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Research paper

Biomass study of *Anabaena sphaerica* cultivated in electroplating industrial effluent

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Abstract

The growth of microalga Anabaena sphaerica in electroplating industrial effluent with reference to the vegetative cells , heterocysts and dry biomass weight brings about oxygenation and mineralization in addition to the increase in biomass which serve as a multipurpose raw material to the industries. From lab scale experiments it is demonstrated that the present investigation could be converted to pilot study for large scale production of biofuel and other commodity chemicals. Cultivation of microalgal biomass as a potential resource / raw material for various industries to produce commodity chemicals could enhance the economy and curtail the environmental hazards.

Keywords: *Microalgae; Anabaena Sphaerica; Electroplating Effluent.*

1. Introduction

The growing global population and the need of large amounts of energy effected a least care about environment. The direct and indirect anthropological activities had a negative impact on the environment and human health leads to the depletion of resources at faster rate and increase of pollutants at high levels. Hence curtailing the load of pollutants in the environment and remediating the polluted sources are the need of hour.Bioremediation by various microbes is the part of green technology and circular economy (Ryckebosch,et al., 2014; Tao, et al .2017 ; Srinuanpan et al.2020; Peter et al.2021).
Several wastewaters from domestic, agricultural, and various industries consist of enormous nutrients . Any waste water can be used as an inexpensive alternative resource for the raw nutrients to cultivate microalgae.
(Metcalf and Eddy, 1997;Li et al 2011; Yuan et al 2013; Sforza et al 2015).
Electroplating industries are characterized based on its infrastructure, production and quality of raw materials used. Source of water in each unit varies. Few industries use distilled water as source for chrome plating and zinc plating industries use tap water or ground water based on their availability. Many small scale units of EP industries release not only waste waster but also the toxic load. Proper treatment has to be provided for the safe discharge of electroplating effluent (Wood and Haselkorn,1980; Wolk et al.1994; Zhu and Lee 1997; Yasodha et al 2009; Kumazaki et al.,2013; Khanzada,2020).
Blue green, filamentous, intercalary heterocystous ,branched trichomes with oxygenic photosynthetic thallus. Anabaena are heterocyst-forming, photoautotrophic cyanobacteria that perform oxygenic photosynthesis. Anabaena grow in long filaments of vegetative cells. The envelopes of these BGA are similar to those of gram negative bacteria.Lipopolysaccaride of these BGA envelop is helpful in ionic exchange properties/intracellular accumulation/adsorption onto cell surface.Yasodha,2009. In Anabaena the cells are ovoid or barrel-shaped, often giving the filaments (trichomes) the appearance. Anabaena possesses heterocysts and can also develop akinetes (thick walled resting cells that can survive in sediments for many years). Biosorption is possible due to the unique and complex structure of the microalgal cell wall (Laamanen and Kuosa, 2005;Corrales-Guerrero et al., 2014 and Tao et al.2017).
With this background the physico chemical characteristics of the electroplating industrial effluent were analysed for its constituents before and after the microalgal treatment. Anabaena sphaerica had been selected to find out its growth response in electroplating industrial effluent with reference to vegetative , heterocyst cells and dry biomass weight.

2. Materials and methods

Physico chemical characteristics of electroplating effluent such as pH,dissolved oxygen,total dissolved solids, total suspended solids ,COD,BOD,sulphate,chloride,nickel,chromium,copper,zinc and ferrous were analysed by the methods of APHA,2012.
Microalga Anabaena sphaerica was cultivated in growth chamber(under 12/12 h light/dark cycle by fluorescent illumination of 40 $\mu Em^{-2}s^{-1}$) in 250 mL flasks with 150 mL, in BM and EPI effluent incubating at 25 ± 2◦ C. (Allen and Arnon ,1955).Triplicates were maintained in each treatment.
Sterilized glass beads were added to the culture flasks .So that microalgal cells sticking on the glass wall and clumping of cells were avoided. Gentle shaking of the cultures was done manually every day to reduce the clumping of cells.

