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The Role of Cyanobacteria in Agriculture

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Abstract—*Microorganisms offer a pool of properties which could be utilized for the benefit of the ecosystem and mankind. They have always played the central role in evolution and shaped out the existing life on earth. The population increase needs to keep pace with the agricultural front to meet the demands with supply. As the crops and other plants are sessile they have to combat biotic and abiotic stresses. The stressful environmental conditions including the stress, soil fertility issues, presence of pathogens etc. can hamper the growth and development of the plants and affect the crop productivity. These conditions could be regulated by using fertilizers specially biofertilizers, as the chemical fertilizers are effective but have deleterious effects on the ecosystem. This article focuses the role of cyanobacteria in agriculture to improve the crop yield. These photosynthetic prokaryotes fix the atmospheric nitrogen into utilizable form and make it available to the plants. They also form symbiotic associations and provide nourishment to the host and in turn get housing either endophytic or exophytic. The growth enhancement was observed when cyanobacteria were inoculated in the fields. This effect was due to hormones like cytokinin, gibberellins and auxin which accelerated growth. Elicitor molecules from these organisms like certain peptides, vitamins, carbohydrates are reported to induce pathogenesis in plants. The exo-polysaccharide of cyanobacteria improves the soil quality and fertility. The recent trend involves the manipulation of the higher plants with the genes from these organisms to improve their production and stress related properties.*

1. INTRODUCTION

According to United Nations estimates, the global human population is projected to reach 8.9 billion by 2050, with the developing countries of Asia and Africa to absorb the vast majority of the increase [1]. Meeting up the demands of ever increasing population creates a pressure on the agricultural front and other related industries to scale up their production. This invites the excessive use of the chemical fertilizers which are effective for short term but have deleterious long term effects on the human health as well as on the ecosystem. Plants being sessile have to face adverse and stressed conditions which affect their productivity. Hence biofertilizers offer an ecological as well as economical substitute for chemical fertilizers. The production could be effected by various kinds of biotic and abiotic stresses like salt, osmotic, pH, temperature etc. "Increased salinization of arable land is expected to have devastating global effects, resulting in 30% land loss within the next 25 years, and up to 50% by the year 2050" [2].

The phyllosphere, rhizosphere and endosphere house a large diversity of microbiota some of which are positive contributors and some are harmful for the growing plantation. Agriculture could be improved and enhanced by the use of variety of microorganisms. These organisms can contribute in a number of ways. The main application is in the form of biofertilizers and green manure. They have been widely used as biocontrol agents. Some of the secretary products act as elicitor molecules. They are also symbionts with the eukaryotic plants which could be an endophytic or an exophytic association. These mainly help in the procurement of basic elements like nitrogen, carbon, sulphur, phosphorus etc. Apart from the application of microorganism in the fields and their symbiotic association, the recent budding technology is the genetic manipulation of eukaryotic plants by introducing the prokaryotic genes to either make them stress resistance or to improve their photosynthetic rates and ultimately their production.

2. BIOFERTILIZERS

Biofertilizers being eco friendly could support the sustainable development and contribute in enhancing the production. They not only help to maintain the natural soil habitat, but also improve the fertility. Soil and crop management practises such as crop rotation, soil fertility restoration, biocontrol of plant diseases etc. used adequately can add up for improving the production rate along with the biofertilizers use. The properties which make an organism to become good biofertilizer are- non-pathogenic, easily cultivable, less input requirements, fixing free nutritional forms in to fixed utilizable state, capable for outdoor/field survivals.

2.1. Potential category of Biofertilizer and Biopesticide organisms

The microbial populations which could be used as a potential fertilizer and as a biocontrol may contain a specific population or a combination of two or more organism. This may consist of plant growth promoting Rhizobacteria, N_2-fixing cyanobacteria, Actinomycetes, soil toxicant degrading microbes, plant disease suppressive bacteria and fungi and other useful microbes. Based upon the source of utilization they can be categorized as- Nitrogen fixers, Sulphur solubilizers, Phosphate mobilizers, Phosphorus solubilizers, Plant growth promoters etc.

3. CYANOBACTERIA- THE GENERAL CHARACTERISTICS

Cyanobacteria are one of the most primitive photosynthetic prokaryote with minimum nutritional requirements for growth. They hold the record for being the oldest fossil dating back to 3.46 billion years ago, found in sedimentary rocks of west Australia [3, 4].

