CHAPTER – I

INTRODUCTION
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A wide range of micro-organisms like, cyanobacteria, eubacteria, fungi, eukaryotic algae etc. are able to synthesize and secrete exocellular polymeric substances (EPS), mainly of polysaccharidic nature. They remain covalently linked or loosely attached to the cell surface or released into the surrounding environment (Neu & Marshall 1990; De Philippis & Vincenzini 2003). Among all the microorganisms cyanobacteria are better suited than macro-algae or higher plants for EPS production since they exhibit higher growth rate and are more amenable for manipulation of conditions for enhancing growth, therefore can be exploited biotechnologically (Ozturk and Aslim 2010). Both unicellular and filamentous cyanobacteria produce exopolysaccharides which may form the compact structures intimately associated with the cell surface giving a characteristic cell shape or as amorphous mucilaginous mass that loosely attached to cells or suspended as colloids in the medium (Otero and Vincenzini 2003).

The microfossil records suggest that cyanobacteria or cyanobacteria-like prokaryotes were present on the primitive Earth in the Archaean era, more than 2.7 billion years ago (Schopf 2000). The exquisite preservation of these microfossils is thought to reflect the intrinsic stability of the exocellular polysaccharide (EPS) and its ability to bind heavy metals as well as to resist degradation (Horodyski 1992). More specifically these polysaccharidic investments have protective role against desiccation, acts as antibacterial agents (e.g. antibiotics, antibodies, bacteriocins, phages, phagocytic cells, and surfactants) or protects against predation by protozoans (Costerton 1981, 1987 and Whitfield 1988).

Cyanobacterial members are Gram-negative prokaryotes possessing the unifying property of performing oxygenic plant-like photosynthesis with autotrophy as their dominant mode of nutrition (Castenholz and Waterbury 1989). However, in spite of their typically aerobic photosynthetic nature, some of the cyanobacterial taxa can grow in the dark on organic substrates (Smith 1983; Stal and Moezelaar 1997) and others under anaerobic conditions with sulfide as electron donor for photosynthesis (Cohen et al 1986). Certain strains have the ability to fix atmospheric dinitrogen into organic nitrogen-containing compounds, so displaying the simplest nutritional requirements in comparison to other microorganisms (Fay 1992; Bergman et al 1997). Representatives of the group have been found frequently in most of the naturally illuminated environments examined so far, both aquatic and terrestrial, including several types of extreme environments. This widespread
distribution reflects a large variety of species, covering a broad spectrum of physiological properties and tolerance to environmental stress (Whitton 1992; Tandeau et al. 1993).

Owing to their ecological and biochemical diversity, cyanobacteria have been regarded as good candidates for various biotechnological applications. Possible exploitation of cyanobacteria has arisen in the last decade by the growing industrial interest towards polysaccharides production of microbial origin, that often show advantages over the polysaccharides extracted from plants or marine macroalgae.

It has been noticed that cyanobacteria from different environments produce different kinds of exopolysaccharides with a great variety of chemical structures and physiological functions. Therefore, a regular search for cyanobacterial strains for high production of new polysaccharides with potentially useful properties are to be conducted (Yunyi 2007). So far, more than 70 cyanobacterial strains have been shown to produce good yields of exopolysaccharides (De Philippis and Vincenzini 1998). It is interesting that most cyanobacterial polysaccharides are characterized by the presence of uronic acids, pentoses, polypeptide moiety or other non-saccharide components, such as organic (e.g. acetyl, pyruvyl and succinyl group) and inorganic (e.g. sulfate or phosphate group) substituent (De Philippis and Vincenzini 1998; De Philippis et al., 1998). Therefore, cyanobacteria may be regarded as a very good source of structurally diverse polysaccharide, and some of them may possess unique properties for special applications, being not fulfilled by the polymer currently available.

The possibility of stimulating polysaccharide release by means of optimisation of the culture conditions have also been studied, mainly devoted in assaying the effects of nutrient deficiencies, which have been shown to stimulate polysaccharide synthesis (Arad et al. 1988 1992; Kroen and Rayburn 1984)

Cyanobacterial polysaccharides also have biotechnological importance. Use of algal polysaccharide in food industry is very common. The sulfated polysaccharides, extracted from cyanobacteria, exhibit a potent broad-spectrum antiviral activity in vitro against HIV-1 and HIV-2, HSV, influenza and a series of other enveloped viruses (Luescher-Mattli 2003). The algal polysaccharides are reported to exhibit antiviral activity in vitro towards mumps and influenza B virus also (Gerber et al. 1958; Feldmann 1999). Ca-spirulan, a sulfated polysaccharide isolated from Spirulina has already been marketed. Other sulfated
polysaccharides like heparin, dextran sulfate, pentosan polysulfate and others show strong anticoagulant activities (Hayashi et.al 1996; Hayashi and Hayashi 1996).

With these background knowledge in the present investigation four cyanobacterial taxa viz. *Spirulina subsalsa, Anabaena spherical, Phormidium valderianum* and *Phormidium tenue* were chosen to study their exopolysaccharide production rate in different nutrient stress like excess and depleted conditions of nitrate, phosphate and NaCl in the experimental media in relation to other growth parameters and in altered physical condition of light intensity, temperature and airflow. Qualitative characterization of the extracted EPS has been done by NMR, methylation and GLC-MS studies. In most of the earlier studies EPS production were studied from soluble fraction of EPS that released in the media, but in the present investigation the bound EPS was also taken for analysis, as this fraction can be collected easily, therefore would be cost effective in further biotechnological exploitation.