CHAPTER 2

REVIEW OF LITERATURE
The Blue Green Algae (BGA), also known as cyanobacteria are the pioneer oxygenic phototrophs on the Earth whose distribution around the world is surpassed only by bacteria. Cyanobacteria and similar organisms produced most of the oxygen found in Earth’s atmosphere, which implies that early photosynthetic organisms would have lived in an atmosphere that was rich in CO$_2$ and poor O$_2$ (David et al. 2005). Fossil evidence points to their presence in geographically diverse regions during the Precambrian (2 or more than 3.5 billion year ago) (Schopf and Walter 1982). They are a large and morphologically diverse group of phototrophic prokaryotes, which occur in almost every habitat on the Earth. Their long evolutionary history has been marked by key geochemical and biotic transitions, including the creation of oxygenic photosynthesis (Holland 1977).

BGA have been proposed to be named ‘Cyanobacteria’ by some bacteriologists under the rules of International Code of Nomenclature of Bacteria (1978). They are microscopic in morphology; they show pigmentation and oxygen evolving photosynthesis in which photosystems, PS I and PS II are connected in series. The genome size of blue green algae, representative of all major taxonomic groups, lies in the range, $1.6 \times 10^9$ to $8.6 \times 10^9$ Da which is comparable to that of other Bacteria ($1.0$ to $3.6 \times 10^9$ Da) (Herdman et al. 1979). BGA are known to occur in oxic and anoxic environments. They are prokaryotic atypical cellular organisms, which share with the bacteria in cellular organization and occupy key position in biological evolution (Weller et al. 1992). Several species can switch to the typical bacterial anoxigenic photosynthesis using sulphide as electron donor; other species assimilate sugars and organic compounds in presence of light (Rippka 1972). In dark, blue green algae gain energy by respiring endogenous carbohydrate, which are accumulated in the light.
However, under anoxic conditions some species maintain this requirement by fermentation whereas in a few cases chemo organotrophy is found. Blue green algae can grow under very low water potential; such species can resist desiccation and grow in arid environments (Deserts) and/or can tolerate high salinity to grow in hypersaline ponds (Thajuddin and Subramanian 1992). It was presumed that cyanobacteria probably in the early Precambrian era were responsible for the significant increase in atmospheric oxygen (Brock 1978) as well as solar energy conversion for proteinaceous food production (Koening 1992). Many cyanobacteria have their ability to fix atmospheric nitrogen in most well oxygenated environments.

A number of BGA are rich in vitamins and many can excrete them into the surrounding environments. Some marine BGA are potential source for large scale production of vitamins of commercial interest such as vitamins of the B-complex group and vitamin E (Borowitzka 1995).

Analysis of extracellular growth-promoting substances liberated by *Nostoc muscorum* and *Hapalosiphon fontinalis* was found to contain amino acids like serine, arginine, glucine, aspartic acid, threonine, lysine, histidine and iso-leucine (Misra and Kaushik 1989a, 1989b).

Above all, under favourable conditions certain species of BGA can fix as much as 80 kg nitrogen per hectare per crop depending upon the ecological conditions (Roger and Watanabe 1986). Nitrogen accumulated through this symbiosis is released into the soil and made available to rice plants. Because of its high nitrogen fixing activity and unique property of being able to retain a significant amount of nitrogenase activity in the presence of combined nitrogen, making the system compatible with inorganic nitrogen fertilization (Ray et al. 1979). In tropical rice fields, biological nitrogen
However, under anoxic conditions some species maintain this requirement by fermentation whereas in a few cases chemo organotrophy is found. Blue green algae can grow under very low water potential; such species can resist desiccation and grow in arid environments (Deserts) and/or can tolerate high salinity to grow in hypersaline ponds (Thajuddin and Subramanian 1992). It was presumed that cyanobacteria probably in the early Precambrian era were responsible for the significant increase in atmospheric oxygen (Brock 1978) as well as solar energy conversion for proteinaceous food production (Koening 1992). Many cyanobacteria have their ability to fix atmospheric nitrogen in most well oxygenated environments.

A number of BGA are rich in vitamins and many can excrete them into the surrounding environments. Some marine BGA are potential source for large scale production of vitamins of commercial interest such as vitamins of the B-complex group and vitamin E (Borowitzka 1995).

Analysis of extracellular growth-promoting substances liberated by Nostoc muscorum and Hapalosiphon fontinalis was found to contain amino acids like serine, arginine, glucine, aspartic acid, threonine, lysine, histidine and iso-leucine (Misra and Kaushik 1989a, 1989b).

