

RESEARCH ARTICLE

Effect of salinity on biomass and biochemical constituents of *Spirulina platensis* (Geitler)

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ABSTRACT

A study was conducted to investigate the impact of NaCl on the growth and biochemical traits of *Spirulina platensis*. In order to determine the impact of NaCl, *Spirulina* was exposed to different concentrations of NaCl ranging from 0.1-0.4M besides control, over a period of 30 days. It was found that biomass, total chlorophyll, phycocyanin and lipid contents stimulated at lower concentrations of NaCl (0.1 and 0.2M) but reduced at higher (0.3 and 0.4M) concentrations. The total protein contents inhibited at all concentrations of NaCl. While β -carotene and carotenoid contents increased upto 0.3M and thereafter declined. The results indicated that *Spirulina platensis* showed diverse response to NaCl stress.

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INTRODUCTION

Although, many species of micro-algae are tolerant to great variations in salinity, their growth and chemical composition can be affected by the rate of respiration, distribution of minerals, ion toxicity, photosynthetic rate and permeability of cell membrane. The ability of microalgae to survive and flourish in saline environment under the influence of osmotic stress has received considerable attention (Sudhir and Murthy, 2004). Against these stresses, cells adapt themselves by undergoing different mechanisms including changes in morphological and developmental pattern as well as physiological and bio-chemical processes. Adaptation to stress is associated with metabolic adjustments which lead to the accumulation of several organic solutes and osmolytes. These osmotic adjustments protect sub-cellular structures and reduce oxidative damage caused by free radicals, produced in response to high salinity (Hiremath and Mathad, 2010). In this present study, an attempt has been made to investigate the effect of different NaCl concentrations on the growth and biochemical constituents of *Spirulina platensis*.

MATERIAL AND METHODS

Spirulina platensis was isolated from the alkaline lake samples of Kodaikanal (Tamil Nadu, India) using Zarrouk's medium (Zarrouks, 1966). In order to evaluate the effect of NaCl levels, *S. platensis* (10%) was added to 250 ml Zarrouks medium with different NaCl concentrations viz., 0.1 M, 0.2 M, 0.3 M and 0.4 M and control. The flasks were kept in shade net under diurnal natural light conditions. Cultures were analyzed for their growth and biochemical (nutrient and pigment) constituents on 30th day after inoculation (DAI).

For biomass content, a known volume of *Spirulina* was filtered in a pre-weighed filter paper and oven dried at 75 °C for 4 to 6 h, cooled and then again weighed. The differences between the initial and final weight were taken as dry weight. Protein content was determined as per Lowry *et al.* (1951), using bovine serum albumin as a standard. The total lipid content was determined from chloroform: methanol (1:2) (v/v) extract (Bligh and Dyer, 1959).

Chlorophyll-a was extracted in methanol as described by Vonshak (1997). For phycocyanin, the culture was precipitated using 50 per cent ammonium sulphate and the pigment was

extracted completely by repeated freezing and thawing (Boussiba and Richmond, 1979). Total carotenoids and β -carotene were extracted as described by Rafiqul *et al.* (2003).

RESULTS AND DISCUSSION

Cyanobacteria inhabit environments which vary drastically in their saline levels. In the last 15 years, many studies were published on the response of cyanobacteria to different saline environments; the specific role of organic compounds as osmoregulators, modification in photosynthesis and respiration activity and variations in the protein synthesis pattern. Different *Spirulina* species have been isolated from a variety of saline environments. In this study, various NaCl concentrations *viz.*, 0.1 M, 0.2 M, 0.3 M and 0.4 M along with control (0.25g) in Zarrouks medium were tried for the optimal growth and chemical composition of *S. platensis* and the results revealed that increase in NaCl concentrations decreased both the growth and chemical composition of *S. platensis*. The present results are in accordance with the findings of Rafiqul *et al.* (2003) who showed that under salt stress conditions, the growth rate of *S. platensis* was slower with decrease in chlorophyll, protein and phycocyanin contents while increase in carotenoids and lipid content.

The results revealed that the growth of *S. platensis* was significantly inhibited in salt stress compared to control. Exposure of *S. platensis* to high NaCl concentrations resulted in an immediate cessation of growth and a decrease in biomass (Fig. 1). An initial lag phase was observed before a new steady state growth was established. This lag period is associated with the decline in chlorophyll and biomass content due to inhibition of photosynthetic and respiratory system after exposed to high salt concentration (Vonshak, 1997). In the present study, total chlorophyll (Fig. 2) contents stimulated at lower concentrations (0.1 and 0.2M) of NaCl when compared to control but it was reduced at higher (0.3 and 0.4M) concentrations. According to Moradi and Ismail (2007), reduced chlorophyll contents at higher salinities are due to decrease in photosynthetic rate because of salt osmotic and toxic ionic stress. Many previous studies reported that the cultivation with higher saline concentrations had lower chlorophyll and protein contents (Vonshak, 1997). It has also been reported that chlorophyll is the primary target to salt toxicity limiting net assimilation rate, resulting reduced photosynthesis and reduced growth.

