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AMINO ACID CONTENT AND BIOCHEMICAL ANALYSIS OF THE METHANOLIC EXTRACT OF *OSCILLATORIA TEREBRIFORMIS*

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ABSTRACT

The pure strain of *Oscillatoria terebriformis* was collected from Vivekananda institute of Algal technology (VIAT). The present study was conducted that the biochemical analysis such as Carbohydrate, Protein and Lipid content of the methanolic extract of *Oscillatoria terebriformis* was carried out in this research work and it is proved that *Oscillatoria terebriformis* contains number of essential and non-essential amino acids are investigated through the High pressure Liquid Chromatography(HPLC) Technique.

Keywords:- Cyanobacteria, Methanol, Biochemical analysis, HPLC, Amino acids.

INTRODUCTION

Cyanobacteria, also known as blue-green algae include a highly diverse group of prokaryotic microorganisms exhibiting oxygenic photosynthesis. Oxygen released by this process gradually changed the original reducing atmosphere of the primitive earth to an oxidizing one (Olsen, 2006) triggering off a dramatic evolution of global biodiversity. The chloroplasts of eukaryotic algae and higher plants have originated from endosymbiotic relationships with cyanobacteria (Martin and Kowallik, 1999 and Raven and Allen, 2003) and this event in the early evolution of life has stimulated the advent of oxygen tolerant flora and fauna capable of aerobic respiration, a highly efficient mechanism of energy utilization. The rapid development of such

organisms resulted in the predominance of oxygenic and aerobic species diversity on earth. Basically, many of the metabolites produced by the organisms are in low amounts. Hence, the present work was carried out; to study the biochemical characteristics such as amino acid content of grown cyanobacteria were investigated.

MATERIALS AND METHODS

Collection and culturing of cyanobacteria.

Oscillatoria terebriformis, a thermophilic cyanobacterium was obtained from the culture collected from the culture collection of Vivekananda Institute of Algal Technology (VIAT), Chennai, Improvised CFTRI medium was used for cultivating thermophilic cyanobacterium and the composition is as follows (Venkataraman

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etal.,1985): The algal samples were dried and the 0.5g of dried algal material was extracted in 20ml of methanol kept in an orbital shaker for overnight. The obtained extracts were filtered with Whatman no.1 filter paper and the filtrate was collected. The solvents were removed under reduced pressure at 50°C to yield a concentrated extract (15%)(Sivasubramanian *et.al.*,2011).

The Biochemical composition of *Oscillatoria terebriformis* of total Carbohydrates, Total protein and total lipids were determined by Anthrone Method (Pons 1981), Lowry's method (1951), Bligh and Dyer (1959) respectively.

Qualitative and Quantitative Estimation of Amino acids

The peptides with N – terminal primary amines get derivated by orthophthaldehyde and the amino acid contents of the cyanobacteria are separated and quantified by reverse phase chromatography (Shimadzu – High Performance Liquid Chromatograph LC6A). Separation of the sample components occur on the column by interaction between the sample components of the stationary and the mobile phase.

RESULTS AND DISCUSSIONS

The cyanobacterium was analyzed for various bio-chemicals such as carbohydrates, proteins, lipids. Experiments were conducted to identify useful bio-chemicals from the biomass of *Oscillatoria terebriformis*. Table-1 shows the levels of the following bio-chemicals.

It has been established that a wide range of cyanobacteria produced large amounts of extracellular polysaccharides which posses unique composition and structure (Parikh *et al.*,2006,Mishra *et al.*,2009).It has also been shown that a number of these polysaccharides have antitumor activities (Gardeva *et al .*, 2009 , Li H , Umemura *et al .*, 2011, Umemura *et al.*, 2003).

Cyanobacteria are a rich source of potentially useful natural products. Over 40 different Nostocales species, the majority of which are *Anabaena* and *Nostoc* sp. produce over 120 natural products (Secondary metabolites)having activities

such as anti-HIV anticancer, antifungal, antimalarial and antimicrobial.