Above all, under favourable conditions certain species of BGA can fix as much as 80 kg nitrogen per hectare per crop depending upon the ecological conditions (Roger and Watanabe 1986). Nitrogen accumulated through this symbiosis is released in to the soil and made available to rice plants. Because of its high nitrogen fixing activity and unique property of being able to retain a significant amount of nitrogenase activity in the presence of combined nitrogen, making the system compatible with inorganic nitrogen fertilization (Ray et al. 1979). In tropical rice fields, biological nitrogen
presence of species of *Tolypothrix, Nostoc, Cylindropermum, Calothrix, Anabaena, Plectonema, Anabaenopsis* and *Schizothrix* are reported (Watanabe 1956, Kobayashi *et al.* 1967). In temperate soils BGA are less abundant than in tropical and subtropical region.

In India the BGA flora of rice soil have been studied by various workers (Tiwari 1972; Loloraya and Mitra 1973; Sarma and Kanta 1978a 1978b; Saha and Mandal 1979, Sardeshpande and Goyal 1981, Bangale 1984). Over 800 taxa belonging to 70 genera of BGA have so far been reported from Indian rice soil. Watanabe and Yamamoto (1971) in India found that in controlled growth *Tolypothrix tenuis* could fix nitrogen at the rate of 2000 lbs. per acre per year, thus potentially increasing rice production by about 20 percent. The production of rice has, therefore, considerably increased in recent years due to the artificial inoculation of BGA in the rice field. The international symposium on Nitrogen and Rice held at “International Rice Research Institute” (IRRI), Philippines in September, 1978 stressed the use of BGA and Azolla as a biofertilizer for rice cultivation. The “Central Rice Research Institute” (CRRI), Cuttack and “National facility for Blue-Green Algal collection”, Indian Agricultural Research Institute, New Delhi are conducting collaborative research and training activities to disseminate knowledge of BGA and Azolla and to encourage their uses as a source of nitrogen for rice cropping through out south and south-east Asia.

Assam is almost a virgin ground where very insignificant information of BGA are available. Bruhl and Biswas (1922), Parukutty (1939), Bordoloi (1974) had reported some information about the algal flora of the state. Devi (1981) put forwarded detailed information of algal flora of Darang district. Hazarika (1988) studied the distribution of BGA of rice field of Golaghat sub-division and reported the presence
of 81 species of BGA belonging to 21 genera from the rice field as well as other habitats including hot spring of Nambar. Altogether 82 species were identified by Deka and Bordoloi (1991), out of these 47 were non heterocystous. Saikia and Bordoloi (1994) had recorded only 28 species of BGA belonging to 12 genera from the rice fields of Barpeta, Nalbari and Kamrup district. Bhuyan (1996) made taxonomic and ecological studies on Myxophycean algae from Nagaon district. Talukdar (1997) explored blue green algal flora from Kamrup district and reported 152 species belonging to 52 genera and 14 families. Ahmed (1999) reported 64 species of BGA belonging to 15 genera from rice field soils of Nagaon sub division. Ahmed et al. (1999) reported a paper on distribution pattern of BGA in Nagaon sub division. Rout and Dey (1999) made a study on algal flora from rice fields of Irongmara (Barak Valley, Assam) and find the algal communities comprised with Chlorophyta, Cyanophyta and Bacillariophyta. Nandi and Rout (2000) reported 66 algal species from different habitats of Daragakona area, Silchar (south Assam); they found 53 species of cyanophyceae belonging to 18 genera. Ahmed and Kalita (2002) collected 53 species of BGA under 9 genera from Hojai sub division of Nagaon district. Chaudhury (2004) studied the taxonomy and ecology of tea garden soil algae of Assam with special reference to pesticidal toxicity. Saha et al. (2007) estimated a total of 29 species representing 18 genera under 12 families of epilithic cyanobacteria from Kakajian reserve forest of Assam.

