INTERNATIONAL JOURNAL OF RESEARCHES IN BIOSCIENCES, AGRICULTURE AND TECHNOLOGY
© VISHWASHANTI MULTIPURPOSE SOCIETY (Global Peace Multipurpose Society) R. No. MH-659/13(N)
www.ijrbat.in

MORPHOLOGICAL AND BIOCHEMICAL STUDIES ON NITROGEN FIXING SOIL BLUE-GREEN ALGA *WESTIELLOPSIS PROLIFICA* JANET

G.S. Shinde
K.J. Somaiya College of Arts, Commerce and Science, Kopargaon, Dist. Ahmednagar (M.S.)
Corresponding author- gsshindebhumi@gmail.com

ABSTRACT:
The present research investigation deals with the isolation and taxonomical characterization of soil blue-green alga Westiellopsis prolifica Janet isolated from agro-practices areas of Kopargaon tehsil of Maharashtra State. Morphological studies revealed that, the alga under investigation showed almost all identical characters as described by (Desikachary, 1959). Biochemical studies with respect to total nitrogen, crude proteins, lipids and chlorophyll-a content was carried out by following standard methods. Biochemical composition showed that soil blue-green alga Westiellopsis prolifica contains 4.70% total nitrogen, 29.37% crude proteins, 5.92 µg/gm Chlorophyll-a on fresh weight basis and 11.5% lipids on dry weight basis.
Keywords: Westiellopsis prolifica, Morphology, Biochemical composition, Kopargaon tahsil.

INTRODUCTION:

Blue-green algae (Cyanobacteria) are unique in reducing the atmospheric nitrogen by the process "Biological nitrogen fixation" (Tiwari *et al*., 2001). The cyanobacteria contain nitrogenase and fix atmospheric nitrogen for which these are used as biofertilizer to maintain and improve soil status (Ahmed, 2001). Most of the heterocystous and non-heterocystous blue-green algae are known to fix nitrogen. The observation in this regard showed that *Calothrix, Hapalosiphon, Aulosira, Anabaena* and *Nostoc* were dominant nitrogen fixing cyanobacteria encountered in various agro-practices areas of Maharashtra state. Such forms hold promise for crops such as maize, rice, mungbean, tomato and sugarcane (Meelu, 1992) and wheat (Genter *et al*., 1995) by fixing nitrogen. The potential impact of these organisms on agriculture through their use as biofertilizers, soil conditioner, plant growth regulators and soil health ameliorators has been well recognized (Venkataraman, 1972 and Metting, 1988).

In recent years, the practice of utilizing blue-green algae as an efficient source of biofertilizer for various crops have been advocated and adopted in India (Venkataraman, 1981; Kannaiyan, 1990). Blue-green algae are often referred to as "Miniature Factories" of the biological world and represent an alternate source of a variety of bioactive compounds, lipids, proteins, enzymes, pigments and compounds of pharmaceutical value. However, little work has been done to exploit the full range of biochemical diversity among these organisms for production of value added products (Marquez *et al*., 1995; Prasanna *et al*., 2004).

By considering all these issues along with societal responsibilities the present investigation was carried out to study the morphology and biochemical content of soil alga *Westiellopsis prolifica* Janet isolated from agro-practices areas of Ahmednagar district, Maharashtra state.

MATERIALS AND METHODS :

BG-11 (Rippka *et al.,* 1979) and modified Fogg's medium (Fogg, 1942 and 1949) was used for isolation of blue-green algae from the soil samples. Soil Samples were collected as per the procedure given by (Singh, 1961). Unialgal isolates were grown and replicated in conical flasks containing 50 ml nitrogen free Fogg's medium and incubated for 28 days at 28±2 ^{0}C under 16/8 hr. light/ dark cycles with 2- 5 K Lux light intensity from white fluorescent tubes. Taxonomic identification was done by following the monographs and keys of (Prescott, 1951) and (Desikachary, 1959). From the raised unialgal cultures, *Westiellopsis prolifica* is used for the present study.

The total nitrogen content was estimated by Micro-kjeldahl method (Jackson, 1985). The crude protein content was obtained by estimating total nitrogen content (Micro-kjeldahl method)

and multiplied it by a factor 6.25 as per the method given by (Sadasivam and Manickam, 1996) and lipids were analyzed by the method of (Folch *et al.*, 1957) while estimation of chlorophyll-a content was carried out by following (Arnon, 1949) method.

RESULT & DISCUSSION :

Thallus filamentous, branching true with primary and secondary filaments, sheath absent, cells in single rows, heterocyst intercalary. Main filament torulose, cells short, barrel shaped, 8.5 µm broad, branch filaments elongate and thinner, not constricted at the cross walls, with elongate cylindrical cell 4.7 µm broad; heterocyst oblong cylindrical, 6.5 µm broad and 9.5- 10.7 µm long, gonidia formed singly in pseudohormocysts.

