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**INFLUENCE OF CYANOBACTERIA BASED INOCULANTS AND
CULTIVATION METHODS ON RICE (*Oryza sativa* L.) YIELD,
SOIL CARBON CONTENT AND AGGREGATION**

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CULTIVATION METHODS ON RICE (*Oryza sativa* L.) YIELD,
SOIL CARBON CONTENT AND AGGREGATION**

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This is certify that the thesis entitled **“INFLUENCE OF CYANOBACTERIA BASED INOCULANTS AND CULTIVATION METHODS ON RICE (*Oryza sativa* L.) YIELD, SOIL CARBON CONTENT AND SOIL AGGREGATION”** submitted to faculty of the Post-Graduate school, Indian Agricultural Research Institute, New Delhi, in partial fulfilment of the requirements for the degree of **Master of science in agronomy** by **Mr. Amit Anil Shahane**, Roll No. 20038 embodies the results of the bonafide research work carried out by him under my guidance and supervision. No part of the study reported here has so far been submitted anywhere for the publication or for any other degree or diploma.

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Dedicated to ...



*My Beloved
Family*



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ACRONYMS

g	: gram
Kg	: Kilogram
µg	: microgram
Mg	: Mega gram
ha	: hectare
cm	: centimetre
m	: Meter
m ³	: cubic meter
q	: quintal
t	: tones
N	: nitrogen
P	: phosphorous
ppm	: parts per million
K	: potassium
hr	: hours
RH	: relative humidity
Rs. /ha	: Rupees per hectare
T	: temperature
max	: Maximum
Min	: Minimum
EP	: Evaporation
AWS	: Average wind speed
Met.	: Meteorological
Ave.	: Average
DW	: Distilled water

1. INTRODUCTION

Rice (*Oryza sativa* L.) is a staple food of more than 50% of the world's population (Fagaria, 2007) and supplies 20% and 31% of total calories required by world and the Indian population, respectively (Zeigler and Barclay, 2008). Agricultural Policy Vision 2020 of Indian Council of Agricultural Research, India has projected 112 million tonnes of rice requirement in 2020, which is 23 million tonnes more than the present rice production (Mahajan *et al.*, 2012). Additional rice will have to be produced from the existing rice area (41.9 million ha.). North West (NW) India covering states of Punjab, Haryana and western Uttar Pradesh are major contributor of food grains procurement by Government of India for its public distribution system (Prasad and Nagrajan, 2004). But during the past 30 years or so the question of production sustainability in this canal and tube well irrigated area has been raised and there are sign of fatigue and decline in productivity. The sustainability of rice production in this area is most threatened by scarcity of water.

With such practices, the rice crop consumes large quantity of irrigation water, ranging between 1500 and 3000 mm (Sharma *et al.*, 2002). So there are twin challenges of producing more rice per unit area with less water as it is crucial for food security in many Asian countries including India. The traditional method of rice cultivation requires a large amount of labour, water and energy. Water and labour, however, are becoming increasingly scarce in the NW India, raising questions about the sustainability of rice production and the overall environment. In this part of country, increasing use of groundwater for rice cultivation has led to a decline in the water table by 0.1 to 1.0 m year⁻¹, resulting in water scarcity and increased cost for pumping water (Hira, 2009; Rodell *et al.*, 2009; Humphreys *et al.*, 2010; Mahajan *et al.*, 2012). Implementation of the Mahatma Gandhi National Rural Employment Guarantee Act, introduced by the Indian government in 2005 (GOI, 2011), promising 100 days of paid work in people's home village, has been creating a labour scarcity in Punjab, Haryana and western Uttar Pradesh as rice transplanting in this region is mostly dependent on migrant labourers from eastern Uttar Pradesh and Bihar. Since rice is primarily grown by transplanting seedlings in flooded puddled fields, it requires a large amount of water (~150 cm), of which 15–20 cm (Singh *et al.*, 2001) is used only for puddling. This suggests that alternatives to traditional puddled transplanted rice are required to save water and increase crop, water, and labour productivity.

Rice production consumes about 30% of all freshwater used worldwide. Flood-irrigated rice uses two to three times more water than other cereal crops such as wheat and maize. In Asia, flood-irrigated rice consumes more than 45% of total freshwater used. System

of rice intensification (SRI) is a new system being developed for lowland areas with water shortage and for favourable upland areas with access to supplementary irrigation. Instead of keeping fields flooded, the soil can be kept near saturation, or alternate wetting and drying regimes are imposed through intermittent irrigation (Tabbal *et al.*, 2002). But due to the advantages like reduced weed problem and higher yield in conventional transplanted rice, giving up the long-term benefits of continuous flooding during rice growth is a risky management strategy, particularly on more fertile, sandy clayey soils found in those areas of India where, at present, water is not in short supply. Any system like SRI, in which the period under flooding is reduced significantly or in which intermittent irrigation causes repeated agitation of biological turnover processes bears the risk of being less sustainable over the long term (Witt *et al.*, 2000).

In several publications, Uphoff (2001), Stoop and Kassam (2002) and Uphoff *et al.* (2002) proposed that the SRI could unlock currently untapped production potentials of rice, allowing farmers to realize yields of up to 15 tonne ha⁻¹ or more with reduced irrigation and mineral fertilizer inputs (Stoop and Kassam, 2002). Most publications on SRI report large yield increases compared to “conventional” or “traditional” management of irrigated rice (Stoop and Kassam, 2002). SRI reports from 17 countries found that, average grain yields in all these studies were 6.8 tonne ha⁻¹ for SRI as compared to just 3.9 tonne ha⁻¹ for the recommended conventional irrigated rice management at these sites. Although yield advantages were claimed for the majority of reports, there were also examples for no yield increases over the traditional practice in Bangladesh, China, India, Myanmar, Nepal and Thailand (Dobermann, 2004; Sheehy *et al.*, 2005; McDonald *et al.*, 2005).

System of Rice Intensification (SRI) was first developed in Madagascar in 1983 by Henri De Laulance and it has been promoted for more than a decade as a set of agronomic management practices for rice cultivation that enhances yield (Uphoff *et al.*, 2002), reduces water requirements (Satyanarayana *et al.*, 2007), raises input productivity (Sinha and Talati, 2007), is accessible to smallholders (Stoop *et al.*, 2002), and is more favourable for the environment than conventional practice with its continuous flooding of paddies and heavy reliance on inorganic fertilization (Uphoff, 2003). Given that water scarcity at field level affects more and more rice growers around the world, SRI has attracted considerable interest, particularly in Asian countries.

SRI was introduced in India in 2000. Today, SRI is known in all rice-growing states in India. It is estimated that as many as 600,000 farmers are growing their rice with all or most of the recommended SRI crop management practices on about one million hectare distributed across more than 300 of the country's 564 rice-growing districts. This is probably the most rapid uptake of new agricultural practices seen in the country, making SRI a national phenomenon with very limited resources devoted to extension. Both on-farm and on-station

evaluations across many states and diverse growing environments have shown clearly that SRI has the potential to improve yield while reducing water use, production costs, and chemical inputs. Farmers see that the main advantages of adopting SRI are considerable savings in seed, water savings of up to 50%, improved soil health, and yield increases of 20–30%. Additional benefits include shorter time to maturity, higher out turn of polished rice when SRI paddy was milled. The major constraints experienced are lack of trained labour, difficulties in planting young seedlings, water management in low-lying areas, and greater requirements for weeding. In the era of climate change, increasing variability of rainfall and growing competition for water and land, SRI offers a new opportunity for increasing the production value per drop of water and for reducing agricultural water demand, which, in many parts of the world, accounts for the largest share.

SRI methods are seen to have the following impacts compared with their conventional counterparts: Depending on current yield, output per hectare is increased usually by 50% or more, with increases of at least 20%, and sometimes 200% or more. Since SRI fields are not kept continuously flooded, water requirements are reduced, generally by 25–50%. The system does not require purchase of new varieties, chemical fertilizer, or agrochemical inputs, although commercial inputs can be used with SRI methods. The minimal capital costs make SRI methods more accessible to poor farmers. Costs of production usually decline by 10–20% and it varied according to the input intensity of farmers' current production. With increased output and reduced costs, farmers' net income increases by more than their augmentation of yield. In an experiment at Hyderabad, SRI crops under organic and inorganic nutrient management resulted in 8.1 and 8.2 t ha⁻¹ grain yields with a 12.6% and 13.7% yield increase over the traditional method, respectively. Careful measurement of water use showed that water productivity was higher under SRI and water savings reached 37.5% and 34.2% under SRI-organic and SRI, respectively.

Rice growers, in general, rely on chemical fertilizers under the belief that they were necessary for high yields. However, achieving high yields based on the prescriptive use of fertilizers (and pesticides) may not be always economically warranted. Moreover, when government subsidies for fertilizers were reduced (e.g., in Indonesia) or eliminated (e.g., in the Philippines, Nepal), small scale rice farmers could no longer afford these inputs. Organic nutrient sources are important components of the nutrient cycle in agro ecosystems and could be utilized along with chemical fertilizers. Integrated nutrient management (INM) practices that rely primarily on judicious use of organic and inorganic sources have drawn the attention of rice scientists and farming communities.

Diazotrophic cyanobacteria are natural renewable biological nitrogen sources that are being used as bio-fertilizer for rice (Jha *et al.*, 1999a, 1999b; Kannaiyan, 2000; Sinha *et al.*, 2002). At least one third of Indian rice fields are amenable to cyanobacterial bio-fertilizer

application. Besides bio-nitrogen, they contribute plant growth-promoting substances (Misra and Kaushik, 1989), prevent the growth of weeds (Subramanyan *et al.*, 1965), add soil organic matter (Roychoudhary *et al.*, 1980), increase phosphorus availability and decrease sulphide injury to rice seedlings (Arora, 1969). The effects on rice yield of soil inoculation by blue-green algae (BGA) were first reported by Watanabe *et al.* (1951), with a 25% increase in yield after inoculation of poorly drained paddy with *Tolypothrix tenuis*. Later on, algal inoculation response was also reported from India, Burma, Egypt, China, Philippines, Bangladesh, West African countries and Russia (Roger *et al.*, 1993; Irrisari *et al.*, 2001; Aziz and Hashem, 2004). In a bibliographic survey covering 634 field experiments, inoculation of rice with cyanobacterial bio-fertilizer was found to have an increase in grain yield by 267 kg ha⁻¹ (Roger *et al.*, 1993).

Biofilm attached to the plant root of some crops help in the cycling of nutrients as well as the bio-control of pests and diseases resulting in improved production of the crop. Biofilms of nitrogen fixing and phosphorus solubilising microorganisms have been developed for the inoculation of various crops. The biofilms showed higher rates of biological nitrogen fixation and organic acid production, which was directly proportional to the synthesis of indole acetic acid like substances, than microbes when used alone. The biofilmed bio-fertilizers improve nitrogen fixing symbiosis in legumes, and could contribute directly to soil N fertility in long term. The biofilms based on cyanobacteria could be beneficial for paddy cultivation as well as for wheat. Cyanobacteria have been shown to colonize the roots of wheat. Biofilms based on bio-control agents like *Trichoderma* could be useful for control of many soil borne fungal diseases. *Agrobacterium tumefaciens* and *rhizobia* can form dense, structurally complex biofilms on root surfaces, extensively coating the epidermis and root hairs, and these bacteria also form elaborate biofilms on abiotic surfaces.

Plant growth-promoting rhizobacteria (PGPR) are naturally occurring soil bacteria that aggressively colonize plant roots and benefit plants by providing growth promotion. Inoculation of crop plants with certain strains of PGPR at an early stage of development improves biomass production through direct effects on root and shoots growth. De Datta (1981) opined that 'the microbial flora causes a large number of biochemical changes in soil that largely determine the fertility of soil. PGPR are commonly used as inoculants for improving the growth and yield of agricultural crops and offers an attractive way to replace chemical fertilizers, pesticides, and supplements (Ashrafuzzaman *et al.*, 2009).

Cyanobacteria are known to contribute to macro-aggregation and result in improved resistance to soil erosion, because as primary producers, they contribute to the enrichment of soil with soil organic matter (SOM) and to the improvement of biological activity (Acea *et al.*, 2003). Cyanobacteria are known to secrete extracellular polymeric substances (EPS) dominated by polysaccharides which can bind soil particles (Issa *et al.*, 2001). Prasanna *et al.*

(2009) reported the significant contribution of cyanobacterial biomass in enhancing microbial activity and plant growth promotion, emphasizing their significance in sustainable management of rice ecosystem. Issa *et al.* (2007) observed improvement in the aggregation of cyanobacteria inoculated soils which could be related to increase in soil carbon and EPS that cause change in the soil micro-morphological characteristics of the aggregates. Maqubela *et al.* (2009) reported that inoculation of degraded soil with strain of *Nostoc* could improve the fertility and structural stability of degraded soil.

With the above background, an experiment entitled “**Influence of cyanobacteria based inoculants and cultivation methods on rice (*Oryza sativa* L.) yield, soil carbon content and aggregation**” was conducted with the following objectives:

1. To identify the most promising cyanobacteria based inoculants enhancing agronomic traits and yield of rice,
2. To evaluate the effect of cyanobacteria based inoculants on soil properties, including soil aggregation,
3. To analyse carbon content in soil as influenced by cyanobacterial inoculants, and
4. To study the performance of SRI over conventional method of rice cultivation.

2. BACKGROUND

In this chapter research work carried out in India and abroad on the influence of cyanobacteria based inoculants and cultivation methods on rice (*Oryza sativa* L.) has been reviewed.

2.1 Methods of rice cultivation

2.1.1 The system of rice Intensification (SRI)

The system of rice intensification (SRI) was developed by Fr. Henri de Laulanié in 1980s over two decades of observation, participatory on-farm research and innovation in Madagascar, mostly on acid soils with low nutrient content and high potential for iron toxicity after submergence (Uphoff, 2006). Stoop *et al.* (2002) stated the major principles of SRI as follows: (1) Rice is not an aquatic plant; it survives under flooded conditions but does not thrive. (2) Rice seedlings lose much of their growth potential if they are transplanted more than 15 days after emergence. (3) During transplanting, trauma to seedlings, especially roots, should be minimized. (4) Wide spacing of plants leads to more root growth and tillering. (5) Soil aeration and organic matter create beneficial conditions for root growth. SRI has been promoted for more than a decade as a set of agronomic management practices for rice cultivation that enhances yield (Ceesay *et al.*, 2006; Kabir and Uphoff, 2007; Namara *et al.*, 2008; Senthilkumar *et al.*, 2008), reduces water requirements (Satyanarayana *et al.*, 2007), raises input productivity (Sinha and Talati, 2007), is accessible to smallholders (Stoop *et al.*, 2002), and is more favorable for the environment than conventional practice with its continuous flooding of paddies and heavy reliance on inorganic fertilization (Uphoff, 2003). Given that water scarcity at field level affects more and more rice growers around the world, SRI has attracted considerable interest, particularly in Asian countries. Rice growers in many countries are trying to find ways to achieve their production goals with less use of water, because of growing water shortages and competing demands, and SRI is one of the rice-farming options. SRI can (a) increase yields and production so that economic and food security goals met at the same time it can (b) reduce farmer's costs of production, enhancing profitability and (c) decrease the amount of irrigation water required (Stoop *et al.*, 2002; Uphoff, 2003; Randriamiharisoa *et al.*, 2006; Thakur *et al.*, 2011).

2.1.1.1 History of SRI

SRI was developed in 1983 by Henri de Laulanie, a French Jesuit working with Madagascar farmers and formed a NGO "Association Tefy Saina", who spent more than three decades in Madagascar trying to devise better production methods that would improve the lives of rural household, who were impoverished and heavily dependent on rice (Laulanie, 1993). Relying as little as possible on external inputs, he sought a methodology that would be

both accessible to poor and marginal farmers and be environmentally friendly. Changes introduced in the management of the rice crop, enumerated below, elicit from different rice genome and more productive phenotypes, including larger root systems. This growth and performance is achieved with substantial reductions (25-50%) in water application, should interest irrigation specialists and policy makers.