2.2 NITROGEN FIXING POTENTIALITY OF BGA IN RICE FIELDS

The first report of nitrogen fixing potential of BGA in rice was presented by De (1939) and Singh (1942). A number of review reports are there which deal with the biochemistry and physiology of nitrogen fixation of BGA (Fogg 1949; Stewart 1966,

Watanabe et al. (1950, 1951) studied the nitrogen fixing ability of BGA in the rice fields in Japan. *Tolypothrix tenuis* was found to be the highest nitrogen fixer among other strains isolated from rice fields in Borneo (Watanabe 1959). The basic information on the biochemical mechanism of nitrogen fixation has come mainly from the studies using cell free extracts of nitrogenase from *Mastigocladus laminosus* (Scheider et al. 1960), which indicated the pyruvate as an essential requirement (Smith et al. 1971). The study made by Singh (1961), and Venkataraman (1972) revealed that the high density of BGA in Indian paddy fields showed the predominance of *Aulosira, Anabaena, Cylindrospermum, Calothrix, Nostoc* and *Tolypothrix*. Again in an analysis of BGA flora from rice fields, *Anabaena, Anabaenopsis, Aulosira, Cylindrospermum, Nostoc, Calothrix, Scytonema, Tolypothrix, Hapalosiphon, Mastigocladus, Stigonema, Westiella, Westiellopsis, Campylonema* were found to be dominant as nitrogen fixer. Stewart (1970, 74, 77) provided the first direct evidence of nitrogenase activity in heterocysts by acetylene reduction technique with isolated heterocysts supplied with ATP and Na$_2$S$_2$O$_4$.

Among the different culture media, the BG$_{11}$ media (Stanier et al. 1971) favoured maximum nitrogenase activity by cyanobacterial culture. According to Verma (1982) BGA can add about 20-25 kg/N/ha to rice fields and to that extent fertilizer can be saved or supplemented. Santra (1993) reported more than 100 species of BGA having nitrogen fixing ability. Roger and Reynaud (1982) revealed that only one third of BGA nitrogen was absorbed by rice plants in the first year and rest of it remained in the soil as residual nitrogen.
Heterocysts are distinctive and specialized cells of some filamentous members of cyanophyceae, which can be distinguished from the remaining cells by their thick walls, often with cyanophycean pigment at one or both end on the cell. The studies revealed that the non heterocystous BGA can fix nitrogen like heterocystous BGA (Fay 1984); but their ability become more widespread under microoxic conditions (Stal 2000). Heterocysts usually develop when concentration of combined nitrogen in the medium has been reduced (Fogg 1944; Castenholtz and Waterbury 1989). Again Stewart and Lex (1970) reported that non heterocystous nitrogen fixing blue green algae under certain ecological conditions showed widespread property of the cyanobacteria. Roger and Kulasooriya (1980) also studied so called auxinic effect of blue green algae mostly in vitro or in pot experiments. Sah (2008) reviewed the various physiological aspects of the nitrogen fixation and simultaneously oxygen evolving mechanisms of blue green algae. The review makes the understanding of cyanobacterial nitrogen fixation in the heterocyst easy, and also links the evolution of free oxygen (g) from the splitting of water by its vegetative cells and ultimately, the role of cyanobacteria in altering the primitive Earth’s atmosphere into present day oxygenating and thus making animal life possible on earth.

2.3. CULTURE MEDIA

For isolation and cultivation of BGA different types of culture media have been reported such as: Chu No. 10 (Chu 1942), modified Beneck’s medium (De 1939), Rodhe VIII (Rodhe 1948), Fogg’s medium (Fogg 1949); Gerloff’s medium (Gerloff et al.1950), Allen and Arnon’s medium (Allen and Arnon 1955), Kartz and Mayer’s media (Kratz and Mayer 1955), Provasoli’s medium (Provasoli et al. 1957), Mineral medium No II (Hughes et al. 1958), Synthetic medium for non-nitrogen fixing BGA
(Watanabe 1960), Bold Basal medium (Nichols and Bold 1965), medium for Spirullina (Tarrok 1966), Cg10 medium (Van Baalen 1967), medium for thermophilic BGA (Castenholtz 1970, Stainer et al. 1971), Mc Lachlan medium (Mc Lachlan 1973) and Woods Hole MBL (RRL, Guillard, quoted from Stein 1973). Above all, however most extensively used media are Fogg’s medium, Chu’s medium and BG11 medium.

2.4. PURIFICATION

Blue green algae in axenic culture have been isolated in solid media by standard plating technique or by taking advantage of properties such as rapid gladding motility and or large cell size. The preparation of solid media is one of the important step of isolation procedure. Concentration of agar in excess of 1.5% may be inhibitory to some cyanobacteria (Allen and Stanier, 1968). Mechanical damage and rapid drying during transfer of algal filaments for purification in solid media also observed (Bowyer and Skerman 1968).