B) Biochemical Studies:

From the experimental results (Table- 1) it was revealed that, the studied alga *Westiellopsis prolifica* contains 4.70% total nitrogen on fresh weight basis. The previously reported range of nitrogen content from lowest amount, 1.72% in *Nostoc linckia* to a greatest 7.28% in *Nostoc passerinianum* studied by (Suseela and Goyal, 1995b) is categorized into three classes as C- 1.85%, B- 3.70% and A- 5.55%. Likewise, the range for nitrogen content from 0.98% in *Scytonema caldarium* to 4.27% in *Calothrix parietina* as reported by (Ahmed and Kalita, 2002) is grouped into C- 1.09%, B- 2.18% and A- 3.27% classes. In comparison with obtained total nitrogen content of alga under investigation revealed that, *Westiellopsis prolifica* fit well in class 'B' of (Suseela and Goyal, 1995b) and also positioned in class 'A' of (Ahmed and Kalita, 2002). Moreover, the obtained value of total nitrogen content (4.70%) in *Westiellopsis prolifica* is much closer to the report of (Pattnaik, 1966). The crude protein content as experienced in *Westiellopsis prolifica* was 29.37% on fresh weight basis. The previously affirmed range of proteins reported by (Mishra *et al.,* 2001) from a lowest amount of 3.72% in *Scytonema tolypotrichoides* to an utmost of 29.82% in *Nostoc paludosum*. This range is grouped into four classes viz. C- 6.52%, B- 13.04%, A- 19.56% and O- 26.08% for the purpose of discussion with the our results. The studied blue-green alga, *Westiellopsis prolifica* estimated for crude protein content, feel right into class 'O'. Neverthless, the recorded value of crude protein content in *Westiellopsis prolifica* (29.37%) is found slightly more than the reported value (22.35%) by (Dash and Mishra, 1999) and much more than the reported value 7.7%, for the same isolate from Pokharan and Achrol by (Tiwari *et al.,* 2005).

Likewise, the chlorophyll-a content of alga *Westiellopsis prolifica* appeared to be 5.92 µg/ gm of fresh weight basis. The pragmatic values obtained in *Westiellopsis prolifica* was found to be elevated than the reported values of (Tiwari *et al.,* 2005) and in agreement with that of the reported values, 5.90 µg/gm by (Suseela and Goyal, 1995a). The affirmed range of chlorophyll-a content minimum of 0.18 µg/gm in *Nostoc carneum* and maximum of 5.22 µg/gm in *Nostoc paludosum* studied by (Mishra *et al.*, 2001) is grouped into C- 1.26 µg/gm, B- 2.52 µg/gm, A- 3.78 µg/gm and O- 5.04 µg/gm categories for the simplicity of discussion. In comparison to the affirmed range, the obtained value of Chlorophyll-a in studied alga, *Westiellopsis prolifica* lies in category 'O'.

The blue-green alga, *Westiellopsis prolifica* also showed significant amount of lipid content i.e. 11.5% on dry weight basis. Vargas *et al.* (1998) revealed the lipid range from a minimum of 1.8% in *Nostoc commune* to maximum of 20.13% in *Scytonema simplex* and is further divided into various classes as C- 4.58%, B- 9.16%, A- 13.74% and O- 18.32% of lipid content for the purpose of discussion with the obtained value. In relation with the previously reported values for lipid content,

Westiellopsis prolifica occupied a position in the class 'B' of lipid content.

ACKNOWLEDGEMENT

I express sincere thanks to Dr. B.S. Yadav, Principal, K.J. Somaiya College, Kopargaon (M.S.) for the constant encouragement and useful suggestions.

BIBLOGRAPHY :

Ahmed, S. U. 2001. Nitrogen fixing potential of cyanobacteria isolated from rice field soils of Nagaon sub- division Assam. *Phykos,* **40** (1 and 2): 53- 59.

Ahmed, S. U. and Kalita, M. C. 2002. Nitrogen fixing potential of BGA isolated from rice field soils of Hojai sub-division, Nagaon, Assam. *Phykos,* **41** (1 and 2): 17- 20.

Arnon, D. I. 1949. Copper enzyme in isolated chloroplasts. Polyphenoloxidase in *Beta vuglaris. Plant Physiol.*, **24** : 1- 5.

Dash, A. K. and Mishra, P. C. 1999. Growth response of the blue green alga *Westiellopsis prolifica* in sewage enriched paper mill waste water. *Rev. Int. Contam. Ambient.*, **15** (2): 79- 83.

Fogg, G. E. 1942. Studies on nitrogen fixation by blue green algae. I. Nitrogen fixation by *Anabaena cylindrica* Lemm. *J. Exp. Biol.,* **19**: 78- 87.

Fogg, G. E. 1949. Growth and heterocyst production in *Anabaena cylindrica* Lemm. II. In relation to carbon and nitrogen metabolism. *Ann. Bot.,* **13**: 241- 249.