2.1.1.2 Key elements of SRI

Stoop *et al.*, (2002) and Uphoff *et al.*, (2002) provided a detailed overview of the rationale and key components of SRI and they also discuss its scope for adoption. SRI is understood as a set of principles and a set of mostly biophysical mechanisms. It originated in the humid highlands of Madagascar with rainfall mostly ranging from 1000 to >2000 mm, mostly on poor soils with low pH, low CEC, low available P, and high concentrations of soluble Fe and Al. Major SRI principles include (1) raising seedlings in carefully managed nurseries, (2) careful transplanting of single, young (8–15 days old) seedlings at wide plant spacing (starting at 25 x 25 cm, but going up to 50 x 50 cm), (3) intermittent irrigation to avoid permanent flooding during the vegetative growth phase, (4) addition of nutrients to the soil, preferably in organic forms such as compost instead of chemical fertilizer, and (5) intensive manual or mechanical weed control without herbicide use. However, SRI is not a “standard package” of specific practices, but rather represents empirical practices that may vary to reflect local conditions (Uphoff, 2002). Variants of SRI have also been tested in which only some of the basic components were practiced.

The key physiological principle behind the principal SRI measures is to provide optimal growing conditions to individual rice plants so that tillering is maximized and phyllochrons are shortened, which is believed to accelerate growth rates (Nemoto *et al.*, 1995). It was also observed that tiller mortality is reduced. Furthermore, intermittent irrigation is believed to improve oxygen supply to rice roots, thereby decreasing aerenchyma formation and causing a stronger, healthier root system with potential advantages for nutrient uptake.

2.1.1.3 The SRI practices

The specific operational practices that derived from Laulanie’s work can be stated as follows (Uphoff *et al.*, 2011):

- Transplant young seedlings, preferably 8–12 days, while still at the 2–3 leaf stage, certainly within 15 days of nursery sowing, before the start of the 4th phyllochron of growth (Nemoto *et al.*, 1995).
- Transplant quickly, within 30 minutes of gently removing seedlings from their nursery, and carefully and shallow (1–2 cm), taking care not to invert the seedlings’ root tips when transplanting, as this increases “transplant shock.”

- Transplant just one seedling per hill, and space the hills widely, in a square pattern. 25 cm x 25 cm was recommended as a typical square planting distance for SRI.
- Apply only a minimum of water during the plants' vegetative growth stage, wetting the field daily but with several drying periods ranging from 3 to 6 days. After panicle initiation, maintain just a thin layer of water, 1–2 cm, until 15 days before harvest, when the field should be drained.
- To control weeds, use of a mechanical hand weeder (rotating hoe) was recommended. This churns up the soil surface and buries young weeds. Weeding should start 10–12 days after transplanting, to preemptively curb weed growth and to aerate and fertilize the soil, and should be done several times before the canopy closes.
- The above SRI methods were initially accompanied by use of inorganic fertilizer, so SRI is not necessarily an “organic” cultivation system. When the Madagascar government removed fertilizer subsidies in the late 1980s, the use of compost became an attractive alternative, giving even better results than mineral fertilizers when it was used with the other practices.

SRI changes the management of rice plants and of the soil, water and nutrients that support them, in simple but specific ways to create optimal growing environments for rice plants so that their genetic potentials are better expressed. SRI is not a set technology; rather it is based on certain insights about how rice plants can be induced to become more productive, particularly by (a) eliciting greater root growth, which is visible and (b) enhancing soil biotic activity (Satyanarayana *et al.*, 2007). The recommended methods derived from SRI concepts are always to be tested and adapted to local conditions. The age of transplanted seedlings, their spacing and the amount and timing of water deliveries e.g., should be evaluated and adjusted in the field for best results. The basic elements of SRI practices are the following:

i. SRI methods give highest yield when young seedlings are transplanted, < 15 days old and preferably only 8-12 days before the start of the 4th phyllochron (Stoop *et al.*, 2002). This preserves plants potential for tillering and root growth that is compromised by later transplanting (Randriamiharisoa and Uphoff, 2002). Transplanting is no longer considered a necessary part of SRI because its concepts and methods are also being adapted for direct seeding to save labour. However, at present, transplanting is the most reliable and widely used practice with SRI.

ii. Transplanting should be done carefully to avoid trauma to the plants, roots and also quickly to avoid their becoming desiccated. Seedlings are raised in an unflooded garden like nursery and then transplanted within 15-30 minute after uprooting. Shallow transplanting is recommended, only 1-2 cm deep, with roots laid in the soil as horizontally as possible.

Plunging the roots into the soil vertically inverts the seedling's root-tips upward, slowing the plants recovery from the shock of transplantation and delaying their resumption of growth.

iii. Plant density is greatly reduced with SRI compared to conventional rice cultivation. Instead of replanting seedlings in clumps of 3 to 6 plants, they are transplanted singly and in a square pattern. Initially, spacing of 25 x 25 cm² is recommended, but as SRI practices improve the soil over time, wider spacing can later give even higher yields. Sparse planting avoids the inhibition of root growth that results from crowding, and by exposing plants to more light and air SRI creates 'the edge effect' (also known as the border effect) for the whole field.

iv. Seedlings are transplanted into a muddy field rather than flooded with standing water. During the vegetative growth phase, paddy soil is kept moist but never continuously saturated because flooding creates hypoxic soil conditions that cause rice roots to degenerate. Under continuous flooding, up to three fourth of roots degrade by the flowering stage (Kar *et al.*, 1974). The SRI recommendation has been to maintain 1-3 cm of standing water in field after panicle initiation. However, this may be more than necessary. Some SRI farmers who practice alternate wetting and drying (AWD) throughout the crop cycle, with no continuous flooding, report good results.

v. To control weeds, hand weeding or use of mechanical weeder is recommended, starting 10 days after transplanting and repeated 2-4 times at 10-12 days intervals until the canopy closes. Rotating hoe that aerates the soil while it prevents weeds growth by churning them into the soil is more suitable. Soil aeration appears to stimulate the growth of aerobic bacteria and fungi and associated organisms in the soil food web. Planting in a square pattern allows farmers to weed their fields in perpendicular directions, which achieves more and better soil aeration.

vi. Nutrient management preferably through organic sources, however, SRI was originally developed using chemical fertilizers to augment soil nutrient supplies. But when the Madagascar government cut subsidies in the late 1980's, SRI farmers were encouraged to apply compost instead used together with other SRI practices; this gave better results and was preferable for cash-poor farmers. The advantages from using compost have been seen from factorial trials (Uphoff, 2003) but if organic matter is not available, SRI practices can be used successfully with fertilizers (Satyanarayana *et al.*, 2007).

vii. Correct implementation of various interdependent components of SRI require considerable field experience in which close observation and timeliness are crucial in minimizing labour requirements while maximizing total factor productivity.

2.1.1.4 Benefits of SRI

SRI performances have raised hope among policy makers, development activists and farmers to solve the food deficit problem in remote areas where modern technologies are not

feasible in terms of cost and accessibility. SRI helps to increase the yields and production, reduces farmers' costs of production, and decreases the water requirement for irrigation up to 50 percent less (Randriamiharisoa *et al.*, 2006). SRI helps resource-poor farmers to attain higher yields despite having infertile soil, no mineral fertilizer input, reduced irrigation and fewer seeds. Benefits of SRI have been demonstrated in 32 countries of Asia, Africa, and Latin America namely China, Indonesia, Cambodia, Vietnam, Philippines, Laos, Myanmar, Thailand, India, Nepal, Bangladesh, Sri Lanka, Pakistan, Bhutan, Afghanistan, Iran, Iraq, Egypt, Gambia, Guinea, Senegal, Mali, Sierra Leone, Benin, Mozambique, Rwanda, Zambia, Cuba, Peru, and Brazil.

Evidences show that SRI method increases water-saving by 65 percent in China (Satyanarayana *et al.*, 2007). Similarly, the economic return was reported 41 percent higher in Cambodia, also there was 44 percent increase in yield in Srilanka. The cost of production was reported to be reduced by 25 percent in Indonesia. The rice production with SRI increased by 28 percent with 53 percent less water in Tamil Nadu in India. The income with SRI was increased by 44 percent in Sri Lanka. Similarly, the harvest increased by 35 to 50 percent in China and 41 percent in Cambodia. As compared to the rice grown through traditional way, the SRI has following benefits:

a. Greater resistance of SRI crop to pest and disease loss: SRI crops are found to have greater resistance against pest and diseases. Chemical means of controlling diseases and pests are neither economic nor necessary, if SRI practice is followed. Zheng *et al.* (2004) stated that SRI plants have less insect and disease problems.

b. Greater resistance to abiotic stress: In coming decades, the farmers have to face various changes in climatic pattern so it is important that the crops can tolerate adverse biotic and abiotic stresses. SRI plants have large and strong root systems which enable them to tolerate adverse climatic influences as water stress, drought, storm damage, cold snaps, heat waves etc. Additionally, despite their larger and heavier grain panicles, SRI plants have greater resistance against lodging i.e. falling over.

c. Higher out-turn of milled rice: SRI plants not only increase the yield of paddy (kg of unmilled rice harvested per hectare), but also offer 15 percent more of milled rice (kg of consumable rice per bushel of paddy), because SRI grains have less chaff (fewer unfilled grains) and less shattering (fewer broken grains).

d. More nutritional value and grain quality: There is 30-65 percent less chalkiness in SRI grains. SRI roots are larger and go deeper into soil so it is believed that SRI plants uptake higher micronutrients from the soil.

e. Reduction in greenhouse gas emission: Flooded rice fields are an important source for green house gas emissions. It is believed that methane emission from SRI soils will be less because the rice is grown under aerobic conditions. Also, application of no inorganic nitrogen

fertilizer may reduce nitrous oxide formation. However, very few studies have been done so far to evaluate this.

2.1.1.5 Criticism regarding SRI

Scientists such as Dobermann (2004), McDonald *et al.*, (2006), Sinclair (2004) have raised the questions regarding the yield benefits of SRI. Dobermann (2004) reported that intermittent irrigation in SRI practice bear short and long term risks. He further reported that SRI favoured rice growth on poor soils but it was likely to have little potential for improving rice production in intensive irrigated systems on more favorable soils. McDonald *et al.* (2006) found no empirical evidence that SRI fundamentally changes the physiological yield potential of rice. Sheehy (2004) has found no major role of SRI in improving rice yields in his experiments carried out in China and reported that the extraordinary high yields may be due to experimental error. Sinclair (2004) claimed that SRI used very low plant densities so suffered from poor light interception. Additionally, he claimed that SRI replaced flooding of rice field simply by maintaining moist soil conditions but the rice fields were flooded so as to assure no water limitations. He stated that ample water maximized rice yields. Furthermore, he claimed that SRI lacked sufficient mineral nutrients in order to achieve high yields. The claimed yield of 15 t ha⁻¹ requires nitrogen from over 50 t ha⁻¹ of organic matter because rice grains contain about 0.013 gm of N per gram of seed. Despite such criticisms, SRI is gaining popularity all over the rice growing countries. Farmers have been able to grow more with less water, less mineral fertilizer and less seeds. There may be multiple benefits of SRI which may have been poorly studied.

2.1.2 System of rice intensification vs. traditional methods

2.1.2.1 Growth components

2.1.2.1.1 Plant growth

The improved root and shoot growth under AWD-SRI rice contributed directly to better plant growth, grain setting (higher percentage of filled grains) and heavier individual grains (higher 1000-grain weight). Zhang *et al.* (2009) also reported that higher grain yield in the “super” rice varieties (Liangyoupeijiu and Huaidao 9) was attributable to improved root and shoot growth, which contributed to the larger sink size (total number of spikelet). SRI practices enhance the plants growth and tillering ability and improve plant/culm height and strength of tillers (greater tiller perimeter). Tillering ability in rice has a close relationship with the number of phyllochrons completed before entering the reproductive stage (Nemoto *et al.*, 1995). SRI plants, due to their early establishment, only suffered minimally from transplanting shock; the subsequent favorable growing conditions allowed the plants to complete a greater number of phyllochrons before the onset of anthesis, while producing a greater number of strong tillers and larger root systems than did scientific management

practices (SMP) plants (Thakur *et al.*, 2010b). Conversely, SMP plants appear to be constrained by their competition for nutrients, space and light during their later stages of vegetative growth, which was illustrated by the slowing down of crop growth rate (CGR) beyond 60 days after germination. Number and size of leaves were significantly increased in SRI plants compared with SMP at the flowering stage which led to a higher leaf area index (LAI) than in SMP. The extensive root systems developed by SRI plants enhanced water and nutrient uptake, resulting in greater leaf elongation rates (LER), which may have contributed for larger leaf size. The higher specific leaf weight (SLW) in SRI plants indicated thicker leaves compared to the leaves on rice plants grown under SMP.

SMP rice plants had a more compact structure, with tillers that were more vertical or less horizontal. In contrast, SRI plants had more open architecture (greater canopy angle), with tillers splayed out more widely and covering more ground area. Greater LAI and a more favorable canopy structure facilitate higher light interception in the SRI crop beyond 50 days after germination (DAG). This study also showed that SRI leaves had higher light utilization capacity (Fv/Fm and UPS II) and a greater photosynthetic rate, especially during the reproductive and ripening stages of the crop. Actively photosynthesizing leaves ensure a sufficient supply of assimilates to the roots for their development and longevity, maintaining active root functioning. Manickasundaram (1992) reported that recouping submergence once in four days gave higher plant height, LAI, shoot and root dry weight and tiller production as that of continuous submergence. Giving irrigation at one day after disappearance of ponded water increased the plant height, LAI, DMP, tiller production, root dry weight and volume (Asokaraja, 1993; Balasubramanian, 1998).

Vijaykumar *et al.*(2006) found that the treatment combination of 14 days old seedlings planted at 25 x 25 cm² spacing + water saving irrigation and SRI weeding significantly recorded taller plants (109 cm), total dry matter production (120 kg ha⁻¹) and LAI (7.69) than conventional weeding. Between panicle initiation (PI) to flowering (FL) and between FL to maturity stage the CGR, RGR and NAR were significantly increased by the treatment combination of 14 days old seedling, wider spacing of 25 x 25 cm², limited irrigation of 2 cm with incorporation of weeds and disturbing the soil through SRI weeding using rotary weeder.

Shrirame *et al.* (2000) reported that the number of functional leaves, leaf area and total number of tillers hill⁻¹ were higher at wider spacing which increased the photosynthesis rate leading to taller plants. Rajesh and Thanunathan (2003) recorded higher total dry matter production under wider spacing. It is due to increased shoot: root ratio and production of more number of tillers on individual hill.

Root system supports canopy growth by providing nutrients and water, while canopies in turn photosynthesizes, some part of which is shared with root systems for their

nutrition. Positive feedback between root growth and shoot growth is easy to understand as a larger root system can support a larger canopy and vice-versa. There are also phytohormone effects, as shoots release auxins which enhance root initiation, while roots produce cytokinins that stimulate the development of shoots (Oborny, 2004). Kumar and Shivay (2004) found 30-50 tillers plant⁻¹, 80-100 possible and sometimes even more from a single plant in SRI. De Datta and Surajit (1987) reported that with SRI seedlings are transplanted before they are 15 days old. This preserves the plants potential for massive tillering and significantly higher yield.

2.1.2.1.2 Yield attributes

Higher root metabolic activity supports a high photosynthetic rate by supplying a sufficient amount of nutrients to the shoot/leaf (Samejima *et al.*, 2004; Yang *et al.*, 2004; Mishra *et al.*, 2006). This interdependent relationship has been referred to as the root–shoot interaction (Samejima *et al.*, 2004), and SRI practices have a substantial effect on such root–shoot interaction. Other workers (Menete *et al.*, 2008; Thakur *et al.*, 2010a, b; Uphoff *et al.*, 2011) reported significant improvements in per-hill performance under SRI, in terms of morpho-physiological characteristics, with resulting enhancement of grain yield. However, since yield is assessed by performance per unit area, greatest productivity is achieved by optimizing the number of plants m⁻² without compromising individual plant performance for maximizing grain yield. Vijaykumar *et al.*, (2004) reported that the yield attributes (panicle length, number of panicles hill⁻¹ and total number of grains panicle⁻¹) were significantly higher in the treatment involving 14 days old seedling + 25 x 25 cm² spacing + water saving irrigation + LCC based N management + SRI weeding than other treatments during wet season.

