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Bioremediation of Petroleum Effluent by Freshwater Microalgae

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A B S T R A C T

Freshwater green microalgae *Chlorella vulgaris Beyerinck* , *Scenedesmus quadricauda, Chlamyomonas reinhardtii* and bluegreen microalgae *Nostoc muscorum,Anabaena ambigua* ,*Oscillatoria animalis, Oscillatoria sancta, Spirulina maxima* and *Spirulina platensis* were obtained from Culture Collection of Algae, Centre for Advanced Studies in Botany, University of Madras, Guindy Campus, Chennai-600025. These microalgae screened for petroleum effluent utilization as carbon source on agar salt medium. The microalgae *Scenedesmus quadricauda, Chlamydomonas reinhardtii, Chlorella vulgaris Beyerinck* and bluegreen microalgae *Anabaena ambigua, Nostoc muscorum, Oscillatoria animalis ,Oscillatoria sancta, Spirulina maxima* and *Spirulina platensis* showed better growth.*Scenedesmus quadricauda ,Chlamydomonas reinhardtii, Chlorella vulgaris Beyerinck* and bluegreen microalgae *Anabaena ambigua, Nostoc muscorum, Oscillatoria animalis* and *Oscillatoria sancta* they were selected for further petroleum biodegradation studies. These microalgae degraded petroleum effluent levels in culture medium studies.

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Introduction

Petroleum hydrocarbons are widely spread in the environment. These hydrocarbons continue to be used as the principle source of energy and are natural products as well as pollutants. Incessant oil spills caused severe damage to aquatic and terrestrial environment. Biodegradation of organic compounds is the complex destruction of their molecular structure by microorganisms (Atlas and Bathra.,1992).In bioremediation process, hydrocarbons were consumed by microorganisms into CO_2 and H_2O.The complete mineralization of the starting compound to simpler ones like CO_2, H_2O, NO_3 and other inorganic compounds. Both aerobic and anaerobic biodegradation have been shown to reduce the concentration of several components of petroleum. Microorganisms like algae, fungi and bacteria play a vital role in biodegradation of toxic chemicals in the environment. Natural populations of these microorganisms are the main process acting in the degradation of hydrocarbon-polluted environments. Hydrocarbon degrading microorganisms play an important role in the aquatic environment. Microalgae have been identified in oil polluted environments. Algae are universally acknowledged as playing a very important role in natural water purification process (Han *et al*., 2000; Olguin, 2003). They were used for domestic and industrial waste water treatment. Microalgae; particularly the green phototropic ones have been used to remove organic matter, inorganic nutrients, heavy metals and even toxic organic contaminants from wastewater, due to their low capital investment, low operation cost and high efficiency.

Material and Methods

Collection of Microalgae

The freshwater organisms *Scenedesmus quadricauda ,Chlamydomonas reinhardtii, Chlorella vulgaris Beyerinck* and blue green microalgae *Anabaena ambigua, Nostoc muscorum, Oscillatoria animalis, Oscillatoria sancta, Spirulina maxima* and

Spirulina platensis were obtained from Culture Collection of Algae, Centre for Advanced Studies in Botany,University of Madras,Guindy Campus,Chennai-600025.The collected algae were maintained in culture medium at culture room conditions.Green algae cultured in bold basal medium and bluegreen algae in BG11medium whereas *Spirulina maxima* and *Spirulina platensis* cultured in Zarrouk's medium.

Screening studies of microalgae

These freshwater organisms *Chlamydomonas reinhardtii, Chlorella vulgaris Beyerinck, Scenedesmus quadricauda* and blue green microalgae *Anabaena ambigua, Nostoc muscorum,Oscillatoria animalis, Oscillatoria sancta, Spirulina maxima* and *Spirulina platensis* were screened on mineral salt medium at 1% petroleum effluent concentration and incubated for one week under culture room conditions.

Total Petroleum Hydrocarbon (TPH) Analysis

At each time of analysis, 50 ml of the sample solution was taken in a 150 ml separating funnel to which 10 ml of hexane was added, shaken manually for 2 minutes and allowed to stand for 20 minutes. The water layer was drained off ; hexane layer collected in quartz curvet and read using UV/Visible spectrophotometer at a wavelength of 350nm. (Medjor O.W *et al* ; 2012).

Results and Discussion

Screening of freshwater green and blue green micro algae for petroleum effluent hydrocarbon utilization and biodegradation

The fresh water green algae *Chlamydomonas reinhardtii ,Chlorella vulgaris Beyerinck, Scenedesmus quadricauda* and fresh water blue green algal species *Anabaena ambigua, Nostoc muscorum, Oscillatoria animalis, Oscillatoria sancta, Spirulina maxima* and *Spirulina plantensis* were used for screening studies of hydrocarbons.*Chlamydomonas reinhardtii* and *Chlorella vulgaris Beyerinck* showed better tolerance than *Scenedesmus quadricauda* (Pic.1a). whereas in blue green algae *Anabaena ambigua, Nostoc muscorum, Oscillatoria animalis, Oscillatoria sancta* showed better tolerance than *Spirulina maxima* and *Spirulina plantensis* (Pic.1b).