Purification by an antibiotic treatment has been adopted by many workers (Spencer 1952; Provasoli 1958; Machlis 1962; Zatewaki and Provasoli 1964; Droop 1967; Venkataraman 1979). Ultraviolet irradiation has been widely used to obtain pure cultures of BGA (Gerloff et al. 1950; Koch 1964; Bowyer and Skerman 1968).

2.5 FACTORS AFFECTING GROWTH AND NITROGEN FIXATION

2.5.1. Light

The BGA are phototropic microorganisms and are usually located in the upper 0.5 cm of the soil (Roger and Kulasooriya 1980). Light availability, which influence BGA growth and nitrogen fixation, depends on the season, latitude, cloud cover, plant canopy and turbidity of water. The growth of BGA is better at higher light intensity.
Singh (1976) reported that field growth of *Anabaena fertilissima* was better under full sunlight. Watanabe *et al.* (1977) reported that under moderate light during wet season, nitrogen fixation was higher in basal soil than in planted soil. Roger *et al.* (1979) had also studied the influence of high light intensity on algal succession and reported highest nitrogen fixation. Roger and Reynaud (1982) reported rice canopy decrease the light availability of BGA by 50% after 15 days, 85% after one month and 95% after two months of transplanting. Nobel (1998) reported the growth rate of light limited cyanobacteria will depend primarily on their capability to capture light and the efficiency with which they use this light energy for growth.

2.5.2. Temperature

Optimum temperature ranges for BGA growth is about 30-35°C. Subramanium (1972) reported that cold temperature inhibited the growth of BGA in paddy fields of India. The lower temperature decreased productivity and favoured eukaryotic algae, whereas higher temperature favoured both phytoplankton and BGA (Roger and Reynaud 1977, 79b). Exposure to a temperature of 42°C for 50 minutes reduced photosynthetic activity of *Nostoc* and *Calothrix* to about 20% of the control (Venkataraman 1964). Singh (1976) reported that in Indian paddy field, high temperature (34 – 39°C) is favourable for the growth of *Aulosira fertilissima*. Venkataraman (1964) reported that exposure to a temperature of 42°C for 50 minutes reduced photosynthetic activity of *Nostoc* and *Calothrix* to about 20% of the control.

2.5.3. pH

pH is the most important factor, which determines growth, nitrogen fixation and distribution of BGA. The optimum pH for BGA growth in culture media ranges from 7.5 – 10.0 and lower limit is about 6.5-7.0. It has been reported that there is a positive
correlation between soil pH and growth (Okuda and Yamaguchi 1956; Singh 1984a; Roger and Reynaud 1977; Sardeshpandey and Goyal 1981) Venkataraman (1988) reported that BGA were relatively salt tolerant. The beneficial effect of pH on BGA growth can be demonstrated by the fact that addition of lime increased the BGA growth and nitrogen fixation (Okuda and Yamaguchi 1956; Yamaguchi 1976). However, reports also exist that some BGA can tolerate even lower pH viz. *Nostoc muscorum* and *Anabaena torulosa* had been found to grow at pH ranging from 5.0-7.0 (Durell 1964). Aiyer (1965) reported that *Aulosira fertilissima* and *Calothrix brevissima* had been found to be ubiquitous in Kerala rice fields, where pH ranges from 3.6-6.5. Madhusoodanan and Dominic (1999) also categorized some cyanobacteria from extreme environment. *Nostoc humiformae* showed maximum growth at pH 6.0. Mc Rae and Castro (1967) reported that high organic matter contents of the soil influenced the algal flora of BGA. Nitrogen fixation by *Calothrix* was slightly affected 1748 ppm of sodium chloride and severely depressed by 3396 ppm. Okuda and Yamaguchi 1952; Subrahmanian et al. 1965 studied the relationship between the physicochemical properties of the soil and response to algalization indicate that low pH limits BGA growth.