During the dry season, greater values of panicle length, no. of panicles hill⁻¹ and filled grains panicle⁻¹ were recorded in treatment combination involving 14 days old seedlings + 25 x 25 cm² spacing + water saving irrigation + SRI weeding. Uphoff (2001) observed that under SRI, number of roots, tillers and grains panicle⁻¹ were increased by having more space between plants, which respond positively in their greater exposure to sunlight and circulatory air. Verma *et al.*, (2002) reported that closer spacing (20 x 10 cm²) gave higher sterility percentage than wider spacing. Ceesay and Uphoff (2003) reported that under SRI, 1000 grain weight and biomass accumulation were higher by 6.7 and 20.1% respectively compared to conventional practice of continuous flooding. The average panicle setting rate (32%) was only about half than that from SRI management (58%).

2.1.2.1.3 Yield

Most publications on SRI report large yield increases compared to “conventional” or “traditional” management of irrigated rice (Stoop *et al.*, 2002). Unweighted average grain

yields in all these studies were 6.8 Mg ha⁻¹ for SRI as compared to just 3.9 Mg ha⁻¹ for control treatments that represented the recommended conventional irrigated rice management at these sites. The extraordinary yields reported from Madagascar have, however, not been achieved elsewhere. Although yield advantages were claimed for the majority of reports, there were also examples for no yield increases over the control in Bangladesh, China, India, Myanmar, Nepal, and Thailand. Numerous uncertainties are associated with these studies. Most of the papers published in Uphoff *et al.* (2002) do not include sufficient descriptions of site characteristics (e.g., soil, climate), experimental protocols, field management, sampling methods, and statistical data analysis to properly assess the validity of the results reported. Many reports represented on-farm experimentation or even unreplicated demonstration plots with limited data collection and statistical analysis. Little information was given on how the “conventional” plots were managed in comparison with SRI, and also with regard to known best practices for water, nutrient, and pest management.

Many comparisons of SRI with conventional management practices also tended to be confounded because total nutrient inputs in SRI and conventional treatments differed, and because the conventional management treatments did not represent average or advanced levels of modern rice production. For example, the average grain yield reported for conventional irrigated rice management (3.9 Mg ha⁻¹) in the SRI studies was significantly less than the current global average irrigated rice yield of about 5.3 Mg ha⁻¹. To resolve such issues, SRI must focus on carefully conducted, replicated field experiments in which known best-management practices are fully implemented at yield levels that approach location-specific ceilings. The studies by Wang *et al.* (2002) are among few published examples in which SRI was compared to a well-managed conventional treatment at high yield levels (8.5–11.5 Mg ha⁻¹). Despite differences in various physiological characteristics measured during the growing season, final numbers of tillers, grain yields, and components of yield of *Indica* varieties or hybrids were either the same or even higher with conventional management than under SRI.

Experimental evidences are forthcoming on why the rice crop under SRI results in higher yields than under conventional or recommended cultivation practices. Thakur *et al.* (2010) showed that alterations in management practices can induce multiple, significant, and positive changes in phenotype from a given rice genotype. The increase in yield with SRI when compared with that obtained using recommended management practices reached 42% and it was associated with various phenotypic alterations such as longer panicles, more grains panicle⁻¹, higher percent of grain filling, increased productivity per plant, deeper and better distributed root systems, higher xylem exudation rates, more open plant architecture with more erect and larger leaves, more light interception, higher leaf chlorophyll content at

ripening stage, delayed senescence and greater fluorescence efficiency, higher photosynthesis rate, and lower transpiration.

The set of practices like SRI when used incompletely usually enhances grain yield with precision and care, yields in the range of 10-15 tons ha⁻¹ or even higher have been achieved. These practices are all known to have positive effects on yield (Horie *et al.*, 2005). When the China National Hybrid Rice Research and Development Centre began evaluating SRI methods in 2000, it found that with careful management, water applications for rice production could be reduced by as much as 65% on SRI plots compared with previous applications while still getting 1-2 tonnes ha⁻¹ more production as top of the record high yields that is could obtain with the its hybrid varieties (Satyanarayana *et al.*, 2007).

Satyanarayana *et al.* (2007) found that even when the Andhra Pradesh farmers were unable to implement all the aspects of SRI in 1st season (Summer 2003), just planting younger seedling carefully at wider spacing with somewhat better water management practices resulted in over 2.0 t ha⁻¹ extra yields as compared to the conventional methods using higher inputs. He predicted that with more experience, higher yields may be possible. There are confirming results from 21 additional countries ranging from China to Peru, with average yields from SRI in 7-8 t ha⁻¹ range, and with yields over 15 t ha⁻¹ reported from at least four countries beyond Madagascar. There are some researchers at leading Institutions in China, India and Indonesia now working on different aspects of SRI. Initial trials have shown high potentials for its acceptance as an improved method of rice production. In the Sichuan province of China, during the 2004 summer, average yield gains were 3 t ha⁻¹ over the usual yield of 7.5 t ha⁻¹. The highest SRI yield was recorded as 20.4 t ha⁻¹ in Yunnan Province, water saving of 42% were recorded in Sichuan, while in Zhejiang province, incidence of sheath blight, a major rice disease in the area, was reduced by 70%. The subsequent harvest, almost 13 t ha⁻¹, set a record for that province (Satyanarayana *et al.*, 2007).

In Andhra Pradesh, results of SRI trials by 50 farmers during *rabi* season 2003-04 showed an increase of yield from 7.1 to 9.7 t ha⁻¹ with the highest measured yields surpassing 15 t ha⁻¹. In Cambodia, evaluation of performance of SRI covering five provinces showed average yield gain of 41% with gross profits ha⁻¹ increased by 74% rather significantly, SRI was found there not to be labour intensive (Satyanarayana *et al.*, 2007).

In Tamil Nadu, SRI evaluation by the crop and soil management center of the Tamil Nadu Agricultural University (TNAU), at Coimbatore started in 2000 with field experiment at the wetland farm of TNAU assessing different methods of crop establishment, spacing and water management. Overall yield increases were realized from combined effects of these management practices, with the highest yield obtained from SRI practices (7.6 t ha⁻¹). Mean grain yield for all water-saving treatments (6.35 t ha⁻¹), indicating that use of younger seedling and soil aerating weeding had a beneficial effect, of particular interest was the finding that in

situ incorporation of weeds into the soil with the rotating hoe, significantly increased yield (6.7 t ha⁻¹) compared to conventional weeding (6.08 t ha⁻¹) (Satyanarayana *et al.*, 2007).

In Sri Lanka, an International Water Management Institute (IWMI) evaluation in two districts found almost a 50% increase in yield. Water productivity increase 90%, reduction in cost of production by 17-27% and 112% increase in net profit. In Indonesia, the Agency for Agricultural Research and Development (AARD), based on evaluation of SRI practices for three years, made SRI part of its new national strategy for integrated crop resource management to restore growth in the rice sector (Uphoff, 2002b). McHugh *et al.* (2002) reported that SRI was associated with significantly higher grain yield of 6.41 t ha⁻¹ compared with 3.4 t ha⁻¹ from conventional practices on SRI plots, grain yield were 6.7 t ha⁻¹ for alternate wetting and drying irrigation, 5.9 t ha⁻¹ with non-flooding irrigation and 5.9 t ha⁻¹ for continuously flooded.

Krishnakumar (1986) observed that irrigation one day after disappearance of ponded water gave yields comparable with continuous submergence besides considerable saving in quantity of water. When examining ICM (integrated crop management), it is important to take into consideration the SRI development in Madagascar in late 1980s (Randriamiharisoa and Uphoff, 2002). Vijaykumar *et al.* (2004) revealed that grain yield and water productivity were significantly increased when applying SRI weeding with 14 days old dapog seedlings planted at 25 cm x 25 cm spacing to achieve yields of 7009 and 5655 kg ha⁻¹ and 0.610 kg and 0.494 kg m⁻³ water, respectively in wet and dry season.

Uphoff (2002b) reported that instead of transplanting relatively mature 3 to 4 weeks old, as is common practice, with SRI seedlings are transplanted before they are 15 days old, even 8 or 10 days old. This preserved the plant potential for massive tillering if the other practices are followed. Khush *et al.* (1996) reported that with SRI method, a reversal of relationship between number of tiller plant⁻¹ and number of grains panicle⁻¹. This supported increase yield from 2 t ha⁻¹ to 8 t ha⁻¹. Thiyagarajan *et al.* (2002) reported that the modified SRI practices, 14 days old seedling should be transplanted in puddled field with the square planting of 25 cm x 25 cm which gave significant effect on grain yield of about 5059 to 7612 kg ha⁻¹.

The SRI is a system of rice production through synergistic interactions for the production of much higher grain yield than usually achieved by conventional practices with new varieties and external inputs (Uphoff, 2001a). It permits resource limited farmers to realize yields up to 15 tons of rice ha⁻¹ on infertile soils, with greatly reduced rates of irrigation and without external inputs. In spite of poor soil fertility, small farmers using the SRI methods on plots that range from 100 to 500 m² have obtained average rice yields of 8 to 9 t ha⁻¹ (Hirsch, 2000). Randriamiharisoa and Uphoff (2002) reported the results of factorial trials evaluating four SRI practices. SRI practices evaluated were young age seedlings (8, 12,

16 and 20 days old), water management (aerated soils and saturated soil), plant density (1 and 3 seedlings hill⁻¹) and fertilization (compost vs. NPK @ 16-11-22 vs. no fertilizer). They reported that these SRI practices were having advantageous effects individually as well as when combined. In fact, these trials showed a high degree of synergy among practices. Although ‘young seedlings’ were found to be the most important practice in these trials, none could be discarded without some loss. With growing conditions controlled, using all SRI practices, young seedlings, one seedling hill⁻¹, aerated soil, and added compost gave yield increase of 140 to 245%, compared to plot where only non-SRI practices (more mature seedlings, three hill⁻¹, saturated with NPK fertilizer were used).

Sinha and Talati (2007) reported that the number of SRI adopters has increased in India in recent years. They evaluated the impact of adoption of SRI practices on rice yields, the economics of paddy cultivation and labour inputs based on field research conducted in Parulia, West Bengal. Paddy yields with SRI were higher than those under conventional paddy cultivation by 32%, net return were higher by 67% and labour input was reduced by 8%. They also reported that, SRI promises to be a significant alternative for not only raising paddy yields, but also for managing paddy based farming in resources-starved region. SRI advocates often maintain that conventional research approaches to SRI evaluation ignore what is happening in farmer’s fields and that the rapid geographic spread of SRI is prima facie evidence of its value (Stoop and Kassam, 2005). Husain (2004) documented a 30% yield advantage for SRI in Bangladesh. An evaluation of SRI in 2003 by farmers fields schools in three communities in Negros occidental province of the Philippines estimated SRI yield of 7.33 t ha⁻¹, which was almost triple the 2.65 t ha⁻¹ that was obtained from standard farmer practices (Lazaro, 2004). At times, yields up to 21 t ha⁻¹ has been reported (Uphoff, 2002b). Researchers have used several concepts to explain the higher yield obtained with SRI. Berkelaar (2001) uses the concept of “synergy” resulting from a combination of management practices. He attributed higher yields to the greater number of phyllochrons with SRI.

2.1.2.1.4 Nutrient uptake

Wang *et al.* (2003) reported that an increase in nitrogen rate increased the number of tillers but decreased dry matter translocation percentage from vegetative organs to grains, suggesting yield increase under SRI should not rely on the excess nitrogen rate. Turner (2006) report on contribution of organic phosphorus in rice nutrition, he found that organic phosphorus concentrations were greater in soils rich in organic matter, but there were not apparent differences between soils under conventional flooded rice cultivation and the system of rice intensification. Additional experiments are now required to assess the role of organic phosphorus in the nutrition of rice growing under a range of management and soil conditions.

Biswas and Mahapatra (1980) found that water logging resulted in significantly higher N content in the plant body at dough stage (1.12%) and total uptake (157.7 kg ha⁻¹) at

the time of harvest compared to alternate wetting and drying. The N uptake was greater under higher percolation in well drained soils than under lower percolation (Khind and Kazibwe, 1983). Wang *et al.* (2003) reported that the increase in the N rate resulted in increase in N uptake, straw N content and yield, and in the reduction of percentage of N translocation (PNT) from vegetative organs, N dry matter production efficiency (NDMPE), and N gain production efficiency (NGPE). N uptake and NGPE decreased under severe water stress (aerobic cultivation system). Mild water stress (SRI) reduced N uptake, and increased PNT from vegetative organs, NDMPE and NGPE. Grain yields were enhanced with the reduction of water stress under 30 g N m⁻², and with mild water stress at 15 g N m⁻². Barison (2002) found that plants of same variety took 90% more N, K and 60% more P under SRI management compared with conventional rice.

2.2. Cyanobacterial bio-fertilizer

Cyanobacteria comprise a large group of structurally complex and ecologically significant gram-negative prokaryotes, which exhibit a wide range of nutritional capabilities ranging from obligate phototrophy to heterotrophy (Vasudevan *et al.*, 2006). Wetland rice fields can provide an ideal condition for the growth of cyanobacteria, which accumulate 19–28 kg N ha⁻¹ crop⁻¹, and can reduce the use of urea fertilizer in rice culture by 25–35% (Hashem, 2001). Experimental results at the IRRI (Los Banos, Philippines) revealed that the amount of N accumulation by cyanobacteria varies among soils, ranging from a few to 50 kg N ha⁻¹ crop⁻¹ (Roger and Ladha, 1992). The literature on the beneficial effects of cyanobacteria on the growth and yield of rice is voluminous (Kannaiyan *et al.*, 1997; Kennedy and Islam, 2001). However, the efficiency of cyanobacteria in increasing rice yields varies depending on soil types. Findings of several field experiments conducted on different types of soils show that cyanobacteria can be used to reduce urea- N inputs by 25–35% for a rice crop in acid, saline and red soils while they are less effective in calcareous and neutral soils (Hashem, 2001).

2.2.1 The Blue green Algae or Cyanobacteria

Algae are the heterogeneous assemblage of plants that includes prokaryotes and eukaryotic organisms. They are the pioneer colonizers both in hydrosphere and xerosphere and occupy the base of the tropic pyramid. These organisms have been found to synthesize 0.8x10¹¹ tonnes of organic matter, constituting about 40 percent of the total organic matter synthesized annually on this planet.

Blue green algae or cyanobacteria constitute the largest, most diverse and widely distributed group of prokaryotic microscopic organisms that perform oxygenic photosynthesis. These are also known as myxophyceae, cyanophyceae and cyanobacteria. These are ubiquitous in distribution, more common in tropics; and are able to withstand extremes of temperature and drought. Ability to withstand adverse ecological conditions,

capacity to thrive well in hostile environments and response to the onset of dry conditions by entering into dormant resistant state, have distinguished this group of organisms as pioneers of plant succession. Nitrogen fixation is carried out in specialized cells known as heterocysts, which have thick walls and hence, physically prevent the entry of oxygen and provide necessary anaerobic conditions for the activity of the enzyme nitrogenase. Heterocysts have a high rate of respiration that scavenges the diffused oxygen, lack photoreaction II of photosynthesis due to which there is no splitting of water and evolution of oxygen during photosynthesis. Therefore, these cells act as ideal site for nitrogen fixation under aerobic conditions. Although, nitrogenase enzyme is reported to be present in vegetative cells also, however, it remains inactive because of the presence of oxygenic photosynthesis.

The basic significance of the ecological observations on the abundance of blue green algae in Indian rice field soils became apparent when it was recognized that heterocystous forms can fix atmospheric nitrogen that is made available to the plants during life cycle and after its death by decomposition of cells which became available to the subsequent crops. Till the findings only forms like *Anabaena*, *Nostoc*, *Cylindrospermum*, *Tolypothrix*, *Calothrix*, *Scytonema*, *Hapalosiphon*, *Westiellopsis* and *Stigonema* belonging to orders Nostocales and Stigonematales were considered to be capable of fixing atmospheric nitrogen. Since then, many of the non-heterocystous forms are also reported to fix nitrogen under aerobic or micro-aerophilic conditions except *Gloeocapsa* that can fix nitrogen under aerobic conditions (Stewart *et al.*, 1979). In the later case, it is accomplished because of the spatial and temporal separation of the processes of carbon and nitrogen fixation.

