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Cyanobacteria to the Effect of NaCl on Growth and Biochemical Characters

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Summary : Effect of NaCl on the growth of the Cyanobacteria *Westiellopsis prolifica* Janet has been studied. Growth was observed in different concentrations of NaCl ranging from 0.1 to 0.9M but, maximum growth, chlorophyll-a ,protein content and ammonia excreation was noted in 0.6 M NaCl.

Keywords : Chlorophyll-a, Cyanobacteria, Growth, NaCl, Protein, Heterocysts

INTRODUCTION

Cyanobacteria are oxygenic photosynthetic prokaryotes which can fix atmospheric nitrogen and are abundant in rice field ecosystem. It is evident that cyanobacteria contribute 25-30 kg N/ha to ricefields besides increasing the yield to the tune of 10 -15 percent. However, the cyanobacteria in rice fields are subjected to various field problems such as salinity, acidity, herbicide application, etc. which affect their growth and function (Gopalaswamy, 2001). Salinity is one of the most important factor in nature leading to severe crop loss every year and it is an ever increasing problem in agriculture. The area which has become unproductive due to accumulation of salts in the upper profile of soils is about 7-12 m ha in our country. The occurrence of cyanobacteria in varying saline situations have drawn much interest in recent years especially on their levels of halotolerance, mechanism of adaptation and role in amelioration of salt affected soils. Hence the present study was undertaken to know the relative tolerance of the cyanobacterium, *Westiellopsis prolifica* to salinity conditions

MATERIALS AND METHODS

The W*estiellopsis prolifica* Janet (CU 45286) was isolated from the paddy field soil of Malappuram District, Kerala state (pH 6.5) and axenic cultures of the organisms were obtained by streak plate method. The cultures were grown under continuous light at 25 ±10 C in BG-11 N- free medium (Rippka *et al*., 1979). An equal volume of exponentially growing homogenized cyanobacteria were inoculated to culture media containing NaCl having 0.1, 0.2, 0.3, 0.4, 0.5, 0.6, 0.7, 0.8, 0.9 M concentration and the experiment was set up to 42 days. The morphology was studied by observing the cyanobacteria through a stereo microscope. The growth was estimated by measuring the optical density in a spectrophotometer set at a wavelength of 760 nm against a reference blank containing a sterile medium. The total chlorophyll, protein content and heterocyst frequency were also estimated as an indicator of growth. The chlorophyll content was assayed after extracting the cyanobacteria in 80% acetone and measured at a wavelength of 665 nm, 645 nm, and 630 nm (Parsons and Strickland, 1965). The total protein content was measured according to Lowry *et al*. (1951) and Price(1965). The heterocyst frequency was measured according to Kaushik,1987.

RESULTS

The cyanobacterium *Westiellopsis prolifica* showed considerable growth in BG-11 liquid medium amended with NaCl. This halotolerant cyanobacteria grow well up to 0.9M NaCl with maximum growth at 0.6M NaCl (Graph 1). The protein content was maximum at 0.6M NaCl and it showed tremendous increase after 35 days of incubation and the amount was double over the control (Graph 2). The pigment, chlorophyll-a content was maximum at 0.6M NaCl and the increase was less when concentration was higher (Graph 3). Morphological studies revealed that the *Westiellopsis prolifica* does not lose its green colour even after 42nd day of incubation and it grows fastly on graded concentrations of NaCl with maximum number of heterocysts at 0.6M NaCl (Graph 4).

DISCUSSION

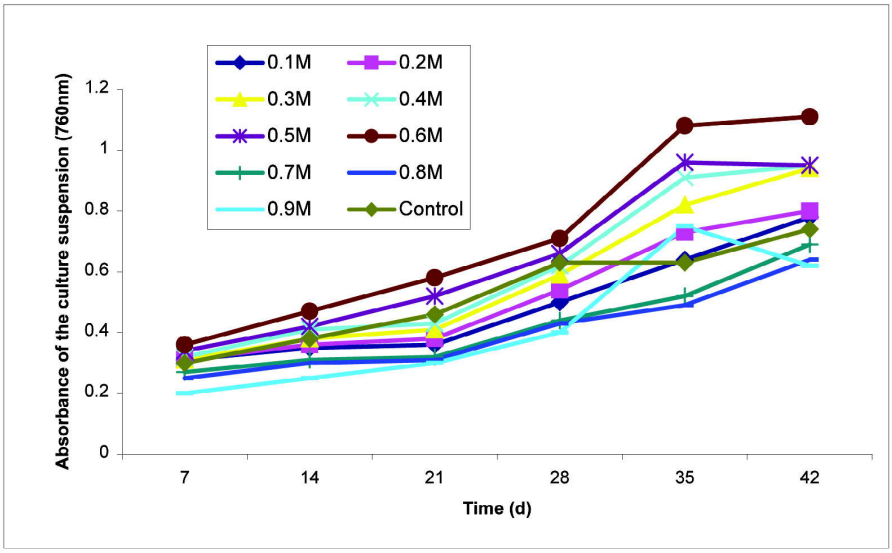
Analysis of results showed that cyanobacterium *Westiellopsis prolifica* of rice fields show a varying degree of tolerance to NaCl in culture which ranges from 0.1 to 0.9M. Earlier reports have shown that supplementation of 0.4M NaCl decreased biomass accumulation, chlorophyll-a content and cell protein levels of *Calothrix brevissima* whereas *Anabaena* sp. of freshwaters tolerated only up to 0.1M NaCl (Kumar and Kaushik, 1994). The unicellular species *Synechocystis aquatilis* showed salt tolerance up to 0.3M NaCl whereas,*Microcystis firma*