Chlamydomonas reinhardtii

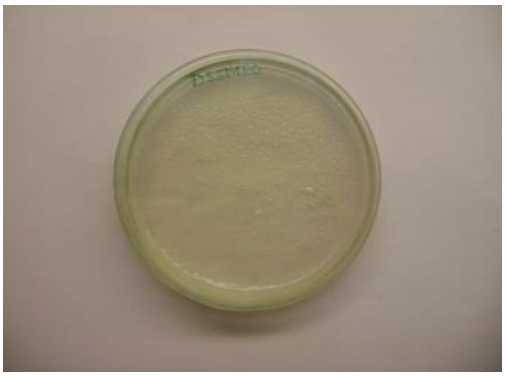
Chlorella vulgaris Beyerinck

Scenedesmus quadricauda

Picture:1.a. Screening for petroleum effluent utilization and biodegradation by freshwater green microalgae.

Anabaena ambigua

Nostoc muscorum



Spirulina maxima

Oscillatoria sancta

Spirulina plantensis

Oscillatoria animalis

Picture: 1.b. Screening for petroleum effluent utilization and biodegradation by freshwater blue green microalgae.

Biodegradation of 7 micro algae *Chlamydomonas reinhardtii, Chlorella vulgaris Beyerinck, Scenedesmus quadricauda, Anabaena ambigua, Nostoc muscorum, Oscillatoria animalis* and *Oscillatoria sancta* were studied at 1% petroleum effluent concentration in culture medium for 30 days.The results showed all microalgae were capable to tolerate and utilized petroleum effluent as a carbon source for biomass and also had the capacity to degrade petroleum effluent.(Picture 2) .

a) Green microalgae

b) Blue green microalgae

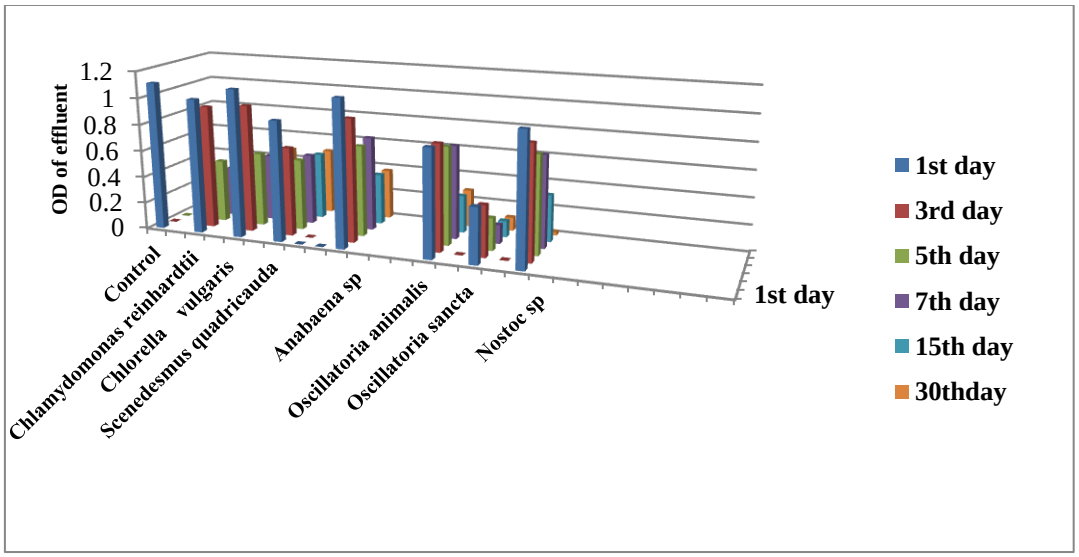
Picture: 2. Biodegradation cultures of 7 microalgae

Conclusion

Screening and biodegradation studies of microalgae showed that microalgae can tolerate to grow in petroleum effluent cultures. *Scenedesmus quadricauda, Chlamydomonas reinhardtii, Chlorella vulgaris Beyerinck, Anabaena ambigua, Nostoc muscorum, Oscillatoria animalis* and *Oscillatoria sancta* were studied at 1% petroleum effluent contain ing medium for 30 days.The results showed that all microalgae have utilized petroleum effluent as a carbon source and also have capability to degrade petroleum effluent (Picture 2) .

Table: 1. Petroleum bioremediation by microalgae.

Samples	1st day	3rd day	5th day	7th day	15th day	30thday
Control	1.110	-	-	-	-	-
Chlamydomonas reinhardtii	1.010	0.924	0.468	0.375	0.361	0.360
Chlorella vulgaris Beyerinck	1.107	0.955	0.555	0.498	0.488	0.479
Scenedesmus quadricauda	0.902	0.665	0.534	0.531	0.501	0.491
Anabaena ambigua	1.101	0.922	0.682	0.707	0.384	0.375
Oscillatoria animalis	0.804	0 .794	0.734	0.702	0.282	0.280
Oscillatoria sancta	0.418	0.385	0.242	0.141	0.122	0.102
Nostoc muscorum	0.982	0.854	0 .737	0.693	0.354	0.025



Graph: 1. Petroleum biodegradation by Microalgae.

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