2.2.2 Use of BGA bio-fertilizer for rice crop

De (1939) was the first to suggest a positive role for the blue greens in the sustenance of the nitrogen status of rice fields of our country. Since then lot of information has been generated in tropics regarding improvement in the fertility status of rice soils to sustain rice yields by utilizing diazotrophic blue green algae as biological input (Singh and Bisoyi, 1989). Singh (1939, 1942) in Uttar Pradesh attributed the enhanced nitrogen contributions to the nitrogen fixing capacity of blue green algae and the nitrogen contribution was calculated as 48 kg N ha⁻¹crop⁻¹. They also suggested the persistence of residual effect of these organisms in the following season. Venkataraman (1966) initiated the work on the algalization of Indian rice fields and much interest was generated in this aspect. Multi-location trials conducted under varying agro-climatic conditions using different rice varieties indicated that the algal inoculation could save 30 kg N ha⁻¹ that, however, depends upon the agro-ecological conditions (Venkataraman and Goyal, 1969). The performance of *Aulosira fertilissima* in water logged rice fields was assessed and a 10-30 % increase in soil nitrogen by BGA incorporation was reported (Singh and Mandal, 2000).

2.2.3 Algal bio-fertilizer versus chemical fertilizer

Bio-fertilizers are renewable sources of plant nutrients to supplement chemical fertilizers in sustainable agricultural system. Chemical fertilizer is the major supplier of nutrients besides organic and green manures. Despite sizeable increase in the use of chemical fertilizers, the gap between the nutrient removal and replenishment is significantly high. The role of biofertilizers in sustainable agriculture assumes special significance particularly in present context of high cost of chemical fertilizers. The effect of bio-fertilizer is not as instantaneously obvious as in the case of chemical fertilizers. It is really difficult to believe the effectiveness of the 'cover crop' of these 'Microbugs'. These can be hardly seen with the naked eye but one must have the patience to realise the potential of this inexpensive input, which is self-regenerating and has sustained cumulative effect. As the crop plants utilize the products of biological activities of a bio-fertilizer, there is always a possibility of their population build up in the soil that may exhibit the benefits after a few superimposed applications.

These are also known to play an important role in soil conditioning and may also be source of growth regulators and organic matter. Algal bio-fertilizers are much cheaper than the chemical fertilizers available in the market. The crop plants are able to utilize more nutrients from the soil in presence of the algal inoculation because of slow release of the fixed and metabolically nitrogen. The chemical fertilizers provide the nutrients in a cyclic manner but the blue green algal biofertilizers provide the nutrients in a gradual and linear manner. This results in the utilization of the applied nutrients more effectively. The diazotrophic cyanobacteria are more compatible with nitrate nitrogen than ammonium nitrogen. This leads to rapid multiplication of blue green algae in the photic zone of rice field where oxidizing conditions tend to keep the level of nitrate nitrogen high. In presence of combined nitrogen the blue green algae not only continue to grow but show concentration dependent increase in the uptake of added chemical nitrogen. This metabolized nitrogen which is prevented from being lost through denitrification, percolation and run off, is slowly released through excretion and autolysis (Singh and Mandal, 2000).

Further, the algal nitrogenase has 'switch on' mechanism, which is activated when the level of combined nitrogen falls below a threshold value, which seems to be around 40 ppm. This enables the increased algal biomass to contribute more of the biologically fixed nitrogen when the level of fertilizer nitrogen in the ecosystem is reduced due to progressive utilization and loss (Singh and Mandal, 2000).

2.2.4 Bacterial inoculant bio-fertilizers

Bacterial inoculant bio-fertilizers are efficient sources of N used to substitute for urea-N in rice production. Some bacteria like *Azotobacter*, *Clostridium*, *Azospirillum*,

Herbaspirillum and *Burkholderia* can supplement urea-N by BNF while *Rhizobium* can supplement urea-N by promoting the growth physiology or root morphology of the rice plant. In addition, by acting as plant growth promoting rhizobacteria (PGPR), they can improve the ability of the rice plant to assimilate soil N. The beneficial effects of PGPR have been well demonstrated in both greenhouse and field conditions (Biswas *et al.*, 2000a, 2000b).

2.3 Biofilm fertilizer

Biofilm attached to the plant root of some crop helps in the cycling of nutrients as well as the bio-control of pests and diseases resulting in improved production of the crop. Biofilms of nitrogen fixing and phosphorus solubilising microorganisms have been developed for the inoculation of various crops. The biofilms showed higher rates of biological nitrogen fixation and organic acid production, which was directly proportional to the synthesis of indole acetic acid like substances, than microbes when used alone. The biofilmed bio-fertilizers improve nitrogen fixing symbiosis in legumes, and could contribute directly to soil N fertility in long term. The biofilms based on cyanobacteria could be beneficial for paddy cultivation as well as for wheat. Cyanobacteria have been shown to colonize the roots of wheat. Biofilms based on bio-control agents like *Trichoderma* could be useful for control of many soil borne fungal diseases. Formation of a biofilm begins with the attachment of free-floating microorganisms to a surface. These first colonists adhere to the surface initially through weak, reversible van der Waals forces. The first colonists facilitate the arrival of other cells by providing more diverse adhesion sites and beginning to build the matrix that holds the biofilm together. Once colonization has begun, the biofilm grows through a combination of cell division and recruitment. The final stage of biofilm formation is known as development, and is the stage in which the biofilm is established and may only change in shape and size. The development of a biofilm may allow for the aggregate cell colony (ies) to be increasingly antibiotic resistant. For example, the bacteria colonize root elongation zones and root hairs, forming dense biofilms. Microscopy of rhizobial cells within curled root hairs reveals small biofilm-type aggregates that provide the inocula for root invasion; the rhizobial cells migrate down infection threads as biofilm like filaments towards the root interior.

Microorganisms synthesize a wide spectrum of multi-functional polysaccharides including intracellular polysaccharides, structural polysaccharides and extracellular polysaccharides or exo-polysaccharides (EPS). Production of exo-polysaccharide is generally important in biofilm formation, and likewise can affect the interaction of microbes with roots and root appendages. Exo-polysaccharides generally constitute of monosaccharide and some non-carbohydrate substituents (such as acetate, pyruvate, succinate, and phosphate). Owing to the wide diversity in composition, exo-polysaccharides have found multifarious applications in various food and pharmaceutical industries. Recent findings suggest that multiple

polysaccharides modulate the chemical and physical attributes of the *Pseudomonas fluoresces* and *P. aeruginosa* biofilm matrix on abiotic surfaces. Such complexity may explain variable observations regarding the requirement for specific exo-polysaccharides in biofilm formation and root association.

A biofilm is an aggregate of microorganisms in which cells adhere to each other and/or to a surface. In fact, archaeal, bacterial, and eukaryotic microbes produce the biopolymers. Biofilms may form on living or non-living surfaces, and represent a prevalent mode of microbial life in natural and industrial. The microbial cells growing in a biofilm are physiologically distinct from planktonic cells of the same organism, which, by contrast, are single-cells that may float or swim in a liquid medium. Biofilm cells respond to nutrient and waste product diffusion gradients, modulate their metabolism as a function of their position within the biofilm, contact adjacent cells, and engage in cell–cell communication. Microbes form a biofilm in response to many factors, which may include cellular recognition of specific or non-specific attachment sites on a surface, nutritional cues, or in some cases, by exposure of planktonic cells to sub-inhibitory concentrations of antibiotics.

These adherent cells are frequently embedded within a self-produced matrix of extracellular polymeric substance (EPS). EPS are biopolymers of microbial origin in which biofilm microorganisms are embedded. Contrary to common belief, EPS are certainly more than only polysaccharides. An HPLC analysis showed similar pattern in the polysaccharide production by *P. polymyxa*. According to stander sugar, suggesting that the biopolymer is a homopolysaccharide, which is consistent of various sugars and sugar derivatives such as glucose, galactose, mannose and xylose. Microbes living in a biofilm usually have significantly different properties from free-floating microbe of the same species, as the dense and protected environment of the film allows them to cooperate and interact in various ways. One benefit of this environment is increased resistance to detergents and antibiotics, as the dense extracellular matrix and the outer layer of cells protect the interior of the community.

2.4 Carbon dynamics in soil

Soils are thought to have a finite carrying capacity for carbon (C) based on parent material (texture), ambient temperature, annual precipitation, and net plant primary production. However, microbial transformation and turnover of soil organic matter (SOM) can also influence the magnitude of soil C storage. The microbially mediated reactions fractionate organic material into various reactive C pools such as labile, slowly decomposable, and resistant with respect to their persistence in soil (Jenkinson *et al.*, 1987; Jenkinson, 1990). The importance of C fractionation to soil C storage is evident by the intriguing possibility of manipulating the system into bypassing the labile pool with transformation directly into the slow and resistant pools. In this scenario, one would

hypothesize that more of the C input would be sequestered in the soil for longer periods of time.

Simple C compounds such as glucose and amino acids have been used for decades in SOM studies to investigate the kinetic turnover of compounds, priming effects and C flow through microbial biomass. Regardless of the complexity of the starting substrate, models of organic matter transformations that exclusively track the percent substrate remaining tend to underestimate the initial decomposition rate. Thus, they underestimate the long-term persistence of substrate C and the amount of that C in long-lived soil C fractions (van Veen *et al.*, 1984). In addition, the priming effect is difficult to incorporate into models due to the numerous factors involved in the mechanisms of priming expression (Fontaine *et al.*, 2003). However, the magnitude of the priming effect can have substantial influence on soil C storage of plant residues (Bell *et al.*, 2003).

After decades of investigating substrate decomposition (Ladd *et al.*, 1992), little is understood about the relationship between the initial decomposition of substrates and longer-term SOM formation. However, it has been hypothesized that substrates that initially decompose rapidly may persist longer in the soil, possibly due to substrate quality and environmental conditions.

Soils constitute the largest pool of actively cycling C in terrestrial ecosystems and stock about 1500–2000 Pg C in various organic forms to a depth of 1 m (Nieder and Benbi, 2008). It has been estimated that cultivated cropland soils of the world have lost 41 to 55 Pg C in the past (Houghton and Skole, 1990; Paustian *et al.*, 1998). Several studies have shown that conversion of forest lands to permanent cropping decreases the soil organic carbon stocks, rapidly in the initial years and at a slower rate thereafter, approaching a new equilibrium after 30 to 50 years (Nieder and Benbi, 2008). Arrouays and Pelissier (1994) demonstrated that soil organic carbon (SOC) storage in the 0–50 cm soil horizon declined by about 50% after 35 years of intensive corn cropping in temperate soils. Decline in organic matter content of tropical soils due to continuous cultivation has also been reported in some studies (Brown and Lugo, 1990; Lugo and Brown, 1993). Any modification of land use or land management can induce variations in soil carbon stocks, even in agricultural systems that are perceived to be in a steady state.

2.5 Effect on soil microbial properties

Microbial biomass and soil enzymes are considered as potential indicator of soil quality due to their relationship to soil biology, ease of measurement, rapid responses to changes in soil management and high sensitivity to changes originated by management and environment factor (Marx *et al.*, 2001). The rationale for the use of microbial and biochemical characteristics as soil quality indicators is their central role in cycling of C and N and their sensitivity to change (Nannipieri *et al.*, 1990). Although, it is now well established that a great

many soil microorganisms can produce plant growth regulating substances (Phytohormones), little has been done to exploit the influence of microbially-produced phytohormones on plant growth and development, partly because our knowledge is still so incomplete, but even more so because the processes involved are so complex.

2.5.1 Soil Dehydrogenase Activity

Dehydrogenase (DHA) predominantly an intracellular enzymes, is used as an index of metabolic activity of microbial community in soil. Dehydrogenase activity represents the intracellular flux of electrons to O₂ and is due to the activity of several intracellular enzymes catalyzing the transfer of hydrogen and electron from one compound to another (Nannipeieri *et al.*, 1990).

2.5.2 Soil microbial biomass carbon (SMBC)

The soil microbial biomass is a living part of the soil organic matter. It contributes significantly in nutrient transformation, xenobiotic degradation, as a source and sink of C, N, P and S; improving the physiochemical properties of the soil (Angers *et al.*, 1992). Due to its dynamic character, it has been shown to be a sensitive indicator of differences in soil quality under sustainable cropping systems (Karlen *et.al.*, 1997). It has also been used to compare microbial carbon and nitrogen content and nutrient cycling between soils under different management systems.

The system of rice intensification (SRI) is a promising technique which includes a set of agronomic management practices for rice cultivation that enhances yield, reduces water requirements, raises input productivity, is accessible to smallholders, and is more favorable for the environment than conventional practice with its continuous flooding of paddies and heavy reliance on inorganic fertilization. Cyanobacterial inoculants can be used to reduce applied urea-N by 10% to 50%. However, there are some limitations of using BNF technology in rice cultivation. The growth and N₂ fixation capacity of cyanobacteria may be erratic due to number of factors. Biofilm bio-fertilizers also have shown promising results.

2.6 Research gap

Limited work is done so far on combined effects of more than one cyanobacteria. There is a need to study the effect of different cyanobacterial inoculants on changes in soil aggregation, microbial properties and soil carbon content of soil under different methods of planting. Comparative study of cyanobacterial inoculants under conventional and SRI method and their effect on performance of rice in both method need to be undertaken. With this background an experiment entitled “**Influence of cyanobacteria based inoculants and cultivation methods on rice (*Oryza sativa* L.) yield, soil carbon content and aggregation**” was conducted and results presented and discussed in this thesis.

3. MATERIALS AND METHODS

The present investigation entitled “**Influence of cyanobacteria based inoculants and cultivation methods on rice (*Oryza sativa* L.) yield, soil carbon content and aggregation**” was conducted at the Research Farm of the Indian Agricultural Research Institute in *kharif* season of 2011. The details of materials used and methods employed during the course of investigation are described in this chapter.

3.1. Experimental site

The experiment was conducted in the ‘Genetics D Block’ at the Research Farm of Indian Agricultural Research Institute (IARI), New Delhi, situated at latitude of 28°40’ N and longitude of 77°12’ E, altitude of 228.6 m above the mean sea level.