In regarding carotenogenesis, at higher NaCl concentration the grown cells contained higher amount of total carotenoids and β -carotene content (Fig. 1) with similar to previous studies. Pisal and Lele (2005) reported that β -carotene is a secondary metabolite and these molecules are produced by the cells in stress condition as cell protecting mechanism. There was an increase in total carotenoids and β -carotene content at higher saline conditions.

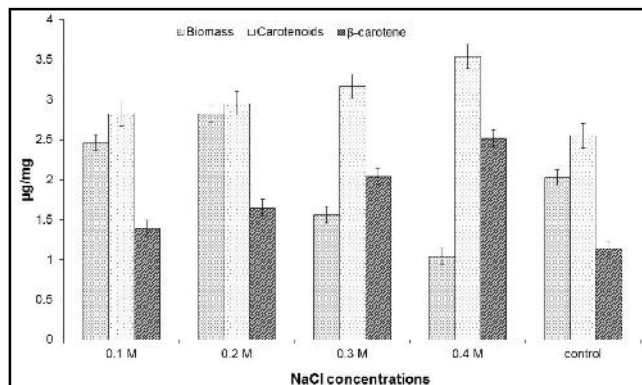


Fig. 1 : Effect of different NaCl concentrations on growth and carotenoids of *Spirulina platensis*

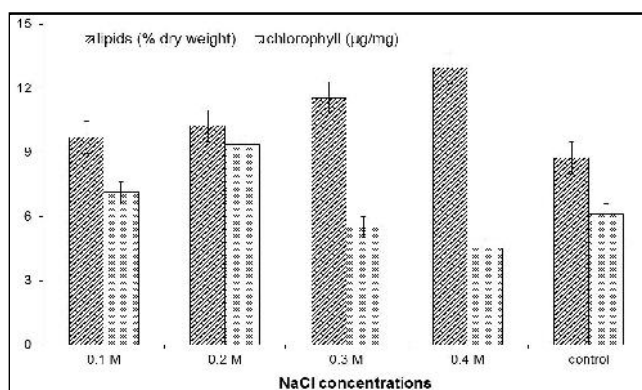


Fig. 2 : Effect of different NaCl concentrations on lipid and chlorophyll contents of *Spirulina platensis*

In addition, *Spirulina* exhibited decline in the total protein contents (Fig. 3) in all the concentrations of NaCl. This is because stress cells have a lower protein synthesis capacity increasing carbohydrate and lipid content. Infact while stimulation of carbohydrate biosynthesis by stress condition is well known, the increase in lipid synthesis has been seldom documented. The lipid accumulation in algae usually occurs during environmental stress, including growth under nutrient deficient conditions. The cells adapt themselves to stress by undergoing changes in morphological and developmental pattern as well as physiological and biochemical processes. The increase in salt concentration affects the rate of respiration, distribution of minerals, ion toxicity, photosynthetic rate and permeability of the cell membranes (Sudhir and Murthy, 2004).

The nutrient and pigment contents (Fig. 3) were also found to be decreased at high saline concentrations. Increasing NaCl levels at 0.3 M and 0.4 M led to decrease in the production of phycocyanin and soluble protein contents in *S. platensis*. The composition of phycocyanin was hanged markedly as results of increasing NaCl levels (Hanaa and Abd El-Baky, 2003). Decrease in the phycocyanin pigments

suggests that the cells may down-regulate their light-harvesting capacity to acclimate their low carbon metabolic capacity.

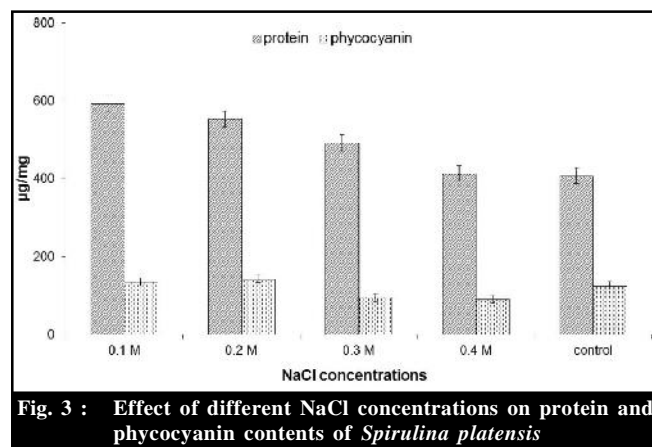


Fig. 3 : Effect of different NaCl concentrations on protein and phycocyanin contents of *Spirulina platensis*

The effect of various concentrations of NaCl on *S. platensis* showed reduced biomass and chlorophyll content mainly at 0.3 and 0.4 M and reduced protein and lipid content at all concentrations of NaCl. While β -carotene, carbohydrates and glycine betaine increased upto 0.3 M where as significant increase in the production of proline was observed at all concentrations of NaCl. These beneficial properties indicated that, adaptation of the alga to salinity was characterized by the accumulation of osmolytes.

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