Their habitat could vary from garden soil, rock, and desert to marine water bodies, freshwater bodies, lakes, rivers, salt marshes, swamps etc. Majorly all of the cyanobacteria are either alkalophilic or alkalotolerant along with or without halophytic characteristics. They have a striking property of survival under high pH and salt concentration [5]. They are capable of tolerating extreme environment of desiccation, light intensity, temperature, pH, salinity and nutrients [6] Cyanobacteria are one of the widely used organisms for organic biofertilizer. After water, nitrogen is the second limiting factor for plant growth in many fields and efficiency of this element could be met by these biofertilizers [7]. They harness the natural and simple inputs like solar energy, water, nitrogen and ensure the soil fertility, thus improving and contributing to the plant growth.

These organisms act as soil conditioners, soil, waste land and water body ameliorants and heavy metal scavengers. They are one of the excellent sources of vitamins, minerals and essential amino acids. They have a great economic importance and industrial application in the areas of food, feed, agriculture, natural pigments, aquaculture, bio-fuels, cosmetics and pharmaceuticals [8, 9, 10].

4. THE BRILLIANT NITROGEN FIXERS

The importance of the blue green algae on the nitrogen economy in Indian agriculture was studied by Singh in 1961 focusing on the paddy cultivation [11]. Watanabe et al were among the earliest groups who devised various methods for mass production of cyanobacteria to be used as biofertilizers. They also proved the non-symbiotic nitrogen fixing properties of these Myxophytes in paddy, which lead to 25% increase in grain yield [12, 13, 14]. Application of blue-green algae on a mass scale in the fields as a biofertilizer was coined as “algalization” by Venkataraman, 1961 [15]. If inoculation of cyanobacteria is done for 3-4 crop season repeatedly, there are chances that they can institute themselves permanently [16]. The main nitrogen fixers in the phyla cyanobacteria are *Anabaena*, *Nostoc*, *Trichodesmium*, *Calothrix*, *Cyanotheca*, *Aulosira*, *Plectonema* and *Tolypothrix*. According to a study of diazotrophy by these organisms in subarctic floodplains, the statistics indicate an increase in the productivity, which was an indirect result of nitrogen fixation. A diversity of blue-green algae was found colonizing the detritus surface in the form of biofilms [17].

Mechanism of nitrogen fixation is enzyme mediated phenomenon, the major role being played by nitrogenase. Since the enzyme is oxygen sensitive, it is protected by the superoxide dismutase activity along with high respiration rate. The filamentous, multicellular forms have spatial separation for nitrogen fixation. Specialized cells called heterocyst are dedicated for nitrogen fixation and vegetative cells perform photosynthesis and other cellular activities [18]. Heterocysts are the thick walled cells which have different morphology and physiology, performing specialized function. In unicellular forms there is a temporal separation. The uptaken nitrogen fixed as ammonia is available to the crops either by the exudates from the organism, autolysis and after the death and from the decaying body. Algalization has been extensively practiced in the paddy as the water is available which increases the working efficiency and exudates release by these Myxophytes. Other plants apart from rice are- tomato, barley, oats, sugarcane, wheat, maize and cotton [19, 20]. Besides fixing nitrogen they also serve as the reservoir of phosphorus, sulphur, carbon and other micronutrients. 5-35% increase in the organic content of field was observed by the cyanobacterial inocula [21].

5. SYMBIOTIC ASSOCIATION

In the symbiotic association cyanobacteria provides nitrogen to the host, fixed carbon is provided in case the host is non-photosynthetic. Cyanobacteria establish association with variety of hosts like fungi, protists, sponges, bryophytes, pteridophytes, gymnosperms, angiosperms and animals. These kinds of associations maintain an overall nutrient and energy account of any area and directly or indirectly play a major role in the agricultural practices. A marine species, *Richelia intracellularis*, may be found within the cells of the diatom *Rhizosolenia*. They are mainly found in the northern Arabian Sea and are the main regulators of the nitrogen pool of that zone along with the energy and nutrient regulation [22]. Many other reports suggest the association of the diatoms with these blue-green algae [23, 24]. Cyanobacterial association with bryophytes is rare. Only four associations were found out of 340 liverworts [25]. In the 13 genera of hornworts such association was omnipresent [26]. Slime cavities in the gametophyte of bryophytes like hornworts and liverworts are occupied by cyanobacteria [27]. The cyanobacteria-bryophyte association is a key regulator of the carbon pool along with being a litter producer and thermal insulators in the northern globe ecosystem. This symbiotic association affects the net primary productivity and heterotrophic respiration [28]. In sub-Antarctic region on Marion Island, 19 bryophytes with epiphytic cyanobacteria were found to reduce acetylene in field [29]. Water fern *Azolla* forms a symbiotic association with *Anabaena*, in which the cyanobacterium fixes nitrogen for the plant and acquires carbon and protected environment in return [30]. This fixed nitrogen becomes available when *Azolla* decomposes [31].