3.2. Climate and weather conditions

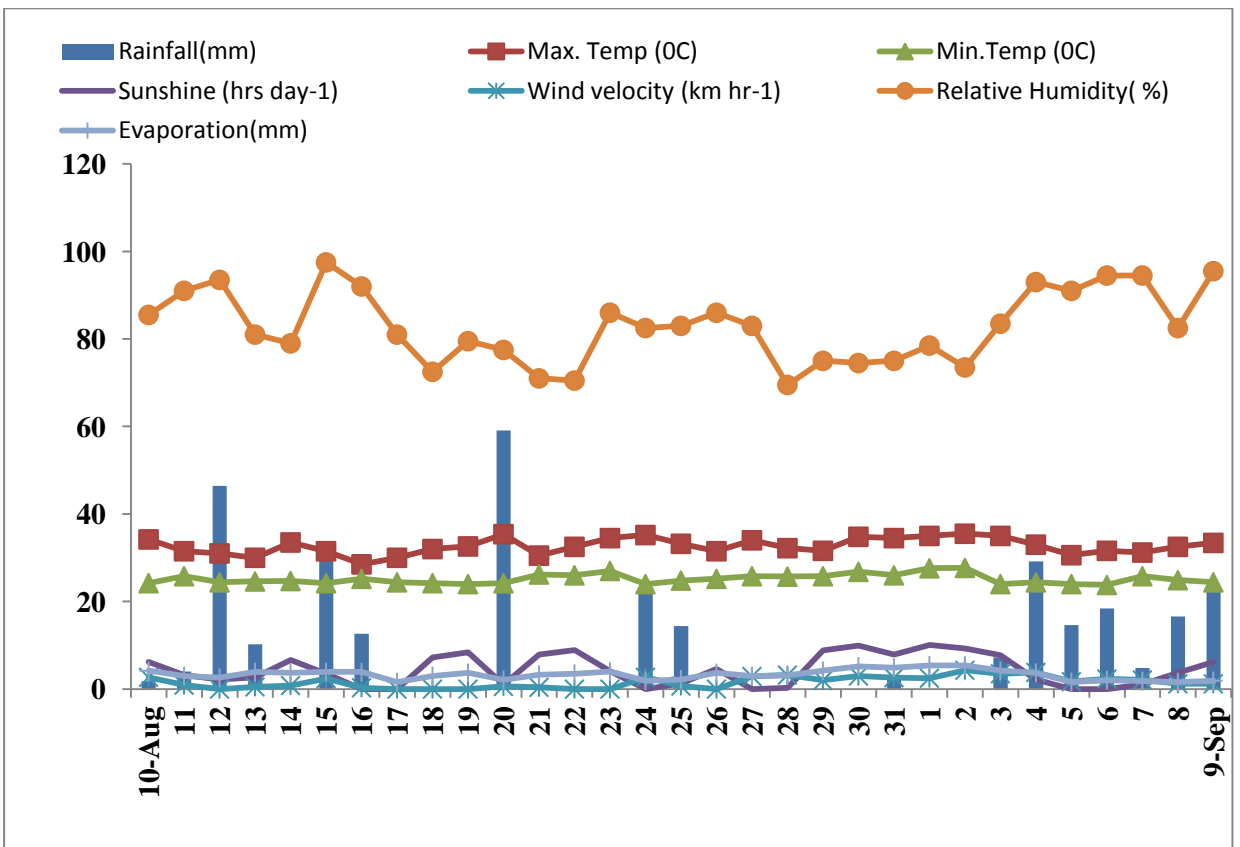
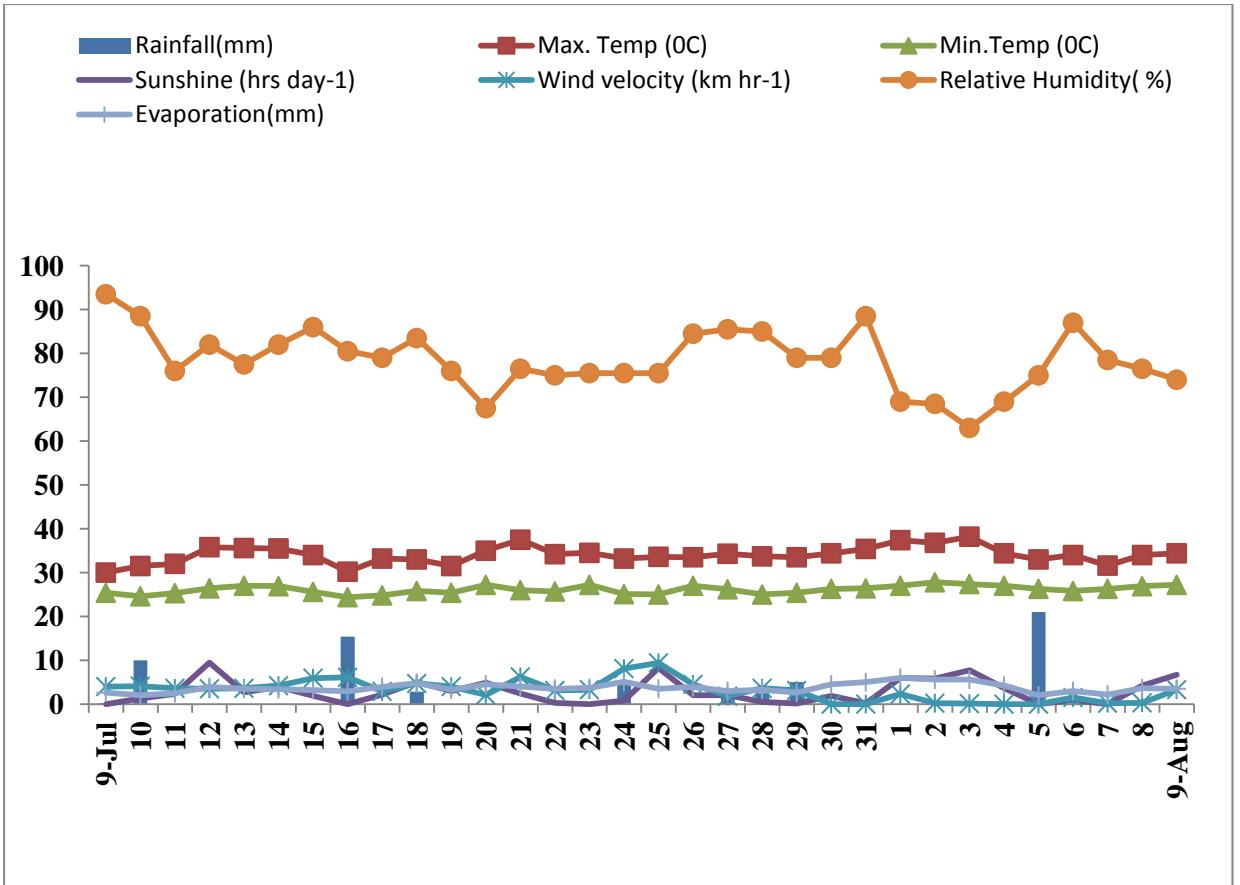
The climate of Delhi is of sub-tropical and semi-arid type with hot and dry summer and cold winter and falls under the agro-climatic zone ‘Trans-Gangatic plains’. Summer months, May and June are the hottest with maximum temperature ranging between 41 and 46°C, while there is a drop in temperature from September onward. January is the coldest month of the year with a minimum temperature ranging from 5 to 7°C. The mean annual normal rainfall is 650 mm, while July and August are the wettest months. The annual mean pan evaporation is about 850 mm. The detailed weather data during rice crop growing season (June - October) recorded at the meteorological observatory of Indian Agricultural Research Institute, New Delhi, are given in Appendix I and depicted in Fig. 3.1. During June to October maximum and minimum temperatures ranged between 43.6⁰C (8 June) to 29.4⁰C (30 October) and 11.9⁰C (28 October) to 29⁰C (7 June), respectively. The year 2011, had less than normal rainfall (539.4 mm during June to October) out of which 43.8, 226.8 and 163.6 mm rainfall which comprised 80.5 % of the total rainfall was received during July, August and September months, respectively. There were 6, 9 and 11 numbers of rainy days during July, August and September months, respectively. There was no rainfall in October. There was late onset of monsoon and July month had much less rainfall than normal. Daily evaporation was also higher during rainy season. Difference in maximum and minimum daily relative humidity was higher in June and October months. In other weather parameters many fluctuations were not recorded over the normal conditions of Delhi.

3.3 Cropping history of the experimental site

The cropping history of the experimental field indicated that only rice-wheat cropping system was practiced in the experimental field during last ten years in *kharif-rabi* seasons.



Plate 1 Location of field



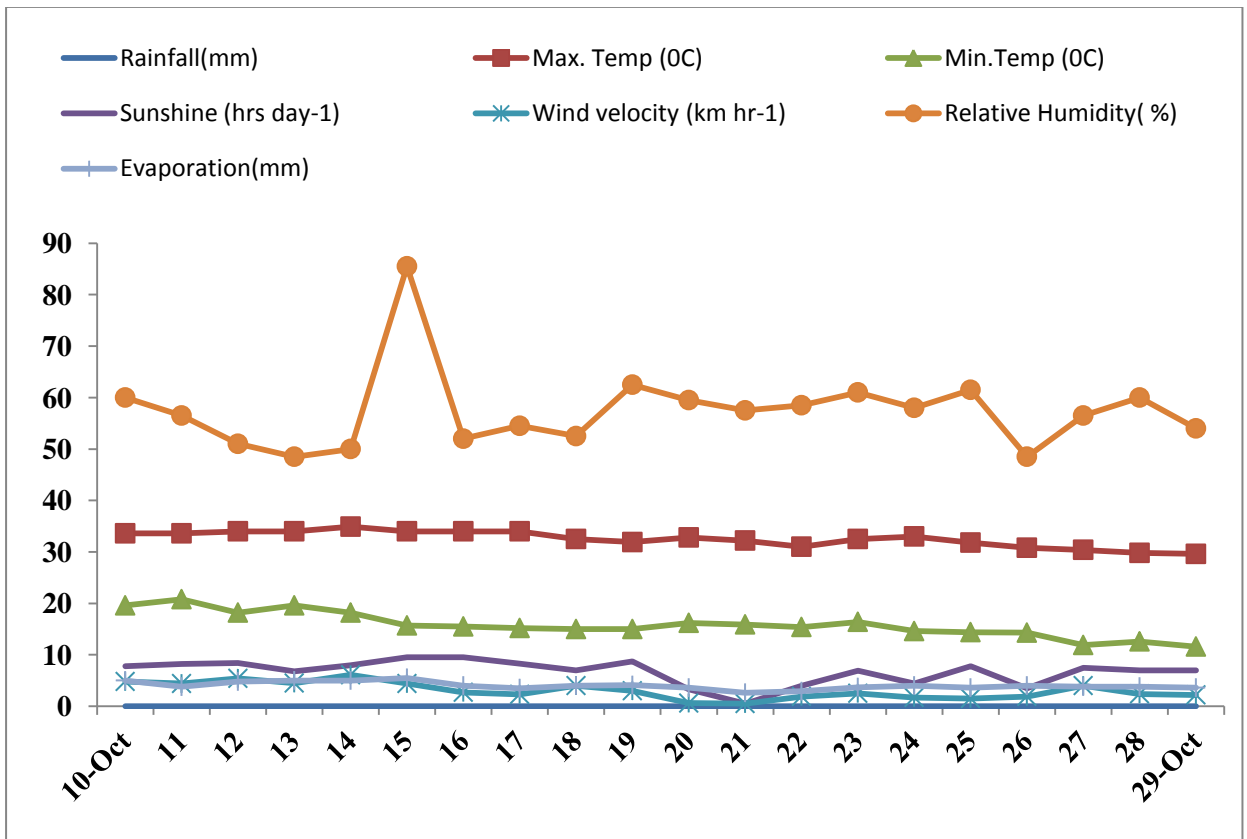
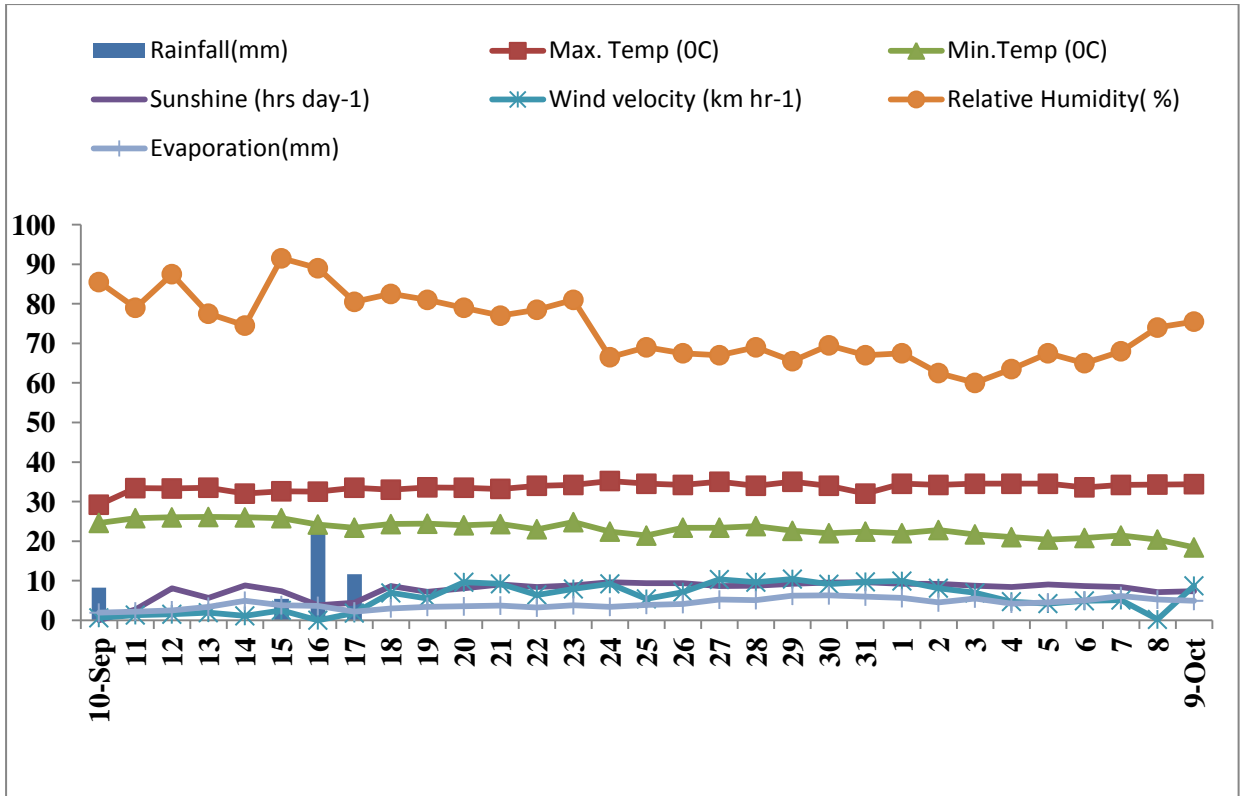


Fig. 3.1: Daily weather conditions during experimental period (July-October, 2012)



Plate 2 general view of experimental field

Table 3.1 Physical, chemical and biological properties of the experimental soil at the start of the experiment

Particulars	Content
A. Physical characteristics	
Mechanical composition	
(Hydrometer method, Bouyoucos, 1962)	
Sand (%)	51.80
Silt (%)	22.20
Clay (%)	26.00
Moisture at 1/3 atmospheric tension (%)	25.40
(Pressure plate apparatus, Richards and Weaver, 1943)	
Moisture at 15 atmospheric tension (%)	12.20
(Pressure plate apparatus, Richards and Weaver, 1943)	
Bulk density (0-15 cm layer) (g/cc)	1.48
B. Chemical characteristics	
Organic carbon (%)	0.53
(Walkley and Black method, Jackson, 1973)	
Alkaline permanganate oxidizable N (Subbiah and Asija., 1956)	134.9
Available phosphorus (P kg ha ⁻¹)	15.90
(Olsen's method, Jackson, 1973)	
Available potassium (K kg ha ⁻¹)	260.83
(Flame photometer method, Jackson, 1973)	
Available iron (Fe mg kg ⁻¹)	10.0
(DTPA extraction, Lindsey and Norwell, 1973)	
P ^H (1:2.5 soil: water)	8.1
(Elico pH meter, Piper, 1950)	
C. Biological characteristics	
MBC (µg microbial biomass carbon g ⁻¹ soil) (Nunan <i>et al.</i> , 1998)	105.45
Dehydrogenase activity (µg TPF g ⁻¹ soil day ⁻¹) (Casida <i>et al.</i> , 1964)	5.90
Acetylene reductase activity (n mole ethylene g ⁻¹ hr ⁻¹)	0.70
Soil chlorophyll (Nayak <i>et al.</i> , 2004)	1.03

3.4 Soil

Composite soil samples were collected from different sites and analyzed for mechanical composition, important physico-chemical and biological properties at the start of the experiment. The soil of the experimental field was sandy clay loam in texture, with moderate water holding capacity and well leveled topography. The data on mechanical composition, physico-chemical and biological properties of soil of the present study are presented in Table 3.1. The soil was medium in organic C, low in N and medium in available P and high in available K.

3.5 Characteristics of rice variety ‘Pusa Basmati 1401’ used in experiment

‘Pusa *Basmati* 1401’ was developed through hybridization between Pusa *Basmati*-1/Pusa 1121-92-8-2-7-1 followed by pedigree method of selection. Pusa 1401 (IET 18005) has been identified with superior quality *Basmati* variety than the traditional *Basmati* variety. It has been found better than the most popular traditional *Basmati* ‘Taraori *Basmati*’ in grain, cooking, eating quality and grain yield (34%). It is a semi-dwarf (85 cm) and will not lodge like traditional tall *Basmati* varieties. Its cooked rice appearance, fluffiness, kernel length after cooking (16.07 mm) with least breadth-wise swelling and pleasant aroma is attractive to catch the consumers' attention in the International and domestic market. In the panel tests conducted over three years it was found better in comparison to ‘Taraori *Basmati*’ and ‘Pusa *Basmati*-1’ in overall acceptability (Singh *et al.*, 2007).