2.5.4 Fertilizers

Several microorganisms are known to be the important components of soil ecosystem which help the better crop nutrient management and maintenance of the soil health while working in a perfect harmony with the nature (Goyal 1993). Of the various soil ecosystems, BGA play a vital role in maintaining soil fertility and sustaining the crop yield even in the absence of any added nitrogen fertilizer in the water logged rice fields (Venkataraman 1981). Nitrogen fixing cyanobacteria have a major advantage
when sources of combined nitrogen are depleted from the water. Biological nitrogen fixation was known to be suppressed in the presence of exogenous sources of combined nitrogen. Chen (1983) showed the effect of nitrate, nitrite and ammonia on nitrogenase activity. *Scytonema schmidlei* did not show nitrogen fixation in presence of ammonium nitrogen (Bottomley *et al.* 1977; Kaushik 1987; Goyal 1993). Ammonium nitrogen has been proved to be toxic for growth of nitrogen fixing cyanobacteria at 75µg/ml (Stewart 1964; Goyal and Marwaha 1985). The effect of nitrate nitrogen on the growth, nitrogen differentiation and nitrogen fixation were studied (Mickelson *et al.* 1967; Pattnaik and Singh 1978; Kaushik 1987a). Soil fertility, specially organic carbon, nitrogen and available phosphorus play a significant role in growth and distribution of BGA in rice field soils (Kannaiyan 1985; Venkataraman 1988). Interaction of different soil samples from Bengal and Assam with tricalcium phosphate was studied and concluded that addition of superphosphate to rice soil would enrich the soil not only with phosphorous but also with nitrogen (De and Sulaiman 1950). The interaction of several species of rice field BGA with the commercial fertilizer was studied and observed the growth and heterocyst frequency (Singh 1975; Anand and Karuppusamy 1987; Goyal 1989; Prosperi *et al* 1993).

### 2.5.5 Effect of Agrochemicals

Predators, pathogens and antagonistic organisms limit the growth of BGA. Certain bacteria, fungi and viruses cause rapid of large BGA blooms (Singh 1973a; Roger and Reynaud 1979; Huang 1982; Grant *et al.* 1983a, 83b, 84, 85). Zooplankton, Cladocerans, Copepods, Ostracods and snails and mosquito larvae can damage BGA establishment. Pesticides have toxicity, which are selective for certain groups of microorganisms (Padhy 1985). Many of the studies on the effect of pesticides on
cyanobacteria have been conducted only in India (Venkataraman 1972; Adhikary 1989; Pabbi and Vaishya 1990; Sahu et al. 1992). This reflects the justifiable concern for the fate of nitrogen fixing cyanobacteria in pesticide treated paddy fields (Mishra et al. 1989).

Insecticides, which constitute 83% of total pesticides produced, have been studied quite extensively. Among the insecticides, organochlorine group is considered hazardous to these organisms due to their toxicity and persistence (Kar and Sing 1979a; Lal and Saxena 1980). Organophosphate insecticides are preferred by farmers for pest management practices for their biodegradability and non-persistency on soil. Effects of these agrochemical are reported by Gangawane (1979); Sardeshpandey and Goyal (1982); Kaushik and Venkataraman (1983); Subramanian et al. (1994). Carbamate group of insecticides have also been used and studied (Kar and Singh 1978, 79a; Adhikary et al. 1984; Vaishampayan 1985).

Fungicides also have potentially serious consequences on the overall productivity of soil by interfering with the activity of cyanobacteria. Research on the interaction of rice field cyanobacteria with fungicides has been undertaken (Moore 1967; Venkataraman and Rajyalakhami 1971, 72; Gangawane and Kulkarni 1979; Gangawane 1980). Many of the herbicides used are known to inhibit photosynthesis.

The growth, cellular nitrogen levels and tolerance of different herbicides in different concentrations were studied (Venkataraman and Rajyalakhami 1971; Khalil et al. 1980; Kapoor and Sharma 1980; Kashyap and Pandey 1982; Chinnaswamy and Patel 1983; Maule and Wright 1983; Pandey and Kashyap 1986; Mishra and Pandey 1989; Mishra et al. 1989; Nagpal and Goyal 1992). Toxicity of heavy metals in combination with pesticides was also investigated (Bisen et al. 1993).
2.6 PRODUCTION OF BGA UNDER NATURAL ENVIRONMENT

The potential significance of BGA in agriculture is obvious, therefore efforts were made in developing techniques for the mass production of BGA biomass (Ley 1992). Watanabe and Yamamoto (1970) and Venkataraman (1981a) reported that BGA could be produced in bulks with ease. Watanabe et al. (1969) developed methods for mass culturing of BGA in illuminated tanks by formulating mineral glucose liquid medium. Venkataraman (1979) and Venkataraman (1980) developed tank and pit method for individual farmers for mass production of BGA. Bisoyi and Singh (1988a) developed a simple method of large scale composite inoculum in the rice fields at the Central Rice Research Institute (CRRI), Cuttack. Venkataraman (1972) reported BGA production in shallow trays; varied from 0.4 to 1 kg dry weight/m² in 15 days. Roger and Watanabe (1986) reported technologies for utilizing nitrogen fixing organisms in lowland rice and the beneficial role of blue green algal inoculation in rice soils of Tamil Nadu (Kannaiyan 1985c).