tolerated up to 1M NaCl. The growth of N_2 fixing cyanobacterium *Nostoc muscorum* was completely inhibited by 0.4M NaCl (Blumwald and Tel-Or, 1982). But according to Jose*et al.* (1996) cyanobacteria can tolerate salinity levels from 10 mM to 3M salt in the environment. All these results showed the varying degree of tolerance to NaCl in different species.

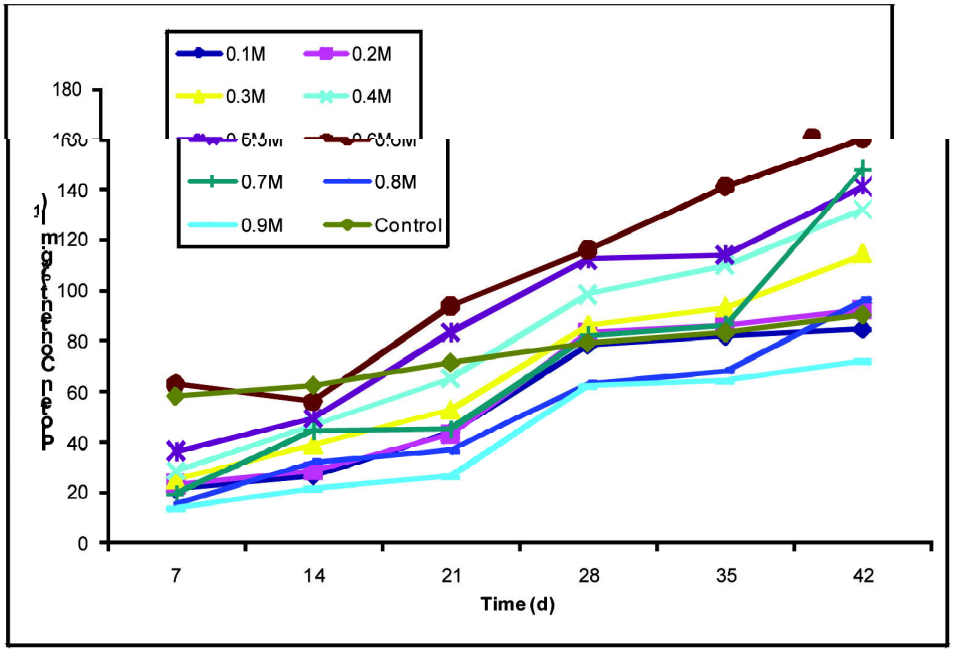
Like the results of the present investigation on the changes of photosynthetic pigments, protein content, decrease in the number of heterocysts at higher salt concentrations is in accordance with the earlier findings that the presence of salt yields a series of processes affecting cell composition, structure and function such as stimulation of photosynthesis, accumulation of sugars, reorganisation of photosynthetic apparatus and modification of cell surface (Blumwald and Tel-Or, 1982). Sodium is required for growth (Allen and Arnon, 1955), nitrogenase activity (Apte and Thomas, 1980) and photosynthesis (Apte and Thomas, 1984). Sodium plays a multiple role in cyanobacteria and may be the main causative factor for stimulated growth of *Westiellopsis prolifica* at low salt concentrations.

Salinity stress in cyanobacteria is also relieved by a variety of exogenous saccharides and alcohols. The low salinity induces soluble sugars and inhibits starch content and thus extracellular saccharides and aminoacids secreted to the medium also provide protection to rice field cyanobacteria from NaCl stress (Padhi *et al*., 1998). In addition, the tolerance is also achieved by maintaining a low ion concentration inside the cell by the use of active transport mechanisms like a Na^+/H^+ antiporter (Packer *et al*., 1987), which is energized by the increased activity of the cytochrome oxidase (Molitor *et al*., 1986). Further consequences of a salt shock observed in cyanobacteria are the induction of stress proteins (Hagemann *et al*., 1991), the short decrease of photosynthesis directly after the shock and the increase of cyclic electron flow around photosystem I (Jeanjean *et al*., 1993). The salt tolerance exhibited by many cyanobacteria has been exploited with some success in reclamation of saline and sodic soils (Kaushik and Venkataraman, 1982). Hence the application of cyanobacterium*Westiellopsis prolifica* as biofertilizer to saline soil enhances the organic carbon content of soil due to the addition of enormous quantity of organic matter in the form of cyanobacterial biomass.

