



ISSN : 0973-7057

Ind. Database Index: 663 www.mjl.clarivate.com

Biochemical analysis of some blue-green algae from Saharsa district, Bihar, India

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Received : 26th October, 2017 ; Revised : 14th December, 2017

Abstract : Three local strains *Oscillatoria*, *Anabaena* and *Spirulina* of Blue-green algae were collected from the local water bodies of Saharsa District. Chlorophyll, protein, carbohydrate and lipid were estimated in each strain.

Keywords : Blue-green algae, *Oscillatoria*, *Anabaena* and *Spirulina*

INTRODUCTION

Saharsa is located at 25.88°N 86.6°E. It has an average elevation of 41 metres (134 feet). Saharsa and its surrounding areas are a flat alluvial plain forming part of the Kosi river basin. This makes the land very fertile. However, frequent changes in the course of the Kosi have led to soil erosion. Flooding is a major reason for the connectivity of the area. A large number of water bodies like Chours, Ponds and Ditches are present in different areas of the district in which water remain logged around the year. These water bodies are being polluted with organic pollutants by local people. Sewage from houses, animal waste and human waste are added in these water bodies. The organic matter added in these water bodies support the luxuriant growth of blue-green algae.

METHODOLOGY

Local strains of *Oscillatoria*, *Anabaena* and *Spirulina* were identified with standard monograph and cultured in BG-11 medium. Chlorophyll, Protein, Carbohydrate and Lipid were extracted from each strain.

Chlorophyll extraction

For the extraction of pigment from filamentous blue-green algae (*Oscillatoria*, *Anabaena* and *Spirulina*) harvested material was suspended in 90% hot methanol and filtered through Whatman's filter paper. After a second and third extraction, supernatants containing chlorophyll a were combined and made up to a known volume obtained the absorption spectra. The remaining material extracted with 90% hot methanol showed characteristic phycocyanin absorption with no trace of chlorophyll a absorption. Pigment analysis was based on optical density measurements, obtained by spectrophotometer, using aqueous extracts and 90% methanol extracts. Percent (%) chlorophyll was determined by multiplying the OD at 665 nm by 1390, the extinction coefficient of Richards and Thompson (1952) as modified by Talling and Driver (1963).

In all extraction procedures, three 20 ml aliquots of freshly harvested algal suspensions were centrifuged at 5000 x g for 5 min and washed three times with glass distilled water. After a third centrifugation of the washed suspensions, one aliquot was quantitatively transferred to pre weighed crucibles, dried for 24 h at 105°C and reweighed for dry weight determination. The second aliquot was used for chlorophyll extraction.

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Estimation of Total Proteins

The estimation of total protein was done by the method of Lowry *et al.*, (1951). The cultures were centrifuged at 7000 xg for 10 minutes. From the pellets 20 mg was treated with 10% TCA and centrifuged at 10,000 xg for 10 minutes. The resulting pellet was resuspended in 0.1N NaOH and boiled for 30 minutes, cooled and then centrifuged to eliminate light scattering materials. The supernatant was made upto known volume. To 0.1 ml of supernatant, 0.9 ml of distilled water and 5 ml of alkaline copper reagent were added and allowed to stand for 10 minutes, finally 0.5 ml of Folin-ciocalteu reagent added. The absorbance was measured after 30 minutes in Spectrophotometer against the Bovin Serum Albumin as blank.

Estimation of total Lipids

The estimation of total lipids was done by the method of Sato, (1988). The cultures were taken and centrifuged at 7000 xg for 10 minutes. From the pellet, 20 mg was taken, homogenized with extraction solvent (Chloroform:

2:1 (v/v)) and filtered through filter paper. The filtrate was vortexed with sodium sulphate to remove moisture. Then it was taken in a pre-weighed bottle and kept this overnight at room temperature in dark place. The dried extracts were reweighed and total lipids were estimated by subtracting the initial weight from the final weight. The amount of total lipid expressed as mg/g -1 dry weight.

Estimation of carbohydrates

The estimation of carbohydrates was done by the method of Dubois *et al.*, (1956). The cultures were centrifuged at 7000 xg for 10 minutes. The supernatant was discarded and 20 mg of pellets was taken in a test tube. Then it was hydrolyzed with 2 ml of Con. H₂SO₄ and mixed thoroughly. The color developed was measured at 490 nm using Spectrophotometer against glucose as blank.

RESULT

Percentage chlorophyll, protein, carbohydrate and lipid in all three local strain is tabulated in the table.

Table 1: Showing % of Chlorophyll, Protein Carbohydrate and Lipid.

Sl. No.	Name of Species	Chl a (%)	Protein (%)	Carbohydrate (%)	Lipid (%)
1	<i>Anabaena</i>	1.27	1.99	1.28	1.46
2	<i>Oscillatoria</i>	1.01	1.24	3.14	1.58
3	<i>Spirulina</i>	1.24	55	14.20	22.5

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