The production technology of mixed algal inoculant in the laboratory as well as plastic sheet covering nursery beds in different parts of India has been studied under various environmental conditions season and also in various soil types (Goyal and Venkataraman 1971; Venkataraman 1972; Kannaiyan 1979, 1983). Kannaiyan et al. (1978) also developed technology for mass scale production of composite culture of blue green algae under rice field condition and the soil based BGA inoculum could survive for more than two years (Kannaiyan et al.1980). Nain et al. (2009) found positive interactions among the cyanobacterial strains and bacterial strains during pot experiments conducted with wheat variety.
2.7 SEASONAL INFLUENCE ON BGA

The climate of the different season is the most important factor, which determines maximum biomass yield and total N-content of cyanobacteria. Moor et al. (1995) and Shimada et al. (1996) reported that cyanobacteria might respond differently to various environmental factors because of their own physiological properties. Gupta and Agarwal (2006) studied the survival of terrestrial blue green algae with respect to environmental stress condition. They observed that Scytonema millei, Phormedium bohneri and Lyngbya mesotricha survived to 100 percent at atmospheric temperature of 5-35°C and relative humidity 55-100 percent in rainy, winter and spring season but the survival was 15-25 percent in summer when temperature reached 48°C and relative humidity was ≤23 percent. The reports of some earlier workers also exhibited the dominance of Anabaena, Nostoc, Calothrix and Aulosira in summer season on rice soils of India.

Nostoc and Anabaena appeared to be the most common algae found in rice soils of Maharashtra during the summer season (Mitra 1951, Singh 1978, Aiyer 1965, Chako 1972, Sinha and Mukharjee 1975, Singh 1978, Sardeshpandey and Goyal 1981). The study on survey of distribution of nitrogen fixing BGA found the predominance of Nostoc in rice field soil of Kerala, Tamil Nadu, West Bengal, Assam and Haryana during summer period (Singh et al. 1996, 97 and Venkataraman 1975).

Wiedner et al. (2007) also reported that environmental factors such as rainfall, temperature humidity and sunshine hour/day play important roles in relation to the growth of algae.
2.8 EFFECT OF ORGANIC COMPOST AS SUBSTRATE ON GROWTH AND N2-FIXATION OF BLUE GREEN ALGAE

The unique application of algal inoculation on the grain yield of many rice varieties has been demonstrated in different parts of the world. In general, these trials indicate that 10-15% increase in grain yield could be obtained through algal inoculation. On an average, there is contribution of 20-30 kg N/ha/season which means that chemical nitrogen fertilizer could be saved to that extent. Venkataraman (1975, 81) demonstrated the advantage of this technology is that farmers can produce the algal flakes for biofertilizer at their own fields and according to their requirements with bare minimum inputs. Beneficial effects of BGA inoculation have also been reported on a number of other crops such barley, oats, tomato, radish, cotton, sugarcane, maize, chilli and lettuce. Their residual effects on any crop that follows rice because of their role in improving soil health (Kaushik and Venkataraman 1979; Dadhick, Verma and Venkataraman 1969).

On reviewing the literature it is seen that tremendous works of BGA mass cultures in soil substrate have been done in India (Pabbi and Singh 2004). A major role of BGA comprises organic matter accumulation in soil which becomes available to the subsequent crops.

The importance of organic compost in agriculture is known since ancient times (Chhonkar 2003). The role of organic compost as a soil humus and plant nutrient in soil is well established. Besides, organic manure also improves physical, chemical and microbial properties of soil. Soil organic compost has a direct role on growth and metabolism of plants (Mathus and Gaur 1977). Though, relatively small in quantity compared to chemical fertilizers organic compost supply almost all the nutrients
Review of Literature

essential for plant growth. It also directly improves seed germination, water holding
capacity, drainage, base exchange capacity and reduce erosion. Organic compost for
substrate can be produced from different plant biomass, animal dung as well as other
agricultural wastes for growth of blue green algae. Such wastes undergo intensive
decomposition under thermophillic and mesophillic conditions in heaps or pits with
adequate moisture and finally converted into brown to dark coloured humified
material which is more stable with narrow C:N ratio.