Mature leaf of *Azolla* house *Anabaena* in the ellipsoidal cavities located on the dorsal side. Both organisms carry out photosynthesis, *Azolla* being C3 plant has higher photosynthetic rate. *Anabaena* has a lower PSII rate [32]. This complex proves to be as the best biofertilizer and green manure. It has also been used for bio-gas production, weed control, mosquito repellent, water purifier, human and animal feed.

Among gymnosperms cyacads are the only ones which undergo a symbiotic association with the blue-green algae. The specialized coralloid root is the site for the symbiotic partner [33]. *Zamia furfuracea* has *Nostoc* as its cyanobiont [34]. Studies done so far suggest that a single population of cyanobacteria occupies the coralloid root of the cyacad. STRR-PCR DNA Fingerprinting results stated that different strains might occupy a single coralloid root [35]. The *Gunnera-Nosto*c is the only known symbiosis of blue-green algae with angiosperm. Housing of *Nostoc* is on the shoot-associated glands [36, 37].

6. ELICITOR MOLECULES OF CYANOBACTERIA FOR PLANT GROWTH ENHANCEMENT

Hormone production and their physiological effect is basically a eukaryotic trait. Certain microbes are reported for the production of plant hormones like cytokinin, auxin and gibberellins. Many recent reports and findings strongly support the production of hormone(s) by cyanobacteria and their stimulating effect on plants. Indole-3-acetic acid (IAA) production by 34 different free living and symbiotic cyanobacterial strains was analyzed, which suggested 38% of free living and 83% symbiotic isolates were capable of IAA production [38]. *Phormidium sp*. MI405019, which is a mangrove root-associated cyanobacteria, was reported to produce IAA. Analysis of the extracellular extract from this blue-green algae was done to study its effects on seed germination and callus differentiation of tobacco [39]. The cyanobacteria in paddy are capable of synthesizing and liberating gibberellic acid for the growth of plant. The gibberellins produced by the cyanobacteria are capable to overcome salt stress in rice plants. Thus apart from growth enhancement, hormones could help in overcoming salinity stress [40]. Cytokinin isopentenyladenine was isolated and identified from a cyanobacterial strain of *Arthronema africanum* using biological chemophysical techniques [41]. Elicitor molecules from cyanobacteria are known to enhance the production of secondary metabolites in plant cell cultures. Majority of these elicitor molecules belong to the groups of carbohydrate, vitamins, proteins, lipids, glycolipids or glycoproteins. These molecules are known to impart beneficial effects on the plants by providing resistance towards biotic and abiotic stresses [42, 43, 44].

Azadirachtin, an important biopesticide, is synthesized in Neem seeds. Cyanobacterial elicitors derived from *Anabaena sp*. and *Nostoc carneum* were treated with the Neem cell suspensions containing *Azadirachtin*. These trials lead to the five-fold increase in the alkaloid production [45, 46]. Phycocyanin is a natural blue pigment that is the major light-harvesting biliprotein in the blue-green alga *Spirulina platensis*. This particular compound was used as an elicitor to enhance the accumulation of capsaicin and anthocyanin in *Capsicum frutescens* and *Daucus carota* cell cultures respectively [47].

One study was conducted wherein hybrids of maize plants were inoculated with different cyanobacterial formulations, in order to evaluate the defense enzyme activity, Zn concentration and soil health. *Anabaena-Azotobacter* biofilm and *Anabaena spp.-Providencia sp.* enhanced the activity of peroxidase, PAL and PPO in roots [48].

