Research Article

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EXTRACTION OF LIPID FROM ALGAE GROWN IN DIFFERENT OPEN CAST MINING AREAS OF JHARIA COALFIELD UNDER DISTRICT DHANBAD, JHARKHAND: AN EXPERIMENTAL STUDY

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ABSTRACT

Introduction: Microalgae have been enjoying the focus of several renewable energy researchers for past few decades, due to their high photosynthetic activity and lipid content. These lipid acts as a precursor for the production of biodiesel, a green fuel.

Aim: The present experimental study was done to estimate the lipid prospect of microalgae growing in the wastewater of abandoned coal mining and open cast areas of Jharia Coalfield of Dhanbad district, Jharkhand, India.

Methodology: The algae samples were collected from wastewater accumulated in different coal mining areas as well as from the municipality water bodies like pond and river. They were then morphologically analyzed under the microscope to identify the most prevailing algal species in the coal mining areas. Lipid was extracted from these species by the traditional Chloroform and Methanol method i.e., Bligh and Dyer method.

Results: The species prevailing in the coal mine waste water were identified as Spirogyra and Oscillatoria. Estimation of the lipid content extracted showed the variation of lipid content from algae grown in mine water to the algae grown in river water and pond water. Algal biomass from coal mining areas contained 16.3% more lipid than the algal biomass from the pond water.

Conclusion: The findings reveal the different algal species which are predominantly found in the coal mine waste water. It also gives an idea about the quantity of lipid that can be extracted from them. Thus, this experimental study gives an idea about the possible use of the barren wasteland of the coal mining areas for the algae cultivation, in order to generate the biofuel.

Key Words: Microalgae, Lipid extraction, Coal mining areas, Wastewater, Open cast mines, Biofuel

INTRODUCTION

The vast difference between the demand and supply of fossil fuels such as coal, petroleum and the natural gas and the increasing concern about their harmful effect on the environment has resulted in turning the focus towards the sustainable renewable energy. Microalgae biomass is one such sustainable energy source which can be easily harnessed in barren lands and wastewater [1] like the open cast mines.

Apart from various other uses, lipid extracted from the algae biomass can be used as the feedstock for biodiesel production [2]. Since long time, algae have been viewed as having a vast potential resource of energy because of their ability to harness solar energy via photosynthesis, which is almost 70% greater than other green plants. Algae are the fastest growing plants in the world and about 50% of their weight is lipid, a feedstock of bio fuel/biodiesel used for running cars, trucks, airplanes, etc. They can grow almost anywhere, even on sewage or salt water and do not require fertile land as food crops; the processing requires less energy than the algae provides. Algae can be a replacement for oil based fuels, one that this more effective and has no disadvantage.

Coal is the dominant energy source in India, accounting for more than half of the country's requirement. Jharia

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coalfield has a large amount of coal deposit and is one of the largest producers of coal in the world. In the coal mining operation, every year huge quantity of water is discharged from the coal mines to the rivers to facilitate safe mining. As per the estimates, the coal mines of Jharia region discharge about 3, 40, 120 gallons per minute of waste water, which amount to 2.22 million cubic meter of water discharged per day approximately. Besides, a huge quantity of unused mine water is already available in all abandoned open cast mines. These abandoned coal areas can be used for the cultivation of microalgae from which the feedstock of biofuel, i.e., lipid can be extracted.

As microalgae have faster growing rates than most terrestrial crops, the per unit area yield of oil from algae is estimated to range from 20, 000 to 80, 000 lakhs per acre per year. This is 7-31 times better than the next best crop, palm oil. The lipid and fatty acid content of microalgae vary in accordance with the culture/growing conditions. Abandoned coal mine possesses ideal conditions for the growth of algae. These coal mines are vast barren land having unlimited sunlight and carbon dioxide, which are the ultimate requirement for algal growth. Cultivating algae in these areas may put these barren lands, in the form of open pit not back-filled, into use as well as reduce the level of green house gases by carbon-dioxide sequestration.

Though there are several constraints when cultivating algae in open cast project, open cast projects are ideal for algae growth in the post monsoon duration, i.e., winter and during rainy season. However, during the summer, algae quantity is greatly reduced. Moreover, the quality and the quantity of the lipid extracted are greatly affected by the deposition of the coal dust as minute particles over the microalgae biomass. These accumulations make the lipid extraction process longer as the algae has to be thoroughly cleaned and exempted from these dust particles before undergoing the lipid extraction process.