The continuous use of chemical fertilizers over a long period may cause imbalance in
the microflora and thereby indirectly affect the biological properties of a soils
(Manickam and Venkataraman 1972). Benefits of compost as a substrate for growth
of BGA in mass culture technology mainly result from its content of organic matter,
plant nutrients, promoting plant growth and inhibiting root pathogens/ soil borne plant

Compost is known to be the product rich in micro organisms that helps the plants to
mobilize and acquire nutrients (Postma et al. 2003).

Temperature, oxygen supply, moisture, pH, available minerals and C:N ratio of the
plant residues are the direct environmental influence which determine the rate of
decomposition of organic matter. Environmental factors such as temperature,
moisture etc, is of much significance in decomposition (Bunnel and Tait 1977;
Meentemeyer 1978). High temperature and water potential are the most favourable
environmental conditions for decompositions of cereal straw (Summerell and Burgess
1989). Positive correlation between temperature, moisture and decomposition of
biowaste for compost preparation was reported by Statt et al. (1986).
The age of the plant, its lignin content and the degree of disintegration of the substrate presented to the microflora also govern the role of decomposition. In general organic matter with high C:N ratio decompose more slowly than those with a low C:N ratio (Parr and Papendick 1978). If the substrate has poor nitrogen content, decomposition is slow. Carbon mineralization is stimulated by supplemental nitrogen (Alexander 1961).

Nature of biowastes also govern the quality of organic manure. It is reported that the resource quality is a major regulatory factor in the rate of decomposition of biowaste. Plant residue with high nitrogen content had faster decomposition rate and nutrient release (Swift et al. 1979; Berendse et al. 1987). The C:N ratio, proportion of culture, hemi cellulose, lignin, temperature, moisture etc, are some of the factors that govern rate of decomposition. As these organic constituents are more resistant to microbial activity, increase in their content decreases the rate of decomposition (Mellilo et al. 1982; Berendse et al. 1987; Gaur 1994). Bajtehchi et al. (2007) found that cyanobacteria were considered as compost supplier for urban garbage composts. They had significant effect on growth rate and nitrogen content on urban garbage composts. Markou and Georgakakis (2010) demonstrated the cultivation of microalgae using wastes and wastewaters for biomass production, treatment of the wastes and wastewaters because they produced biomass in satisfactory quantity and can be harvested relatively easily due to their size and structure.

It is reported that addition of a nutrient rich wastes like neemcake, cattle dung, bio activators etc. can speed up the decomposition of cereal straw by lowering C:N ratio (Gaur et al. 1992). Dutta et al. (1999) observed that recycling and reuse of coir pith to promote plant growth in crops such as cowpea, green gram, cluster beans etc. was
found to be useful to enhance crop and soil productivity. Malliga et al. (2007) reported that *Anabaena azollae* while being used as biofertilizer exhibited lignolysis and released phenolic compounds which induced profuse sporulative of the organisms. They also reported the usefulness of coir waste as carrier for cyanobacterial biofertilizer with supporting enzyme studies on lignin degrading ability of cyanobacteria and use of lingo cellulosic coir waste as an excellent and in expensive carrier for cyanobacterial biofertilizer. Chu-Tung lin and Chi-Ting Huang (1980) demonstrated the effects of decaying rice straw on growth and nitrogen fixation of a blue green algae. Palaniappan et al. (2010) describes the effect of a coir pith immobilized formulation of the cyanobacterium Phormidium and its aqueous extracts on the germination and growth of cowpea. Evaluation of the various concentrations of the aqueous extract obtained from a 30 day old immobilized culture of the cyanobacterium revealed that 5% aqueous extract significantly improved the germination of cow pea seeds under *in vitro* conditions. It is also reported that the application of immobilized cyanobacterial formulation also increased the seed germination, plant height, plant weight, number of flowers, root nodules and biomass over control.