3.6 Agronomic management

3.6.1 Nursery bed preparation

For conventional transplanting (CT) raised (about 10 cm) nursery beds of 1.0 m width and 4.0 m length were prepared. Around these beds 50 cm drainage cum irrigation channels were prepared. Nursery beds were prepared keeping in view the requirement of 600 m² nursery to transplant 1.0 ha area. On these nursery beds 1:1 soil: decomposed FYM mixture was spread to 4-5 cm thickness as well as recommended dose of N,P and K was applied. Pre soaked (for 24 hours) and incubated seeds in moist gunny cloth for another 24 hours were broadcasted uniformly on nursery beds. Nitrogen, P₂O₅, K₂O and ZnSO₄ were applied to the nursery @ 1.0, 0.5, 0.5 kg and 100 g per 100 m² through urea, single superphosphate, muriate of potash and zinc sulphate, respectively.

Water was allowed frequently into channels that were formed around the nursery beds. Sprinkling of water using rose cane was also done as and when required. These practices were aimed at better aeration as well as avoiding the flooding of nursery bed. Weeding was done twice during the period. For SRI, seedlings were sown 12 days before proposed transplanting date. Seed rate @ 6 kg ha⁻¹ was taken for SRI. After broadcasting the

sprouted seeds 1:1 soil-FYM mixture was spread in a thin layer of 1-2 cm. Irrigation were given as and when required to keep the nursery bed moist.

3.6.2 Land preparation

Main field was pre irrigated, ploughed with tractor-drawn disc plough followed by harrowing after the soil reached to tilth conditions and leveling was done with land leveler. Puddling operation was done twice with using wheel mounted tractor drawn puddler. Water was drained out of the field at the time of transplanting. Field was demarcated into plots providing irrigation and drainage channels according to layout plan (Fig. 3.2).

3.6.3 Transplanting

For conventional transplanting seedling were uprooted and cleaned with water and then transplanted in prepared field. For SRI, before lifting the seedlings, nursery beds were irrigated. Seedlings were not pulled from the nursery rather they were lifted from nursery bed along with soil, intact seed sac and roots, while lifting the seedlings, precautions were taken to have least damage to roots. These uprooted seedlings were carried to transplanting site, separated carefully to avoid any damage to roots. These separated seedlings were immediately transplanted in the main field with gentle placement but not with harsh pushing, which may revert root direction to cause transplanting trauma. On the day of transplanting there was no standing water in the field. One seedling of 12 days old was transplanted hill⁻¹ in a square pattern with spacing of 25 cm x 25 cm. Marked ropes at equal distances were used to achieve square planting.

3.6.4 Gap filling

Gap filling was done wherever seedlings failed to establish. Under conventional transplanting gap filling was done 5 days after transplanting. Under SRI, gap filling was done twice (5 and 10 DAT). For the purpose of gap filling, some seedlings used for respective methods were kept alongside of the channels at the time of transplanting. For gap filling, seedlings along with soil intact roots were used.

3.6.5 Harvesting and threshing

Two rows on all the sides were removed as border and remaining hills were harvested as net plot. For proper air drying the harvested crop was left in the field for a week's time and then weighed total biomass plot wise. Threshing was done manually.

3.7 Experimental details

Layout

The experiment was laid out in a Split Plot Design (SPD) with 14 treatment combinations and three replications. The plan of layout is presented in Fig. 3.2 and treatment details are given below:

Treatment details:

- **Rice cultivation method :**

C1: Traditional transplanting

C2: SRI method

- **Cyanobacterial inoculants:**

B1: N₀

B2: Recommended dose of N (120 kg ha⁻¹)

B3: Compost application (1/3 N) + 2/3 N *

B4: Biofilm based BGA biofertilizer + 2/3 N *

B5: BGA + PGPR + 2/3 N*

B6: Compost based BGA mixture inoculant + 2/3 N*

B7: *Multani mitti* based BGA inoculant + 2/3 N*

*Nitrogen was applied through chemical fertilizer (urea)

Recommended dose of P (60 kg P₂O₅ ha⁻¹) and K (60 kg K₂O ha⁻¹) were applied to all the treatments.

B4 treatment contains *Anabaena spp.* and *Pseudomonas spp.*

B5 treatment contains *Ochromobacterium spp.* (PR-10) and *Anabaena spp.* (CR-2).

B6 treatment contains *Anabaena spp.* (BF1, BE4) and *Noctoc spp.* (BF2, BF3).

B7 treatment contains *Anabaena spp.*, *Noctoc spp.*, *Tolipothrics spp.* and *Allocera spp.*

Experimental design:

Design : Split-plot design

No. of treatment combinations: 14

Gross plot size : 6.2 m x 2.46 m (15.25 m²)

No. of replication : 3

Variety : 'Pusa Basmati 1401'

3.8 Agronomic management in SRI *vis-a-vis* conventional transplanting (CT)

Management practices	SRI	Conventional management
Seedling age at transplanting	Transplanted young seedlings of 12 days after germination (before the fourth-leaf stage). Precautions were taken to have least damage to roots while lifting the seedlings. Seedlings were transplanted in the main field with gentle placement. On the day of transplanting there was no standing water in the field.	Transplanted seedlings of 21 days after germination. On the day of transplanting standing water was maintained in the field.
Plant spacing and density	Transplanted one seedling per hill quickly after uprooting in a square pattern at 25 cm x 25 cm spacing.	Transplanted two seedlings per hill in a square pattern at 20 x 10 cm spacing
Weed control	To control weeds, mechanical hand weeder (rotating hoe) was used. This churned up the soil surface and buried young weeds. First weeding was done 12 days after transplanting, to pre-emptively curb weed growth and to aerate and fertilize the soil, and it was done two times before the canopy closes.	Did hand-weeding twice (25 and 45 days after transplanting) to control weeds. Early flooding also helped in reducing weed population.
Water management	Under SRI management, plots were not kept continuously flooded. Instead, mostly aerobic soil conditions are maintained throughout the vegetative growth period, either by adding small amounts of water regularly, or by alternate wetting and drying (AWD). After panicle initiation, a thin layer of water (1-2 cm) was maintained, until 20 days before harvest, when the plots were drained.	3-5 cm water level was maintained in the plots after transplanting. After panicle initiation, plots were kept under saturated soil conditions up to maturity.
Nutrient management	As per treatments nutrient dose was applied.	Nutrient doses were applied as per treatments.

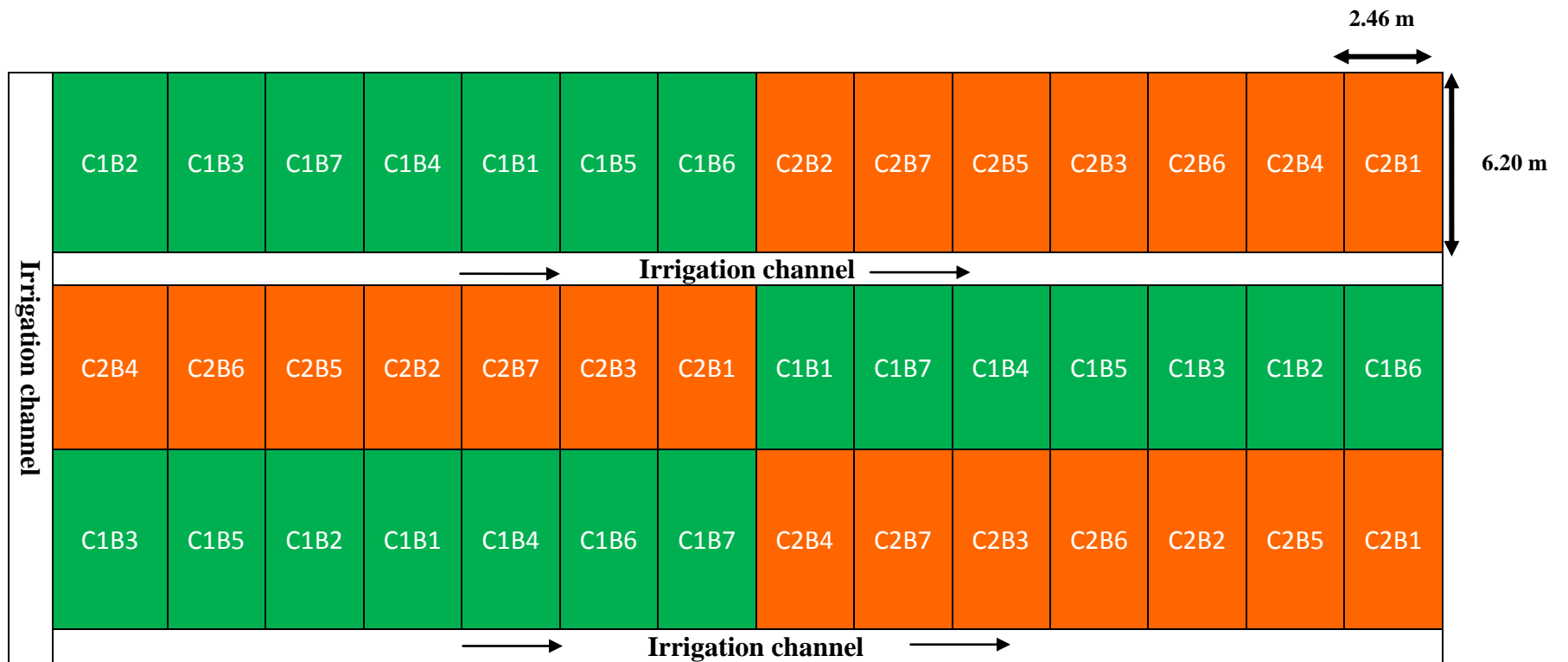


Fig. 3.2: Layout of experimental field



3.9 Details of field operations

The details of the field operations are given in Table 3.2.

Table 3.2 Field operations during experimentation

Name of operations	Date
1. Preparation of nursery	
a. For conventional transplanting	15.06.11
b. For SRI	24.06.11
2. Sowing of nursery	
a. For conventional transplanting	17.06.11
b. For SRI	26.06.11
3. Ploughing of main field	02.07.11
4. Puddling and levelling	04.07.11
5. Layout, bunding & fertilizer application as basal	06.07.11
7. Transplanting (both in CT and SRI)	09.07.11
6. Gap filling	14.07.11 & 19.07.2011
7. Hand weeding	20.07.11
8. Top dressing of N fertilizer in CT and SRI(25 DAT)	04.08.11
9. Biometric observation	09.08.11
10. 2 nd weeding	29.07.10
11. Top dressing of N fertilizer	03.09.11
12. Biometric observations	09.09.11
13. Biometric observations	09.10.11
14. Biometric observations at harvest	29.10.11
15. Harvesting	04.11.11
16. Threshing and drying	09.11.11

3.10 Biometrical observations

3. 10.1 Growth characteristics

3. 10.1.1 Plant height

The heights of the five sample plants was measured from the ground level up to the tip of the apical leaf at 30, 60 and 90 DAT and from ground level to the tip of the panicle/ leaf at harvest and mean values were computed.

3. 10.1.2 No. of tillers

Number of tillers of one m² area were counted at 30, 60 and 90 DAT and at harvest and averaged into tillers m⁻².

3. 10.1.3 Dry matter accumulation

Plants of five hills were collected by cutting at ground level from the second row onwards leaving border rows on all sides. These plants were oven dried at 60°C and dry weight was recorded in g m⁻² at 30, 60 and 90 DAT.

3. 10.1.4 Leaf area index (LAI)

Leaf area index expresses the ratio of total leaf area to the total ground area in which the crop is grown. The leaf area was measured from the leaves of five sampled plants by using leaf area meter (Model LI-COR-3100) and leaf area index was worked out using the following formula:

$$\text{LAI} = \frac{\text{Total leaf area (cm}^2\text{) plant}^{-1}}{\text{Land area (cm}^2\text{) plant}^{-1}}$$

3. 10.2 Yield attributes

3. 10.2.1 Number of panicles

Panicles were counted from selected one m² area where total tillers were counted and mentioned as the number of panicles m⁻².

3. 10.2.2 Panicle length

The length of the panicle was measured from a sample of ten panicles drawn at random from the marked five hills. The length was measured from neck at the tip of the panicle and average panicle length was counted.

3. 10.2.3 Number of grains panicle⁻¹

The grains of ten panicles selected for measuring the panicle length were separated out, cleaned and grains were counted using a grain counting machine. Mean value of number of grains was computed.

3. 10.2.4 Weight of panicle

The weight of 10 panicles was measured and means panicle weight computed and expressed as g per panicle.

3. 10.2.5 1000-grain weight

One thousand seeds were counted with the help of electric seed counter, the sample was weighed and the weight was expressed as test weight in gram.

3. 10.2.6 Grain and straw yield

The net plots (two rows on all side were removed as border) were harvested and sun dried for 5 days in the field and then the total biomass yield was recorded. After threshing, cleaning and drying, the grain yield was recorded. Straw yield was obtained by subtracting grain yield from total biomass yield. Yields were expressed in t ha⁻¹.

3. 10.2.7 Harvest index

Harvest index is the proportion (percentage) of filled grain to the whole above-ground biomass sample expressed in weight. Harvest index was calculated by using the following expression:

$$\text{Harvest index} = \frac{\text{Economic yield (kg)}}{\text{Biological yield (kg)}} \times 100 = \frac{\text{Grain yield}}{\text{Grain yield} + \text{Straw yield}} \times 100$$

3.11 Chemical analysis

3. 11.1 Soil chemical analysis

Soil samples were collected from five locations in each replication using soil auger from the depth of 0-15 and made a composite soil sample, before the start of the experiment. The composite soil samples were analyzed for organic carbon, soil pH, total N, available N, available phosphorus, available potassium, available Fe, mechanical composition and soil physical constants as per methods given in Table 3.1.

3. 11.2 Soil biochemical parameters

3. 11.2.1 Soil Acetylene reductase activity (ARA)

Fresh soil sample was collected from field at initial, 60 DAT and at harvest using steel auger and place in a glass voils. Also keep similar glass voils with 20 ml water same as control. The voils tightly sealed using stoppers and injected with 3.5ml (10% of gas phase) of acetylene (C_2H_2) after removing equivalent amount of air. The samples are than incubated under field condition for a period of three hours and then ethylene produced is measured using gas chromatography. The ARA value is expressed per unit dry weight of soil. Calculation of ethylene concentration in standard ethylene gas: Standard ethylene gas is supplied as compressed gas in air tight cylinder. Std. ethylene comes as mixture with inert gas like argone (Ar). To calculate ethylene concentration formula used as follows:

ARA (n mole ethylene g^{-1} soil hr^{-1}) = 0.1643 x concentration read by gas chromatography.

3.11.2.2 Dehydrogenase activity

Dhydrogenase activity of soil samples was estimated by the method described by Casida *et al.* (1964). For estimation of dehydrogenase activity three reagents namely Triphenyl tetrazolium chloride (TTC), Methanol (AR grade) and standard triphenyl formazan ($100 \mu g ml^{-1}$) were taken.

For estimation of dehydrogenase activity, 6 g fresh air-dried soil sample was saturated with 1.0 ml freshly prepared TTC (3% w/v) solution in a screw capped test tube. Pinch (0.1 g) of $CaCO_3$ was added and care was taken that no air bubble remains during packing of soil sample. It was rotated gently by shaking. These test tubes were incubated at $28 \pm 1^\circ C$ (28-

30°C) for 24 hours. After 24 hours TPF extracted (pink layer). Methanol (10 ml) was added to these test tubes and rotated it well for 1 minute. Supernatant was taken out carefully after allowing standing for 10 minutes. Absorbance of supernatant was recorded by Spectrophotometer at 485 nm. A standard curve was prepared with TPF (0-50 µg ml⁻¹). Concentration of TPF in sample was calculated with standard curve. Dehydrogenase activity was calculated and expressed in terms of µg TPF liberated µg TPF g⁻¹ soil day⁻¹.

3. 11.2.3 Microbial Biomass Carbon (MBC)

Microbial biomass carbon in soil samples was estimated by the method described by Nunan *et al.* (1998). For its estimation two reagents namely Chloroform and 0.5M K₂SO₄ were taken.