The objective of the present work is to explore the lipid prospect from microalgae found in abandoned open cast coal mines/pits, which involves;

a. The analysis of conditions required for higher algal growth in the waste water pits found in the abandoned open cast coal mines.
b. Determination of algal lipid potential in the open cast coal mines/ abandoned pits.

METHODOLOGY ADOPTED

Standard operating protocols were followed for the sample collection, microscopic identification of the algae species and lipid extraction from them. All the samples were collected with utmost care in the sterilized jars and brought to the laboratory under aseptic conditions. In the laboratory, the algae samples and the chemicals were used as defined under the standard protocol.

Collection of Algae Samples

The algae samples were collected in sterilized jars from the water accumulated in different abandoned open cast mines of Jharia Coalfield situated at Dhanbad, Jharkhand, India. The temperature details and the environmental data were carefully noted for each day of the sample collected. They were brought to the laboratory under aseptic conditions to carry out the further experiment.

Species Identification

The samples collected were first taken for the microscopic identification of the algal species present, under the standard operating protocols.

The purity of the culture was maintained through microscopic observations and isolates were grown under 30°C with 300 lux light intensity for 12 days and night cycle in all 9 algae water samples collected. Further, the isolates were extracted for lipid production from all the samples collected.

Lipid Extraction

The method adopted for the extraction of lipids from the algae samples collected from different locations of the coalfield area of Jharia, Dhanbad was the traditional Chloroform and Methanol extraction method as proposed by Floch et al [3] and Bligh and Dyer [4]. This is a rapid method of extraction of lipids by means of phase partition of a ternary mixture of chloroform and methanol (methyl alcohol).

RESULT AND DISCUSSION

In the present study, the cyanobacteria and green algae were isolated from 9 different sites in district Dhanbad of Jharkhand, India of which 7 sites were from Jharia Coalfield and 2, which included 1 pond and 1 river, were from the Dhanbad Municipality. Totally 2 isolates, i.e., Cyanobacyeria e.g., *Oscillatoria* sp. (Figure 1) and green algae e.g., *Spirogyra* sp. (Figure 2) were morphologically identified from the algal samples (Table 1).

The occurrence of green algae and cyanobacteria in wastewater has been supported earlier by the works of Ramachandra *et al* [5] and Sriram and Seenivasan [6].

In the present study, the lipid content from *Oscillatoria* sp. and *Spirogyra* sp. was extracted in combination and compared with all the 9 algae samples collected from different areas.

All the 9 algae samples containing algae species were dried and 100g dry weight were taken evenly for the extraction of lipid, which was found to be 0.627, 0.671, 0.785, 0.546, 0.582, 0.733, 0.669, 0.796 and 0.574 ml per 100 g of dry weight (Figure3). This result was similar to the work done by Sheriff Hossain and Shellah [2].

Therefore, the result proved that lipid can be produced from both cyanobacteria and green algae. But *Spirogyra* sp. contains higher lipid content than the *Oscillatoria* sp. Kamalbasha [7] reported and agreed with the experiment.

The 3 types of water taken for the study were coal mine water, river water and pond water. As stated by many researchers, mine water contains various chemicals and micro particles which act as nutrients for the effective growth of microalgae. The air surrounding the coal mining area tends to be highly polluted having high level of carbon dioxide. These add to the growth of the microalgae and thus, also help in the effective algal biomass production from them.

As agreed by Convertis *et al* [8], decrease in nitrogen concentration and increase in other micronutrients in the water lead to the increase in the lipid content of the microalgae. The same was agreed upon by this experiment. Figure 4 below shows the variation in the lipid content extracted from the algae samples which were collected from 3 different types of water-locations, viz., coal mine, river and pond.

Figure 4 depicts that in the present study, there was 15.6% reduction in lipid production from river water than the coal mine waste-water. Moreover, there was reduction of 1.3% lipid concentration in pond water compared to the river water lipid production. However, there was a reduction of 16.3% lipid concentration in pond water when compared to the lipid production from coal mine wastewater, from the algal biomass growth.

Table 1: Morphological Identification of Cyanobacteria and Green Algae from Jharia Coalfield, Dhanbad, Jharkhand, India.