2.9 BGA BIOFERTILIZER AND BIO-CARRIER MATERIALS

Algalization technology was developed for the welfare of small and marginal farmers. This technology was based on the ecology and potency of microorganisms and could be adopted by the farmers for production of BGA and enhancing paddy productivity. Therefore, efforts were made in developing techniques for the mass production of BGA biomass since 1958 (Ley 1992). Watanabe et al. (1959) developed methods for mass production of BGA in illuminated tanks by formulating mineral glucose liquid
Venkataraman (1972) reported BGA production in trays varied from 0.4 - 1.0 kg dry weight/m² in 15 days. Depending on requirement, four methods viz. tank method, pit method, field method and nursery-cum-algae method were developed (Venkataraman (1918); Bisoyi and Singh (1988a) reported a record production 15 tonnes/ha of BGA in field method within three months in certain localities. Begum et al. (2011) reported impact of blue green algal inocula and urea -N on the growth and yield components of two HYV; of Rice (BR-28, BR-29) variety. They found that application of cyanobacterial composite inoculum produced significantly higher number of tillers, panicles, weight and yield of grain and straw as compared to the control. Tiwari et al (2010) found that exploitation of a potential starter culture of cyanobacterial biofertilizer contribute nitrogen for the growth of the crop in various ways. Several cyanobacteria such as Anabaena, Nostoc, Cylindrospermum, Aulosira, Tolypothrix, Gloeotrichia, Wollea etc are potent free nitrogen fixers (Dart and Day 1977; Peters 1978).

The production technology has been substantially improved with introduction of new and cheap carrier materials that support microbial load with longer shelf life thus considerably reducing the quantity of inoculum per unit area. Peat as carrier material was recognized in 1948, use of Indian peat as carrier was reported by Ishwaran (1969), charcoal was used as carrier in India in 1951 and combination of porous gravel with wholly fused magnesium phosphate is suitable carrier for long distance transportation (Watanabe et al. 1969). Use of coal as alternative carrier to peat was reported by Dubay (1975). Straw has been used successfully as a carrier material. Shanmugasundaram (1996) prepared the biofertilizer inoculum ‘Cyanostraw’ by growing the desired algal strains along with the carrier in polythene bags under...
controlled conditions to get a product of high quality, free from soil fungi, protozoan or contamination algae. Another method involves the growth of algae in an indoor production unit that may be a polyhouse or glasshouse. Once fully grown, the culture is harvested, mixed with the carrier material i.e. rice or wheat straw and sun dried (Kaushik 1996). Multani mitti (Fuller’s earth) has also recommended as a carrier material for the purpose. The harvested wet biomass is mixed with equal amount of multani mitti. The algal-mitti paste is sun dried and ground (Goyal et al. 1997). The technology has been further refined by growing the culture in a polyhouse under semi-controlled conditions for round the year production (Pabbi et al. 2000). The growth in a polyhouse is faster. Kannaiyan et al. (1998, 2000) has shown that cyanobacterial cultures immobilized in PUF (Polyurethane foam) and PVF (Polyvinyl foam) increase the quality and viability of cyanobacteria and could survive for a period of 2 to 3 years. The literature also suggest the use of other substrate like sugarcane waste, coir waste, paper waste, hollow fibres, rice bran etc. (Aruna and Kannaiyan 1998; Suresh Babu and Kannaiyan 1998; Hall et al. 1998; Malliga and Subramanian 2001; Kumar 2003). Despite all the advantages and simplicity of algalization technology, it has not become popular with the farmers (Pabbi and Kaushik 1997). There are a number of technical, environmental and socio-economical constraints that limit wider adaptation of this technology and problems are multifaceted (Singh and Pabbi 2001). Dhar et al (2007) carried out field experiments to compare the efficiency of two newly developed carrier based blue green algal biofertilizers (wheat straw and multani mitti), with the traditional soil based BGA biofertilizer, on the grain yield of rice for a period of three years. They reported that straw based and soil based biofertilizer treatments showed highest yield in rice.
Hegazi *et al.* (2010) found that application of cyanobacterial inoculant enhance the soil biological activity in terms of increasing the total bacterial, total cyanobacterial counts, CO$_2$ evolution, dehydrogenase and nitrogenase activities. They suggested that $\frac{1}{4}$ or $\frac{1}{2}$ of the recommended dose of nitrogen mineral fertilizer could be saved by using some species of nitrogen fixing cyanobacteria. Although the rice field soils may have some of these beneficial algae, artificial inoculations ensure the establishment of most promising strains employed as inoculants in addition to their nitrogen fixing ability. The strains suitable for one agroclimatic area may not be suitable for other agroclimatic region. It is essential to isolate and select promising isolates of BGA in order to have a germplasm collection of ecologically adopted, efficient BGA with their known biology (Sardeshpandey and Goyal 1981; Kolte and Goyal 1985; Kannaiyan 1985c, 1990; Gopalaswamy 2001). The report of Bhuvaneshwari *et al.* (2011) confirmed the comparative application of Cyanopith and Cyanospray like bio fertilizers with chemical fertilizers increased the growth parameters on sunflowers. Such kind of use of cyanobacterial inoculants enhance the nitrogen status of irrigated plantation crops also (Abd-alla *et al.* 1993).