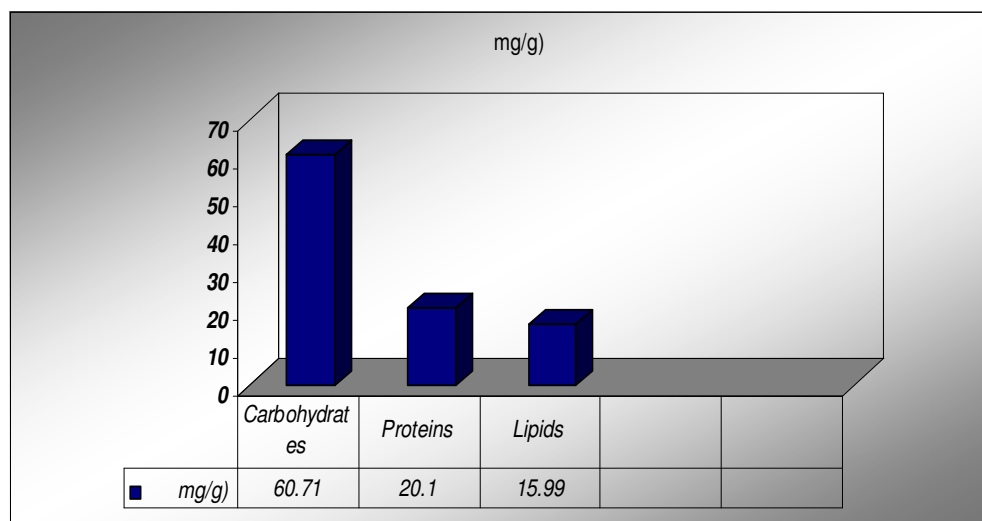
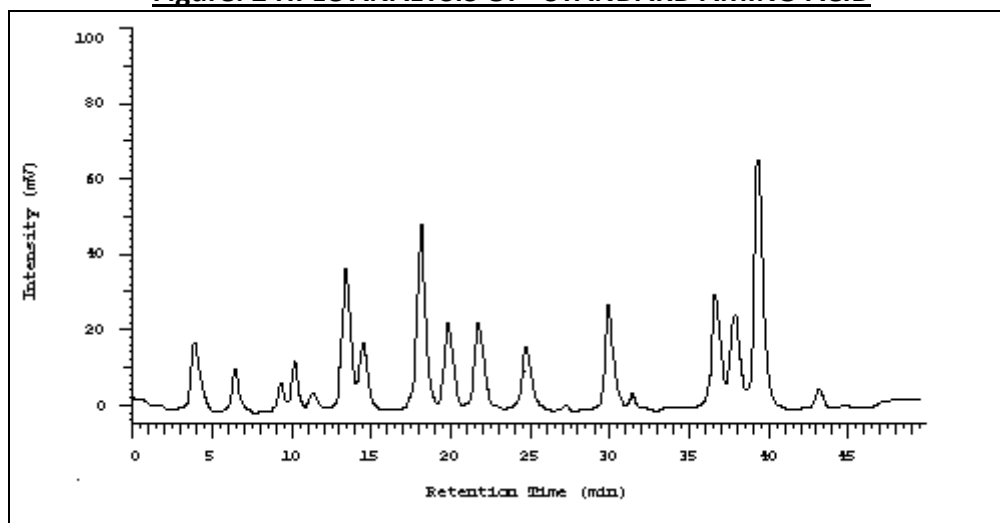
Cyanovirin (CV-N, cyanoviorin-N), a 101 amino acid protein extracted from *Nostoc ellipsosporum* was found to have potent activity against all human immunodeficiency viruses such as HIV-1, M and T tropic strains of HIV-1, HIV-2, SIV (Simian), and FIV (Feline) (Burja *et al* 2001).

Cyanobacteria have been identified as one of the most promising group of organisms from which novel and biochemically active natural products are isolated.Cyanobacteria such as *Spirulina*, *Anabaena*, *Nostoc* and *Oscillatoria* produce a great variety of secondary metabolites (Shalaby *et al.*,2010).The only comparable group is actinomycetes, which has yielded a tremendous number of metabolites. The rate of discovery from traditional microbial drug producers like actinomycetes and hyphomycetes, which are in the focus of pharmaceutical research for decades, is decreasing and it is time to turn to cyanobacteria and exploit their potential. This is of paramount importance to fight increasingly resistant pathogens and newly emergent diseases (Hermann *et al.*, 2002).