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Treatments followed for microalga cultured in BM and EPI effluent

T1-Concentrated Basal Medium
T2,T3 and T4-diluted Basal Medium

T5-Concentrated Electro plating industrial effluent
T6,T7,T8 - diluted Electro plating industrial effluent
Growth Characteristics of Anabaena sp in terms of number of vegetative cells and number of heterocysts were observed in both BM and EPI effluent and dry weight of algal biomass were recorded during the experimental period of 20 days. Growth was observed and subsequent readings were taken on 4^{th} ,8^{th} ,12^{th} ,16^{th} and 20^{th} days of growth period. Microscopic observation: From the 100 randomly selected filaments, the total number of cells, number of vegetative cells and heterocysts were counted.
For biomass study, a known amount of culture was harvested on the last day (20th day) of the study and oven dried at 105^0C until constant weight was obtained and was expressed in mg/l.
All the data were analysed statistically using Duncan's test as per the method suggested by Snedecor and Cochran,1991.

3. Results and discussion

Results of physico chemical characteristics of electroplating effluent recorded before and after algal treatment were predicted in Table-1.The heavy load of pollutants from the EPI effluent were absorbed by Anabaena sphaerica effected a sustainable way to treat the EPI effluent.The previous investigations based on waste waters and microalgal treatments are in agreement with Dufosse et al. 2005;Desbois et al 2009; Razzak el al.2017.

Table 1: Physico Chemical Characteristics of Electroplating Effluent BM

S.No	Parameters	Electroplating effluent (before treatment)	Electroplating effluent (after treatment)
1	pH	5.7	7.9
2	dissolved oxygen (ppm)	7.4	8.1
3	Total dissolved solids (mg/ml)	5	3
4	Total suspended solids (mg/ml)	6	5.2
5	Chemical Oxygen Demand (ppm)	38.4	18.4
6	Biological Oxygen Demand (ppm)	150	193
7	Sulphate(mg/ml)	3.1	2.19
8	Chloride (mg/ml)	1.4	0.4
9	Nickel (mg/ml)	1.2	0.2
10	Chromium (mg/ml)	3.7	0.19
11	Copper (mg/ml)	2.81	0.21
12	Zinc (mg/ml)	1.02	0.04
13	Ferrous (mg/ml)	2.76	0.06

Table 2: Composition of Basal Medium

Medium component	Weight (g/l)
Calcium Chloride	0.2
Magnesium Sulfate (anhydrous)	0.098
Potassium Chloride	0.4
Sodium Bicarbonate	2.2
Sodium Chloride	6.8
Sodium Phosphate Monobasic (anhydrous)	0.12
L-Arginine • HCl	0.021
L-Cystine • 2HCl	0.016
L-Histidine (free base)	0.008
L-Isoleucine	0.026
L-Leucine	0.026
L-Lysine • HCl	0.03647
L-Methionine	0.0075
L-Phenylalanine	0.0165
L-Threonine	0.024
L-Tryptophan	0.004
L-Tyrosine • 2Na • $2H_2O$	0.02595
L-Valine	0.0235
D-Biotin	0.001
Choline Chloride	0.001
Folic Acid	0.001
myo-Inositol	0.002
Niacinamide	0.001
D-Pantothenic Acid (hemicalcium)	0.001
Pyridoxal • HCl	0.001
Riboflavin	0.0001
Thiamine • HCl	0.001
D-Glucose	1
Phenol Red • Na	0.011

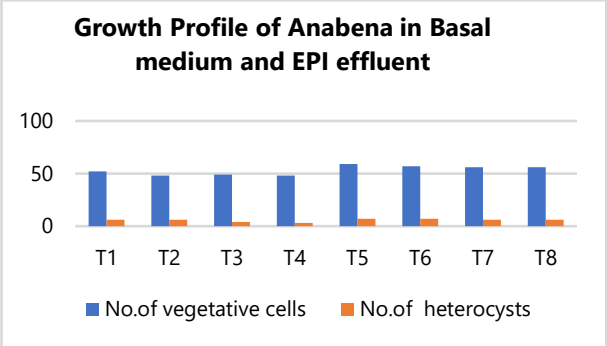


Fig. 1: Dry Biomass of Anabaena SP Cultured from Basal Medium and EPI Effluent.