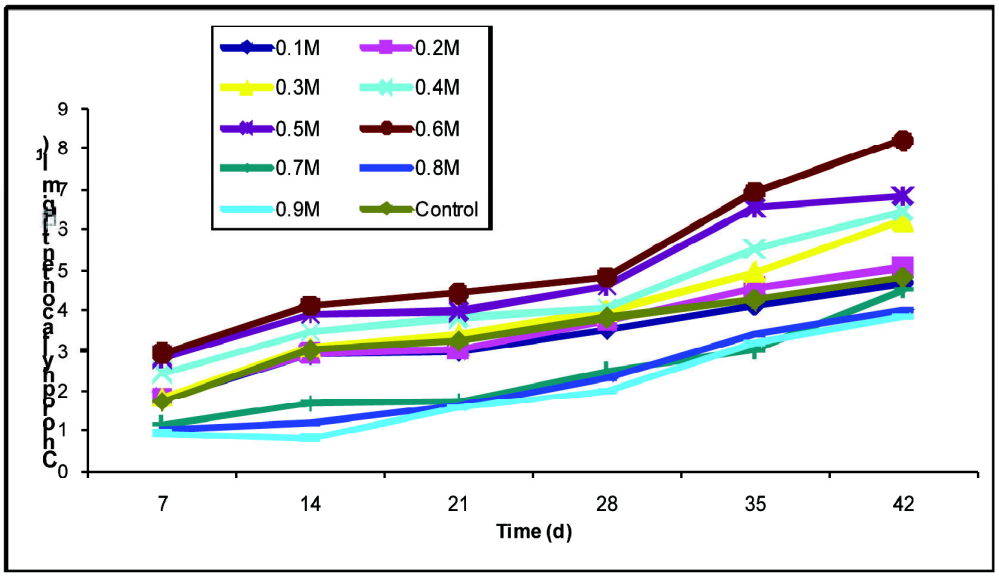
GRAPHS



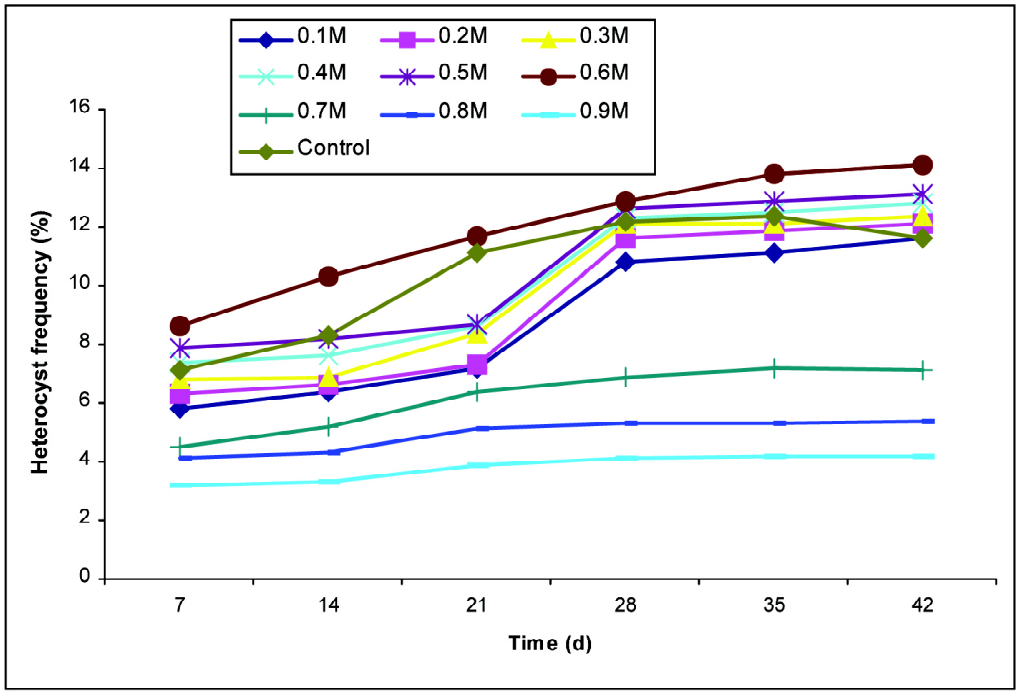
1. **Graph 1.** Effect of NaCl on the growth (absorbance of the culture suspension at 760nm) of *Westiellopsis prolifica* up to 42 days of incubation.



Graph 2. Effect of NaCl on protein content of *Westiellopsis prolifica* up to 42 days of incubation



Graph 3. Effect of NaCl on chlorophyll-a content of *Westiellopsis prolifica* upto 42 days of incubation.



Graph 4. Heterocyst frequency (%) of *Westiellopsis prolifica* at various concentration of NaCl upto 42 days of incubation.

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