Oscillatoria terebriformis was analyzed for amino acid contents by High Performance Liquid Chromatography (HPLC) and the results are tabulated (Table 3). The essential amino acids which are required for dietary supplements are present in high amounts in *Oscillatoria terebriformis*.Thus, result shows that cyanobacterium isolated from effluents has also very high nutritional value. *Oscillatoria terebriformis* was analyzed for amino acid contents by High Performance Liquid Chromatography (HPLC). The essential amino acids and non - essential amino acids in the Cyanobacterium were estimated. Studies were undertaken to explore the presence of useful biochemicals in the *Oscillatoria terebriformis*

TABLE: 1 BIOCHEMICAL COMPOSITION OF MACROMOLECULE

S. No.	BIOCHEMICAL	AMOUNT
1.	Carbohydrates (mg/g)	60.71
2.	Proteins (mg/g)	20.10
3.	Lipids (mg/g)	15.99

FIGURE: 1. TOTAL CONTENTS OF CARBOHYDRATES, PROTEIN AND LIPIDS IN *OSCILLATORIA TEREBRIFORMIS***Figure: 2 HPLC ANALYSIS OF STANDARD AMINO ACID****TABLE: 2. HPLC ANALYSIS OF STANDARD AMINO ACID**

S.No.	Components	R.T	Area	Area
1.	ASPARTIC ACID	4.03	406656	6.343
2.	GLUTAMIC ACID	6.47	178776	2.546
3.	ASPARAGINE	9.23	78655	1.365
4.	SERINE	10.22	118913	3.067
5.	GULTAMINE	11.67	75454	1.275
6.	GLYCINE	13.45	644324	11.152
7.	THREONINE	14.89	226751	3.565
8.	ARGININE	18.08	774343	13.565
9.	ALANINE	19.53	405654	6.921

10.	CYSTINE	21.67	395644	6.787
11.	TYROSINE	24.76	264544	4.556
12.	HISTIDINE	27.25	14087	0.245
13.	VALINE	29.90	496755	8.343
14.	METHIONINE	31.41	44465	0.756
15.	ISO-LEUCINE	36.62	441967	7.554
16.	PHENYLALANINE	37.93	332334	4.620
17.	LEUCINE	39.34	954523	14.41
18.	LYSINE	43.39	63909	1.105
19.	PROLINE	45.11	4344	0.616
20.	TRYPTOPHAN	46.12	5434	0.656
21.	TAURINE	41.11	3433	0.545
	Total		5791740	100.000

Figure: 3 HPLC ANALYSIS OF AMINO ACID CONTENT IN *OSCILLATORIA TEREBRIFORMIS*

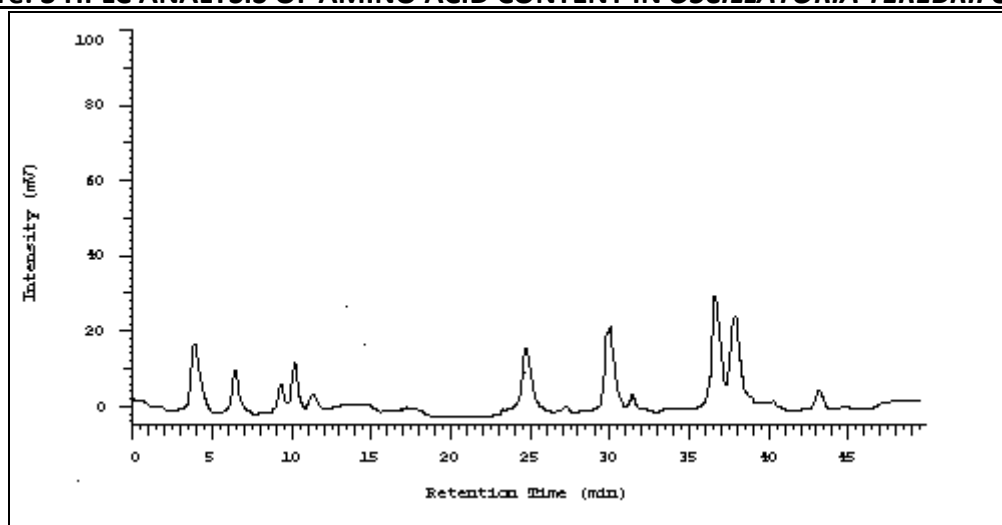


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TABLE:4. AMINO ACID PROFILE OF *OSCILLATORIA TEREBRIFORMIS*.

AMINOACIDS	Amount in µg/0/5g dry weight
IsoLeucine	1.13
Leucine	1.14
Valine	1.14
Lysine	4.03
Methionine	2.04
Tryptophan	2.34
Threonine	1.13
Arginine	1.13
Histidine	0.34
Essential Amino acids	14.42
Glycine	0.34
Tyrosine	1.13
Cystine	0.34
Alanine	3.24
Asparagine	3.13
Glutamate	1.34
Proline	1.14
Serine	0.34
Non-Essential Amino acids	11.00

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