For estimation of MBC, 17.5 g soil sample was taken in a in closed-capped bottle and added 1.0 ml of chloroform and fumigated these samples. And also one non fumigated set prepared in a 250 ml flask. Then these samples were incubated in dark for 24 hours. After 24 hours of incubation, evaporated the chloroform at 50°C in BOD i.e. open the caps for next 20-24 hours. After that 70 ml 0.5M K₂SO₄ added to samples and put on shaker for 30 minutes. Supernatant was taken out by filtering the samples with Whatman No. 42 filter paper. Absorbance of supernatant was recorded immediately for both fumigated and non-fumigated at 280 nM. Soil microbial biomass carbon (MBC) was calculated by using the formula given below:

$$\begin{array}{r}
 \text{MBC} \\
 (\mu\text{g of MBC g}^{-1} \text{ of soil}) = \frac{\text{O.D. with fumigated sample} - \text{O.D. with unfumigated sample}}{17.5} \times 15487
 \end{array}$$

3. 11.2.4 Soil chlorophyll

Soil chlorophyll was assayed using pre weighed soil cores (from 0-20 cm depth); acetone: DMSO (1:1) was added at a rate of 4 ml g⁻¹ soil. The contents were thoroughly shaken and incubated for 48-96 h in the dark at room temperature. Intermittent shaking every 24 h extracted the chlorophyll completely. Optical density values were taken at 663, 645, 630 and 775 nm and the *chlorophyll a* concentration determined (Nayak *et al.*, 2004).

3. 11.3 Plant chemical analysis

3. 11.3.1 Nitrogen concentration and uptake

Plant samples collected at harvest were dried in hot air oven at 60°C for 24 hours after sun drying. The oven dried samples of plants and air dried sample of grain having moisture content (12%) were ground to pass through 40 mesh sieve in a Macro-wiley mill. From each replication, 0.5 g samples were taken for chemical analysis to determine the N concentration.

The nitrogen concentration in grain of rice samples was determined by modified Kjeldahl method (Jackson, 1973). N uptake was calculated by multiplying grain and straw yields with corresponding values of N concentration and expressed in kg ha^{-1} .

3. 11.3.2 Crude protein content in grain (%)

Crude protein content in grain was obtained by multiplying N concentration with a factor 5.95. This factor is based on the nitrogen content (16.8%) of the major rice protein (glutelin).

3. 11.3.3 Fe, Zn, Mn and Cu concentration and uptake in plant samples

The Fe, Zn, Mn and Cu in dry matter of grain and straw of rice crop were determined as per the procedure described by Prasad *et al.*, (2006). Zn, Mn, Cu and Fe uptake in rice grain and straw were calculated by multiplying the grain and straw yield of rice with their respective concentrations and expressed in g ha^{-1} . The total Zn, Mn, Cu and Fe uptake were determined by adding their uptake in grain for the individual treatment.

3.12. Soil carbon content

The determination of oxidisable carbon was repeated using 5 and 10 ml. of concentrated sulphuric acid instead of the 20 ml specified by Walkley and Black (1934). The resulting three acid-aqueous solution ratios of 0.5:1, 1:1 and 2:1 (which corresponded respectively to 12 N, 18 N, and 24 N of H_2SO_4) allowed comparison of oxidizable organic carbon extracted under increasing oxidizing conditions. The amount of oxidizable organic carbon determined using 5,10,and 20 ml. of concentrated sulphuric acid when compared with total carbon concentration allowed separation of total organic carbon into three fractions of decreasing oxidizability: Fraction 1 (12 N H_2SO_4)—organic carbon oxidizable under 12 N H_2SO_4 , Fraction 2 (18 N-12 N H_2SO_4)—the difference in oxidizable organic carbon extracted between 18 N and 12 N H_2SO_4 and Fraction 3(24 N-18 NH_2SO_4)—the difference in oxidizable organic carbon extracted between 24N and 18 NH_2SO_4 .The 24 NH_2SO_4 is equivalent.

3.13. Soil aggregation property

Soil sample was collected from field at 60 DAT at soil moisture near to field capacity and air dried. Air-dried samples were passed through 8 mm sieves. Wherever clods were bigger and the amount of soil passing through was small, larger clods are gently broken apart at their natural cleavages. A known amount of soil sample (50 g) is immersed in water for short period of time for wetting .The wetted sample is sieved through the nest of sieves in Yoders apparatus (Yoder, 1936) and mean weight diameter (MWD) was determined as:

$$\text{MWD} = \sum X_i W_i$$

Where, W_i is the proportion of each aggregate class i in relation to the weight of soil sample taken for analysis and X_i is the mean diameter of the class (mm).

3.14 Economics

Cost of cultivation was calculated based on the prevailing market prices of the inputs. Gross returns were calculated based on the grain and straw yield and their prevailing market prices during the respective crop season. Net returns were calculated by subtracting cost of cultivation from gross returns. Benefit: Cost (B: C) ratio was calculated by dividing the net return from cost of cultivation.

$$\text{Net returns (₹)} = \text{Gross returns (₹)} - \text{cost of cultivation (₹)}$$

3.15 Statistical analysis

The data relating to each character were analyzed as per the procedure of analysis of variance and significance of a Split Plot Design (SPD), tested by “F” test. Standard Error of Means (SEM_±) and Least Significant Difference (LSD) at 5% level of significance were worked out for each parameter.

Effect of planting methods and cyanobacterial inoculants on plant growth, productivity and economics of rice

Abstract

A field experiment was conducted during the rainy (*kharif*) season of 2011 at the Research Farm of the IARI, New Delhi to study the influence of methods of crop establishment and cyanobacterial inoculants on plant growth, productivity and economics of rice (cv. 'Pusa *Basmati* 1401'). The experiment was laid out in split plot design (SPD) with fourteen treatments combinations comprising two methods of crop establishment viz., conventional transplanting (CT) and system of rice intensification (SRI) and seven cyanobacterial inoculants viz., N₀; Recommended dose of N (120 kg ha⁻¹); Compost application (1/3 N) + 2/3 N; Biofilm based BGA bio-fertilizer + 2/3 N; BGA + PGPR + 2/3 N; Compost based BGA mixture inoculant + 2/3 N and *Multani mitti* based BGA inoculant + 2/3 N. 2/3 N (80 kg ha⁻¹) and recommended dose of nitrogenous fertilizer (urea) were applied in 3 splits through urea and equal doses of P and K fertilizers were applied. In CT and SRI, 21 and 12 days old seedlings, respectively were transplanted in puddled fields. Results showed that the effect of conventional and SRI method was significant at 30 DAT on the rice plant growth parameters like plant height, tillers, dry matter accumulation and leaf area index (LAI). However, both methods showed statistically at par growth at 60 and 90 DAT and at crop harvest. The treatment BGA+PGPR and 2/3 nitrogen through fertilizer had highest values in all the parameters studied. But this treatment was statistically at par over with three treatment including RDN (120 kg ha⁻¹), compost application (1/3 N) + 2/3 N through fertilizer and compost based BGA mixture inoculant + 2/3 N through fertilizer. Treatment BGA+PGPR and 2/3 nitrogen through chemical fertilizer showed highest yields but these were statistically at par with recommended dose of N, compost application (1/3 N) + 2/3 N through fertilizer and compost based BGA mixture inoculant + 2/3 N through fertilizer. Grain and straw yield under these four treatments was significantly higher than Biofilm based BGA biofertilizer + 2/3 N through fertilizer and *Multani mitti* based BGA inoculant + 2/3 N through fertilizer. Higher net return and B : C ratio was given in SRI compared to conventional method. Highest net return and B : C ratio was given by BGA + PGPR + 2/3 N through fertilizer followed by the recommended dose of N through fertilizer.

Keywords: Biofilm based inoculants, conventional transplanting, economics of rice, plant growth promoting rhizobacteria (PGPR), rice yield, system of rice intensification (SRI).

4.1 Introduction

Rice (*Oryza sativa* L.) is the major food crop of nearly half of the world's population. To keep pace with the increasing demand for food, increases in rice yield and production will be required. Rice plants require large amounts of mineral nutrients including nitrogen (N) for their growth, development and grain production. Rice crops remove around 16–17 kg N for the production of each ton of rough rice including straw (Datta, 1981; Sahrawat, 2000). However, most of the rice soils of the world are deficient in N, so fertilizer N applications are required to meet a rice crop's N demand. Generally, urea is applied as the N source for rice production. But the efficiency of added urea-N is very low, often only 30–40%, in some cases even lower (Choudhury and Khanif, 2001; Choudhury *et al.*, 2002a). This low N-use efficiency is mainly due to denitrification, NH₃ volatilization and leaching losses (De Datta and Buresh, 1989). NH₃ volatilization and denitrification cause atmospheric pollution through the production of greenhouse gases like N₂O and NH₃ (Reeves *et al.*, 2002). NO₃⁻ leaching causes groundwater toxicity (Shrestha and Ladha, 1998). These problems are of great concern to soil and environmental scientists around the world. Alternate sources of N like bio-fertilizers and plant growth-promoting rhizobacteria (PGPR) should be applied to minimize these problems.

Biological N fixation (BNF) technology can play an important role in substituting for commercially available N fertilizer use in rice culture, thus reducing these environmental problems to some extent. Use of bio-fertilizers can prevent the depletion of the soil organic matter. Rice crops are grown in both wetland and upland cultures. However about 85% of the total rice-cropped area is under wetland culture. Cyanobacteria (blue-green algae) are photosynthetic prokaryotic microorganisms capable of fixing atmospheric N₂ using sunlight as the sole energy source (Stewart, 1980). Cyanobacteria can play a major role in improving the soil environment in addition to N fixation. Wetland rice fields can provide an ideal condition for the growth of cyanobacteria, which accumulate 19–28 kg N ha⁻¹ per crop, and can reduce the use of urea fertilizer in rice culture by 25–35% (Hashem, 2001). Experimental results at the IRRI (Los Banos, Philippines) revealed that the amount of N accumulation by cyanobacteria varies from a few to 50 kg N ha⁻¹ crop⁻¹ among different soils (Roger and Ladha, 1992). The literature on the beneficial effects of cyanobacteria on the growth and yield of rice is voluminous (Ladha and Reddy, 1995; Kannaiyan *et al.*, 1997; Kennedy and Islam, 2001).

A biofilm is an aggregate of microorganisms in which cells adhere to each other and/or to a surface. Biofilms may form on living or non-living surfaces, and represent a prevalent mode of microbial life in natural and industrial (Hall-Stoodley *et al.*, 2004). The biofilmed bio-fertilizers improve nitrogen fixing symbiosis in crops and could contribute directly to soil N fertility in long term. The biofilms based on cyanobacteria could be

beneficial for rice cultivation. The beneficial effects of PGPR have been well demonstrated in both greenhouse and field conditions (Yanni *et al.*, 1997; Biswas *et al.*, 2000a, 2000b).

The traditional method of rice cultivation requires a large amount of labour, water, and energy. Water and labour, however, are becoming increasingly scarce in the region, raising questions about the sustainability of rice production and the overall environment. In the north-west (NW)-IGP of India, increasing use of groundwater for rice cultivation has led to a decline in the water table by 0.1 to 1.0 m year⁻¹, resulting in water scarcity and increased cost for pumping water (Hira, 2009, Rodell *et al.*, 2009) and (Humphreys *et al.*, 2010). Implementation of the Mahatma Gandhi National Rural Employment Guarantee Act, introduced by the Indian government in 2005 (GOI, 2011), promising 100 days of paid work in people's home village, has been creating a labour scarcity in Punjab, Haryana and some other states as rice transplanting in this region is dependent on migrant labourers from eastern Uttar Pradesh and Bihar. Since rice is primarily grown by transplanting seedlings in flooded puddled fields, it requires a large amount of water (~150 cm), of which 15–20 cm (Singh *et al.*, 2001) is used only for puddling. Alternatives like system of rice intensification (SRI) are required to save water and increase crop, water, and labour productivity (Satyanarayana, 2005; Uphoff *et al.*, 2011; Suryavanshi *et al.*, 2012).

The system of rice intensification (SRI) offers many insights into ways that production can be increased efficiently and water saved by managing rice crops with more attention to biology and agro-ecology, as summarized in a recent joint publication by Africare, Oxfam America and the WWF/ICRISAT Project, 2010 (Uphoff *et al.*, 2011). The validity of SRI concepts and methods has been seen now in 42 countries, from Panama to the Democratic People's Republic of Korea. The governments in China, India, Indonesia, Cambodia, and Vietnam, where two-thirds of the world's rice is produced, have come to accept and promote these alternative methods based on their own evaluations and experience. Over the last several years, there has been a substantial increase in the scientific knowledge base built up to account for the remarkable results being reported (Uphoff *et al.*, 2011). Despite the apparent yield barriers, the quest for higher yield potential continues (Peng *et al.*, 1999; Fagaria, 2007). With above background experiment was conducted to understand the effect of cultivation methods and cyanobacterial inoculants on biological processes, functions and interactions on improving rice yield.

4.2 Materials and methods

The experiment was conducted during rainy (*khari*) season (June to October) of 2011 at the Research Farm of Indian Agricultural Research Institute, New Delhi, situated at a latitude of 28°40' N and longitude of 77°12' E, altitude of 228.6 meters above the mean sea level (Arabian Sea). The mean annual rainfall of Delhi is 650 mm and more than 80% generally occurs during the south-west monsoon season (July-September) with mean annual

evaporation 850 mm. During rice cropping season (June to October) maximum and minimum temperatures ranged between 43.6⁰C (8 June) to 29.4⁰C (30 October) and 11.9⁰C (28 October) to 29⁰C (7 June), respectively. The year 2011, had less than normal rainfall (539.4 mm during June to October) out of which 43.8, 226.8 and 163.6 mm rainfall which comprised 80.5 % of the total rainfall was received during July, August and September months, respectively. There were 6, 9 and 11 numbers of rainy days during July, August and September months, respectively. There was no rainfall in October. There was late onset of monsoon and July month had much less rainfall than normal. Daily evaporation was also higher during rainy season. Difference in maximum and minimum daily relative humidity was higher in June and October months. In other weather parameters many fluctuations were not recorded over the normal conditions of Delhi. The soils of experimental field had 134.9 kg ha⁻¹ alkaline permanganate oxidizable N (Subbiah and Asija., 1956), 15.90 kg ha⁻¹ available P (Olsen's method, Jackson, 1973), 260.83 kg ha⁻¹ 1 N ammonium acetate exchangeable K (Prasad *et al.*, 2006) and 0.53 % organic carbon (Walkley and Black, 1934). The pH of soil was 8.1 (1: 2.5 soil and water ratio).

The experiment was laid out in split plot design with fourteen treatments combinations comprising of two methods of rice cultivation viz., conventional transplanting (CT) and system of rice intensification (SRI) and seven cyanobacterial inoculants viz., N₀; Recommended dose of N₁₂₀ (120 kg ha⁻¹); Compost application (1/3 N) + 2/3 N; Biofilm based BGA bio-fertilizer + 2/3 N; BGA + PGPR + 2/3 N; Compost based BGA mixture inoculant + 2/3 N and *Multani mitti* based BGA inoculant + 2/3 N. 2/3 N (80 kg ha⁻¹) and recommended dose of N (120 kg ha⁻¹) was applied through urea and applied in three splits. *Basmati* rice variety 'Pusa *Basmati* 1401' was taken in the experiment and these treatments were replicated three times. In CT and SRI 21 and 12 days old seedlings, respectively were transplanted in puddled fields.

The experimental field was disc-ploughed twice and leveled and after puddling. Seedlings of rice variety were transplanted under conventional transplanting at 20 cm × 10 cm spacing keeping 2 seedlings hill⁻¹. For SRI, 12 days old seedlings were gently transplanted in main field at the spacing of 25 cm x 25 cm keeping one seedlings hill⁻¹. In conventional transplanting method 5 cm water was maintained from transplanting to grain filling stage of crop and later only moist soil conditions were maintained. However in SRI, 2 cm water was applied and alternate wetting and drying conditions was maintained throughout the cropping season. In the experiment equal dose of P₂O₅ and K₂O ha⁻¹ @ 60 kg ha⁻¹ were applied in all the plots. The gross plot size was 6.2 m × 2.46 m for each treatment. Rotary weeder was used in SRI for weed management and interculture while in CT manual hand weeding was done twice at 25 and 45 DAT. Crop was harvested in the last fortnight of October.

Observations on plant growth, yield attributes, yield and economics were recorded.