Folch, J. M., Less, M. and Stoare-Stanley, G. H. 1957. A Simple method for the isolation and purification of total lipids from animal tissues. *J. Biol. Chem.*, **226**: 497- 507.

Genter, M., Kerby, N. W., Rowell, O. P. and Scrimgeour, C. 1995. Colonization of Wheat (*Triticum vulgare* L) by nitrogen fixing cyanobacteria. IV. Dark nitrogenase activity and effect of cyanobacteria on nature Super sN abundance in the plants. *New Phytol.,* **129** (2): 337- 343.

Jackson, M. 1958. Soil chemical analysis. Pub. Prentice Hall, New Jersey, O.S.A.P. 183.

Kannaiyan, S. 1990. Blue green algae biofertilzers in: Biotechnology of biofertilzers for rice crop, (ed) S. Kannaiyan, Tamilnadu Agri. University Publication (Coimbatur, India). PP. 212- 225.

Marquez, F. J., Nishio, Nagai, S. and Sakai, K. 1995. Enhancement of biomass and pigment production during growth of *Spirulina platensis* in mixotropic culture. *J. Chem. Technol.,* **62**: 159- 164.

Meelu, O. P. 1992. Biofertilizers and their potential in crop prouduction. In: *Changing scenario of our environment* (Eds.) Dhaliwal, G. S. Hansra, B. S. Jerath, N. Pub. Punjab Agricultural University Ludhiana, India. pp. 281- 286.

Metting, B. 1988. Microalgae in agriculture. In: M. A. Borowitzka and L. J. Borowitzka (Eds.). *Microalgae Biotechnology,* pp. 288- 304.

Mishra, U., Pabbi, S. and Singh, P. K. 2001. Cyanobacterial diversity in Terai belt of Uttar Pradesh, India-II. Growth and nitrogen fixing potential of local heterocystous isolates. *Phykos*, **40** (1-2): 23-28.

Pattnaik, H. 1966. Studies on growth and nitrogen fixation by *Westiellopsis prolifica* Janet. *Ann. Bot.,* **30**: 231- 238.

Prasanna, R., Pabby, A. and Singh, R. K. 2004. Effect of glucose and light/ dark environment on pigmentation profiles in *Calothrix elenkinii. Folia Microbiol.,* **49**: 26- 30.

Prescott, G. W. 1951. Algae of the Western Great Lakes Area. Publ. Otto Koeltz Science Publishers, Koeningstein. : 1- 935.

Rippka, R., Derulles, J., Waterburry, J., Herdman, M. and Stanier, R. 1979. Genetic assessments, strain histories and properties of pure cultures of Cyanobacteria. *J. Gen. Microl.* **111**:1-61.

Sadasivam, S and Manickam, A. 1996. Biochemical methods. Second Edition. New Age International (P) Limited, Publishers. Pp. 34- 37.

Singh, R. N. 1961. *Role of blue-green algae in nitrogen economy of Indian agriculture*. Publ. I.C.A.R. New Delhi.: 1- 175.

Suseela, M. R. and Goyal, S. K. 1995a. Effect of ammonium nitrogen on growth and nitrogen fixation by Cyanobacteria. *Phykos,* **34** (1- 2): 123- 130.

Suseela, M. R. and Goyal, S. K. 1995b. Growth and nitrogen fixating potential of Cyanobacteria. *Phykos,* **34** (1- 2): 131- 134.

Tiwari, D., Patric, J. M. and Singh, S. 2001. Algal dynamics of the river Ganga at Kanpur. *Phykos,* **40** (1- 2): 45- 51.

Tiwari, O. N., Singh, B.V., Mishra, U., Singh, A. K., Dhar, D. W. and Singh, P.K. 2005. Distribution and physiological characterization of cyanobacteria isolated from arid zones of Rajasthan. *Tropical Ecology,* **46** (2): 165- 171.

Vargas, M. A., Rodríguez, H., Moreno, J., Olivares, H. and Del, J. A. 2002. Biochemical composition and fatty acid content of filamentous nitrogen-fixing cyanobacteria. *Journal of Phycology,* **34** (5): 812- 817.

Venkataraman, G. S. 1972. Algal biofertilizers and rice cultivation Pub. *Today and tomorrows*. Printers and Publishers, Faridabad, India.

Venkataraman, G. S. 1981. Energetics and economics of blue-green algal contribution to rice crop system. *Current Science,* **50** (2): 94- 95.

Table- 1: Biochemical composition of blue-green alga *Westiellopsis prolifica* Janet.

Sr. No.	Biochemical composition	Observed Values (± S.E.)
1.	Total Nitrogen	4.70% ± 0.02
2.	Crude Proteins	29.37% ± 0.01
3.	Chlorophyll-a	5.92 µg/gm ± 0.05
4.	Lipids	11.5% ± 0.1

*The values represented are mean of three observations. (± S.E.- Standard Error.)