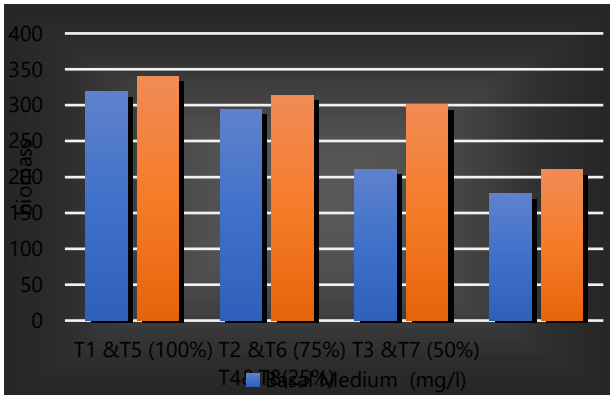


Fig. 2: Dry Biomass of Anabaena SP Cultured from Basal Medium and EPI Effluent.

Results pertaining to the nutrient uptake by the vegetative cells and heterocyst cells was faster and enhanced the efficient growth of wet biomass of experimental microalga. Similar kind of experiments with reference to cell differentiation and heterocyst cells were conducted by Luther 1990 ;Chen et.al,1997; Laamanen and Kuosa ,2005;Bothe et al.,2010.

Similar culture studies were performed by Helena et al.1986 ; Cellular differentiation studies revealed the faster nutrient uptake of microalgae from different nutrient sources are well documented by earlier studies of : Valladares et al.,2007; Kumar et al.,2010; Ehira,2013.

Studies on growth of microalgae in various nutrient composition and metabolic profile were revealed by Cardona et al., 2009; Flores et al., 2010; Herrero et al., 2013; Sforza et al.,2015;Santamaría-Gómez et al., 2018.

Growth performance with reference to the heterocyst cells of different microalgae in various waste waters are well documented by Meeks et al. 2002 and Carey et al. 2012.

The result of present investigation showed an increasing trend in the growth of microalga Anabaena sp biomass grown in all concentrations of electroplating industrial effluent. In 100% basal medium, dry biomass weight was good but a decreasing trend in other dilutions of basal medium. This might be due to the less supply of nutrient impact on the dry weight. Assessments were recorded for the Anabaena sp biomass grown in the electroplating industrial effluent revealed that in all concentrations there was an increasing trend. The results for dry biomass weight production are in agreement with Jais et al.2017; Lima,2020.

Dry biomass of Anabaena sphaerica cultivated from the treatments T5 and T6 showed high performance compare to the dry weight obtained in basal medium in which sufficient nutrients are supplied. Hence it had been concluded that the EPI effluent is effectively enhancing the growth microalga which could be an alternative and sustainable nutrient source.

Dry microalgal biomass yield from waste waters by Zhu and Lee, 1997;Yasodha 2009; Yuan et al,,2013 and Sforza et al.,2015 supported the view of the present study . Brandao et.al, 2023 reviewed about various microalgae yield in terms of biomass production from domestic waste waters revealed the potentiality of nutrient uptake of micaroalgae.

4. Conclusion

The growth of microalga Anabaena in electroplating industrial effluent with reference to the vegetative cells and heterocysts brings about oxygenation and mineralization in addition to the increase in biomass which serve as a multipurpose raw material to the industries. The enhancement of dry biomass was ranging from 11.5% to 45% for Anabaena revealed the potentiality of its nutrient uptake from EPI effluent and cleared the pollutants such as the heavy metals . The dry biomass of microalgae Anabaena sphaerica grown in electroplating industrial effluents could be utilized as an economical resource potential for the commodity chemicals.

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