Algae species Identified	Description	Size (μm)	Algae Type
Spirogyra sp.	Filamentous, unbranched; form slippery filmsy green aggregate on the water surface.	2.50 – 4.38	Green
Oscillatoria sp.	Filamentous unbranched algae with mucilaginous sheath, occurring singly or in tangled mats; named for its oscillating movement.	20 - 40	Green

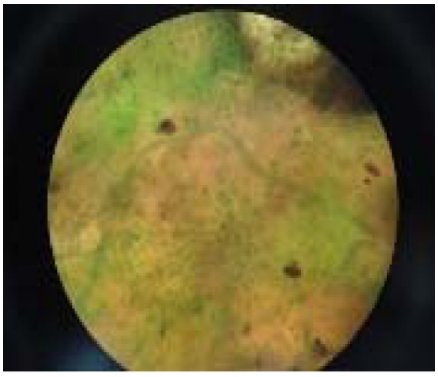

Figure 1: Microscopic View of *Oscillatoria* sp.

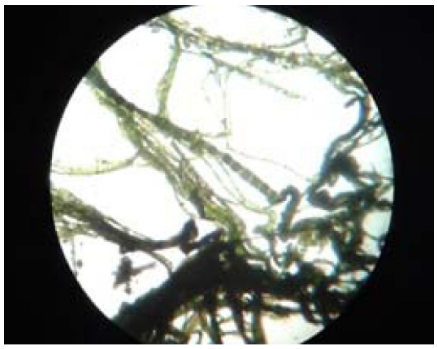

Figure 2: Microscopic View of *Spirogyra* sp.

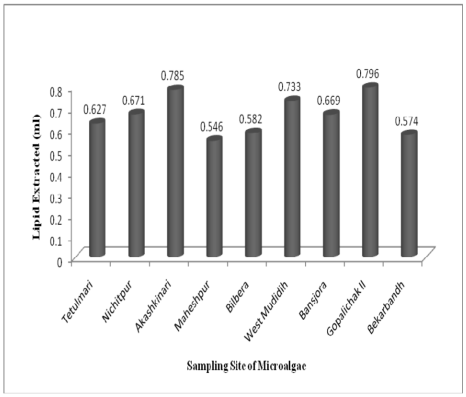

Figure 3: Lipid extracted from different sites in district Dhanbad, Jharkhand.

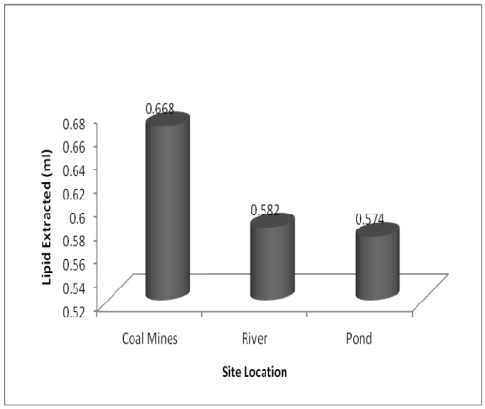

Figure 4: Comparative study of lipid content in algae samples from different types of site locations.

CONCLUSION

The findings reveal predominance of microalgae species *Spirogyra* and *Oscillatoria* in the coal mine wastewater and the presence of high quantity of lipid content in them.

The result from the above study gives an idea about the high potential of the abandoned coal mine areas for the cultivation of microalgae. It also states that the algae which are grown in the coal mining environment have the capacity of high lipid content in comparison to the algae grown in fresh water or the stagnant water.

This study and the work done will help in finding out the economic and environmental prospect of using the vast abandoned open cast mines of the coalfields. This would not only help in the carbon dioxide sequestration by microalgae (thus, reducing the carbon dioxide pollution level of the atmosphere) but also lead to the effective and efficient utilization of those barren waste lands containing the waste water. This would also prove to be an alternative source of revenue generation for the organization as well as source of employment for the local population.

Though, this experiment requires a lot of validation to quality and quantify the lipid produced, for net result biodiesel production for the compatibility of fuel vehicles, it would prove to be a step ahead in the biopurification of mine wastewater through algal biomass to generate profitable sources like lipid and electricity.

However, this study is a partial experimental approach for the utilization of barren and infertile land as a source of sustainable and renewable energy generation, in the form of biofuels. More intensive and continuous study and experiments are required to be conducted to prove this hypothesis in future.

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