Certain elicitors from the blue-green algae are capable of inducing anti-pathogenic responses. Extract of ***Anabaena*** sp. BEA0300B strain, when treated for *Zucchini* leaves, induced systemic defense responses on plants. Accumulation of Chitinases, ***β***-1,3-glucanase and peroxidases was observed in the leaves after treatment. Furthermore, constitutive acidic isoenzymes were increased upon treatment. Hence, this extract showed an indirect antifungal activity by inhibiting the pathogen sporulation [*49*].

7. CROP IMPROVEMENT USING CYANOBACTERIAL TRANSGENICS

The autotrophs such as microalgae and cyanobacteria are more efficient in performing photosynthesis when compared with C3 and C4 plants, this is because of two parameters: **action of carbonic anhydrase (CA), both extracellular and intracellular, and the CO_2 concentrating mechanisms (CCM) [50].** The CCM mechanism works efficiently as it is capable of accumulating CO_2 at carboxylation site. Many structural genes have been identified which are essential for the CCM, yet incomplete information is available regarding their regulation. “The transcriptional regulators, CmpR (Sll0030), CcmR (aka NdhR, Slr1594), and Sll0822 are implicated in the control of expression of the low carbon (LC) inducible genes of the CCM” [51].

The genetic approach was tried mainly in wheat and rice for improving their photosynthesis and water-use efficiency (WUE). Components of the carbon concentrating mechanism were integrated in the genome of the C3 plant chloroplast. “Mathematical modeling indicates that improvements in photosynthesis as high as 28% could be achieved by introducing these two genes- *bicA* and *sbtA*, which are cyanobacterial bicarbonate transporters, in to C3 plant chloroplasts” [52]. Expressing cyanobacterial fructose-1,6-biphosphatase and sedoheptulose-1,7-bisphosphatase show enhanced photosynthetic efficiency and growth characteristics[53].

The response of plants to different stress conditions varies, and different kinds of stress often lead to identical or similar responses. At the cellular level, abiotic stresses, especially water deficit (drought and salinity) causes a decrease in pressure potential. Certain groups of cyanobacteria are recognized on the basis of their organic osmotica and upper salinity limit for growth. They have been known to accumulate disaccharides, betaines, sucrose synthase (SS) and synthesize heteroside glucosylglycerol in response to salt stress [54, 55]. ***Nostoc muscorum***, follows certain mechanisms which leads to salt tolerance in the cell. The primary mechanism is a combination of higher photosynthetic activity with accumulation of sucrose for the purpose of osmoregulation. The secondary mechanism is Nitrogen fixation and protein synthesis [56]. Such organisms which have robust machinery to work under biotic stress can be used in fields to support the agriculture and improve soil quality. The specific genes synthesizing the osmoregulators can be transferred to higher plants and expressed.

The Δ12-acyl-lipid desaturase gene from the cyanobacterium ***Synechocystis*** sp. PCC 6803 was inserted into the leaf cells of the potato plant. This transformation stabilizes the composition and physical properties of biomembranes by promoting polyunsaturation of fatty acids, which averts the accelerated generation of O_2 and suppresses lipid peroxidation during hypothermia. These changes in the membrane provide cold resistance to potato plants, as membranes play important role in stressed conditions [57].

At present many trials are going on to develop robust and healthy plants which could tolerate the extreme environments utilizing the cyanobacterial genes.

8. CONCLUSIONS

Cyanobacteria play a promising role in the field of agriculture. They are one of the best substitutes to the chemical fertilizers. These photosynthetic prokaryotes are capable to play multiple roles at a time in order to improve the crop productivity, maintaining the soil integrity and fertility, preventing the colonization of the pathogenic microorganisms, stimulating growth in the plants. They are cheap, easily cultivable with minimum input requirements. Hormone production by the blue green algae can prove beneficial to improve and raise the plants naturally. Some of these hormones are also effective in overcoming stress and helping in seed germination. They have excellent photosynthetic capabilities which could be extrapolated to the higher plants through recombinant technologies leading to the production of transgenic varieties with enhanced production abilities. Apart from fulfilling the macronutrient and micronutrient requirement, cyanobacteria also secretes certain exudates which directly or indirectly help in plant growth promotion. The polysaccharide secretions from the cyanobacteria improve the soil efficiency by binding the particles together, adding organic material, preventing soil erosion etc. Thus they provide multidirectional benefits towards agriculture.

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