage

# **Nutrient Removal**

The nutrients such as COD, Ammonia-Nitrogen, Phosphorous, Nitrates-Nitrogen, and Total solids(TS), Total suspended solids(TSS) and Total dissolved solids(TDS) have been analyzed and the results are shown in Table 1.

Parameters	Before	After	Removal (%)
(mg/L)	Microalgal Growth	<b>Microalgal Growth</b>	
COD	160	21	86.875
Phosphate	108	10	90.74
Nitrates-Nitrogen	1.558	0.623	60.01
Ammonia-Nitrogen	1.7	0.7	58.82
Total solids	220	30	86.364
TSS	20	10	50
TDS	200	20	90

#### Table 1: Nutrient Analysis of Raw Sewage

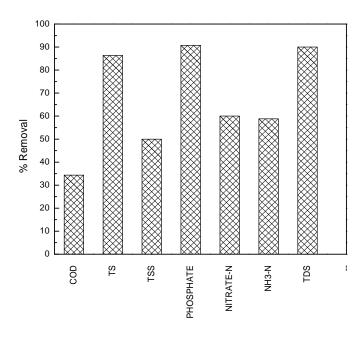


Fig. 3: Nutrient Removal

The nutrient removal characteristics of microalgae are shown in Fig. 5. It can be observed from the figure that among the parameters phosphate and TDS removal is maximum, i.e., 90.7 % and 90%, respectively. Further, the nitrate was reduced from 1.55 mg/L to 0.623 mg/L which is about 60% of removal. Also, COD was reduced from 160 mg/L to 21 mg/L i.e., 86.875% of removal after the 9 days of algal growth, whereas ammonia has been reduced from 1.7mg/L to 0.7 mg/L i.e., 58% removal.

## Conclusion

The main aim of this study was to check the ability of microalgae (*Chlorella sp*) to grow in raw sewage (100%) without sterilization at a laboratory scale batch process under continuous illumination of light and to study the nutrient removal after the algal growth period. The nutrients such as phosphate, nitrates are having the major role in supporting the growth of algae in sewage. Based on the experimental results, it can be concluded that the microalgae could easily grow in the raw sewage and can be taken for large scale process such that it can be used for waste water treatment and as a source of commercial feedstock for energy production.

## Abbreviations

- TSS -Total suspended solids (mg/L)
- TDS -Total dissolved solids (mg/L)
- COD -Chemical Oxygen Demand (mg/L)
- NH<sub>3</sub>-N Ammonia-Nitrogen (mg/L)

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