Plant height of the rice was measured from the base of the plant at ground surface to the tip of the tallest leaf/ panicle. Numbers of tillers were noted by counting from the sampling unit. Dry matter accumulation was calculated from five hills taken from sampling area and after oven drying at $60 \pm 2^{\circ}\text{C}$ dry weight was calculated in g m^{-2} . Selected ten panicles were taken for panicle length and these panicles were also used to record the weight of the panicles, and number of grains panicle⁻¹ was also counted. The 1,000-filled grains, taken from sampled panicles, were first counted by a seed counter and then weighed to compute the 1,000-grain weight. After harvesting, threshing, cleaning and drying the grain yield was recorded at 14% moisture. Straw yield was obtained by subtracting grain yield from the total biomass yield. Yield was expressed in t ha^{-1} . Gross and net returns were calculated based on the grain and straw yield and their prevailing market prices of rice during the respective crop season. B:C ratio was calculated by dividing the net returns from total cost of cultivation.

All the data obtained from the experiment conducted under split plot design were statistically analyzed using the *F*-test as per the procedure given by Gomez and Gomez (1984). LSD values at $P = 0.05$ were used to determine the significance of difference between treatment means. Interaction effect was also statistically analysed in the same design.

4.3 Results

4.3.1 Plant growth parameters

4.3.1.1 Plant height

Effect of cultivation methods and cyanobacterial inoculants on the rice plant height was measured by taking observations at 30, 60 and 90 days after transplanting (DAT) and at harvest stage (Table 4.1; Fig. 4.1 and 4.2). It was observed that at 30 DAT conventional transplanting (CT) method recorded significantly higher plant height (49.5cm) as compared to system of rice intensification (SRI) (46.6 cm). However, no significant difference was observed in plant height under CT and SRI at 60 and 90 DAT and at harvest stage. At harvest stage plant height at CT and SRI was 105.7 cm and 104.5cm, respectively. In sub-plots, treatments containing cyanobacterial inoculants and recommended dose of nitrogen (RDN) showed significantly higher plant height over N control however, differences between the effect of cyanobacterial inoculants along with 2/3 N and recommended dose of N was non-significant at 30, 60 and 90 DAT and at harvest. Treatment containing BGA+PGPR and 2/3 nitrogen through chemical fertilizer shows highest plant height (84.9 cm) but this treatment was statistically at par with three treatment including RDN, compost application (1/3 N) + 2/3 N through chemical fertilizer and compost based BGA mixture inoculant + 2/3 N through chemical fertilizer. All these four treatments were statistically superior over treatments containing Biofilm based BGA biofertilizer + 2/3 N through chemical fertilizer and *Multani mitti* based BGA inoculant + 2/3 N through chemical fertilizer at all the observations.

Treatment N₀ recorded lowest height being 42.7 cm, 67.8 cm, 84.33 cm and 86.7cm at 30, 60 and 90 DAT and at harvest stage, respectively while the respective values for the treatment BGA+PGPR and 2/3 N through recorded were 49.3 cm, 84.9 cm, 108.4 cm and 111.4 cm.

4.3.1.2 Plant tillers

Tiller is the important factor which decides the numbers of panicle m⁻² and ultimately yield of rice. Plant tillers as affected by the method of cultivation and cyanobacterial inoculants are measured at 30, 60 and 90 DAT and at harvest stage is given Table 4.2 and Fig. 4.3. The difference in number of tillers in CT (212) was statistically significant over SRI (198.7) at 30 DAT. However, this superiority of CT over SRI in tiller count was not recorded at 60, 90 DAT and harvest. Number of tillers in both the methods were statistically non-significant at 60, 90 DAT and harvest. Number of tillers at harvest was 356.5 and 353.1 in CT and SRI, respectively. It was recorded that the tillers per hill were higher in SRI compared to CT. This might be due to higher spacing in SRI (25 x 25 cm) which makes more space available for growth of individual hill compared to CT. However, on m⁻² basis, both methods recorded same number of tillers. In subplots, there is no significant difference in tiller number among all cyanobacterial inoculants and recommended dose of N (RDN) however, all these treatments showed statistically significant tillers over no nitrogen at 30 DAT.

Difference in number of tillers within sub-plot treatments was observed only after 60 days of transplanting. At 60 DAT, treatment having BGA+PGPR and 2/3 N through chemical fertilizer showed highest tillers (379.7). But this treatment was statistically non-significant over other three treatment including RDN (120 kg ha⁻¹), Compost application (1/3 N) + 2/3 N through chemical fertilizer and Compost based BGA mixture inoculant + 2/3 N through chemical fertilizer recorded 373.7, 369.2 and 367.5 tillers, respectively. All these four treatments were statistically superior over Biofilm based BGA biofertilizer + 2/3 N through chemical fertilizer and *Multani mitti* based BGA inoculant + 2/3 N through chemical fertilizer which recorded 348.5 and 346.2 tillers, respectively. Similar trend was observed in number of tillers at 90 DAT and at harvest. N₀ recorded lowest number of tillers which were 275.2, 281.7 and 247.7 at 60 and 90 DAT and at harvest, respectively. Treatment BGA+PGPR and 2/3 N through chemical fertilizer recorded highest number of tillers being 379.7, 406.3 and 382.8 at 60 DAT, 90 DAT and at harvest, respectively.

Interaction between planting methods and cyanobacterial inoculants on number of tillers m⁻² was found significant at harvest stage (Table 4.2.1). Within conventional method, BGA+PGPR and 2/3 N through chemical fertilizer showed highest tillers (391) which was statistically significant over treatment containing no nitrogen. This treatment also stands significant over no nitrogen treatment in SRI method. In SRI method, same treatment showed a highest tiller (374.7) which was statistically significant over treatment containing no nitrogen. This treatment significantly higher tillers over no nitrogen treatment in conventional

method. But BGA+PGPR and 2/3 N through chemical fertilizer stands non-significant in both conventional and SRI method of cultivation.

4.3.1.3 Dry matter accumulation

Significantly higher dry matter accumulation was recorded in conventional transplanted rice (160 g m^{-2}) as compared to SRI method (149.8 g m^{-2}) at 30 DAT but at 60 and 90 DAT difference in dry matter accumulation was non-significant (Table 4.3). Dry matter of plants increased with age. In sub-plots, dry matter accumulation was non-significant among different cyanobacterial inoculants and RDN at 30 DAT. Treatment having nitrogen control recorded lowest dry matter being 141.1, 221.8 and 473.1 g m^{-2} at 30, 60 and 90 DAT, respectively. Treatment containing BGA+PGPR and 2/3 N through fertilizer showed highest dry matter accumulation being 305.2 g and 760.6 g at 60 and 90 DAT.

But dry matter accumulation at 60 and 90 DAT due to this treatment was statistically non-significant with other three treatment including RDN (120 kg ha^{-1}), compost application ($1/3 \text{ N}$) + $2/3 \text{ N}$ through fertilizer and Compost based BGA mixture inoculant + $2/3 \text{ N}$ through fertilizer. All these four treatments were statistically superior than treatment having Biofilm based BGA biofertilizer + $2/3 \text{ N}$ through fertilizer and Multani mitti based BGA inoculant + $2/3 \text{ N}$ through fertilizer.

Table 4.1 Effect of planting methods and cyanobacterial inoculants on rice plant height

Treatment	Plant height (cm)			
	30 DAT	60 DAT	90 DAT	At harvest
Planting method				
Conventional	49.5	81.8	103.9	105.7
SRI	46.6	80.4	101.6	104.2
SEm±	0.28	0.39	0.72	0.74
LSD (P=0.05)	1.72	NS	NS	NS
Cyanobacterial inoculants				
N ₀	42.7	67.8	84.3	86.7
Recommended dose of N (120 kg ha^{-1})	49.2	84.6	107.8	110.7
Compost application ($1/3 \text{ N}$) + $2/3 \text{ N}$	49.1	84.4	107.3	110.4
Biofilm based BGA biofertilizer + $2/3 \text{ N}$	48.7	81.2	102.4	103.6
BGA + PGPR + $2/3 \text{ N}$	49.3	84.9	108.4	111.4
Compost based BGA mixture inoculant + $2/3 \text{ N}$	49.1	84.4	106.6	109.8
Multani mitti based BGA inoculant + $2/3 \text{ N}$	48.5	80.4	102.7	102.2
SEm±	0.63	0.95	1.35	1.52
LSD (P=0.05)	1.85	2.78	3.95	4.45
Cyanobacterial inoculants x Planting method				
SEm±	0.89	1.34	1.91	2.15
LSD (P=0.05)	Sig.	Sig.	Sig.	Sig.

Table 4.2 Effect of planting methods and cyanobacterial inoculants on number of tillers m⁻²

Treatment	Plant Tillers (m ⁻²)			
	30 DAT	60 DAT	90 DAT	Harvest Stage
Planting Method				
Conventional transplanting	212.0	353.8	379.0	356.5
SRI	198.7	349.0	376.0	353.1
SEm±	1.62	2.13	3.37	1.75
LSD (P=0.05)	9.88	NS	NS	NS
Cyanobacterial inoculants				
N ₀	142.8	275.2	281.7	247.7
Recommended dose of N (120 kg ha ⁻¹)	217.5	373.7	397.2	378.5
Compost application (1/3 N) + 2/3 N	215.4	369.2	395.3	376.2
Biofilm based BGA biofertilizer + 2/3 N	214.5	348.5	386.5	362.8
BGA + PGPR + 2/3 N	220.2	379.7	406.3	382.8
Compost based BGA mixture inoculant + 2/3 N	214.2	367.5	394.5	375.0
<i>Multani mitti</i> based BGA inoculant + 2/3 N	212.7	346.2	383.8	360.8
SEm±	3.43	5.76	5.23	4.01
LSD (P=0.05)	10.00	16.81	15.05	11.71
Cyanobacterial inoculants x Method				
SEm±	4.84	8.14	7.29	5.67
LSD (P=0.05)	NS	NS	NS	Sig.

Table 4. 2.1 Interaction between planting methods and cyanobacterial inoculants on number of tillers m⁻² at crop harvest

Planting methods	Bacterial inoculants						
	T ₁	T ₂	T ₃	T ₄	T ₅	T ₆	T ₇
Tillers m ⁻² at harvest							
Conventional	254.0	383.0	368.0	364.3	391.0	381.0	354.3
SRI	241.3	374.0	384.3	361.3	374.7	369.0	367.3
S. Em±	5.67						
LSD (P=0.05)	16.55						

Interaction between planting method and cyanobacterial inoculants on dry matter accumulation was found significant at 30 DAT but non-significant both at 60 and 90 DAT (Table 4.3.1). Within conventional method, BGA+PGPR and 2/3 N through chemical fertilizer showed highest dry matter accumulation (165.6 g) which was statistically significant over treatment containing no nitrogen. This treatment also stands significant over no nitrogen treatment in SRI method. In SRI method, same treatment showed highest dry matter accumulation (151.9 g) which was statistically significant over treatment containing no N.

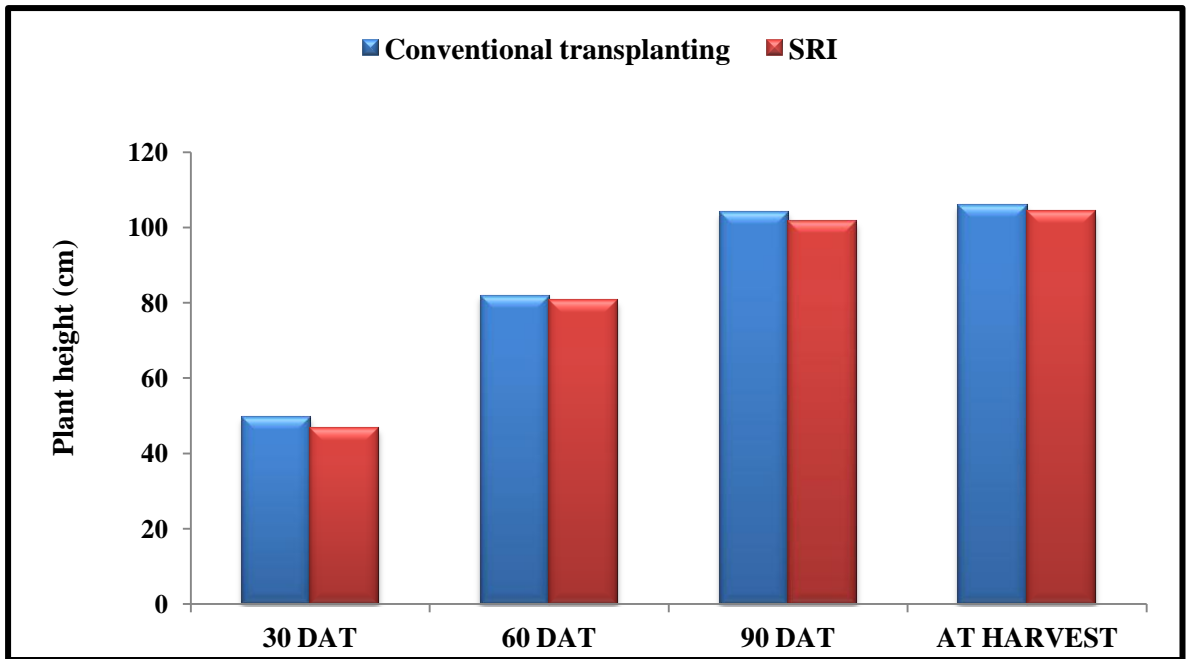


Fig. 4.1: Plant height as influenced by method of planting over crop growth period

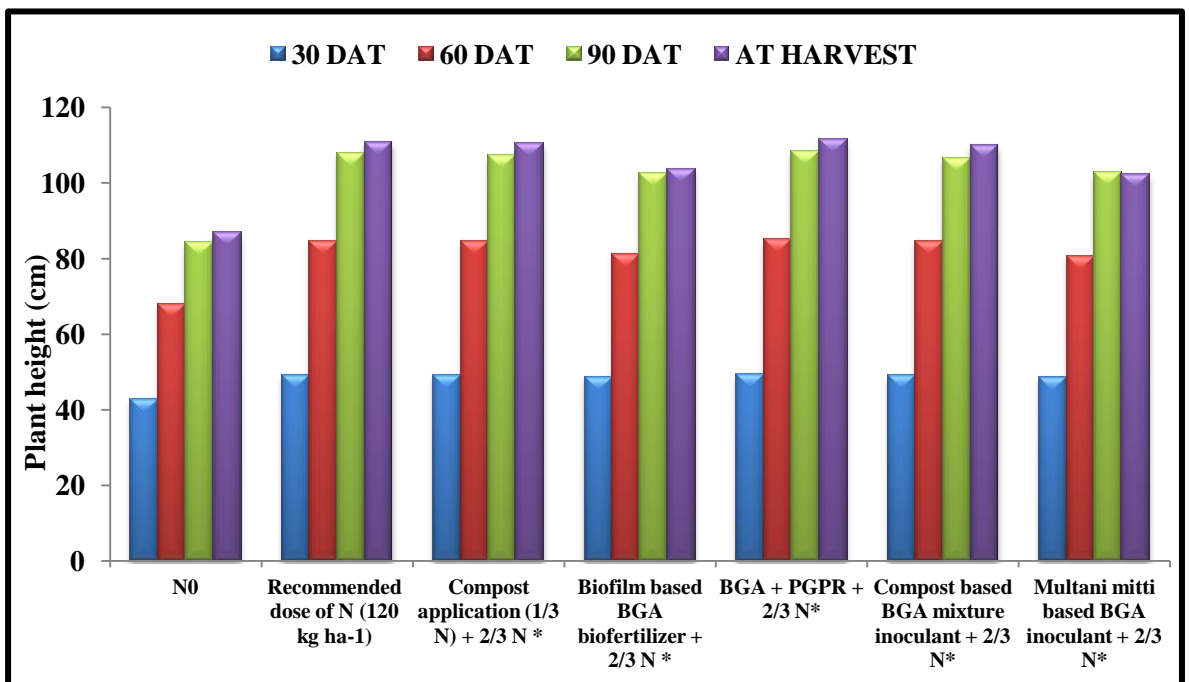


Fig. 4.2: Plant height as influenced by cyanobacterial inoculants over the crop growth Period

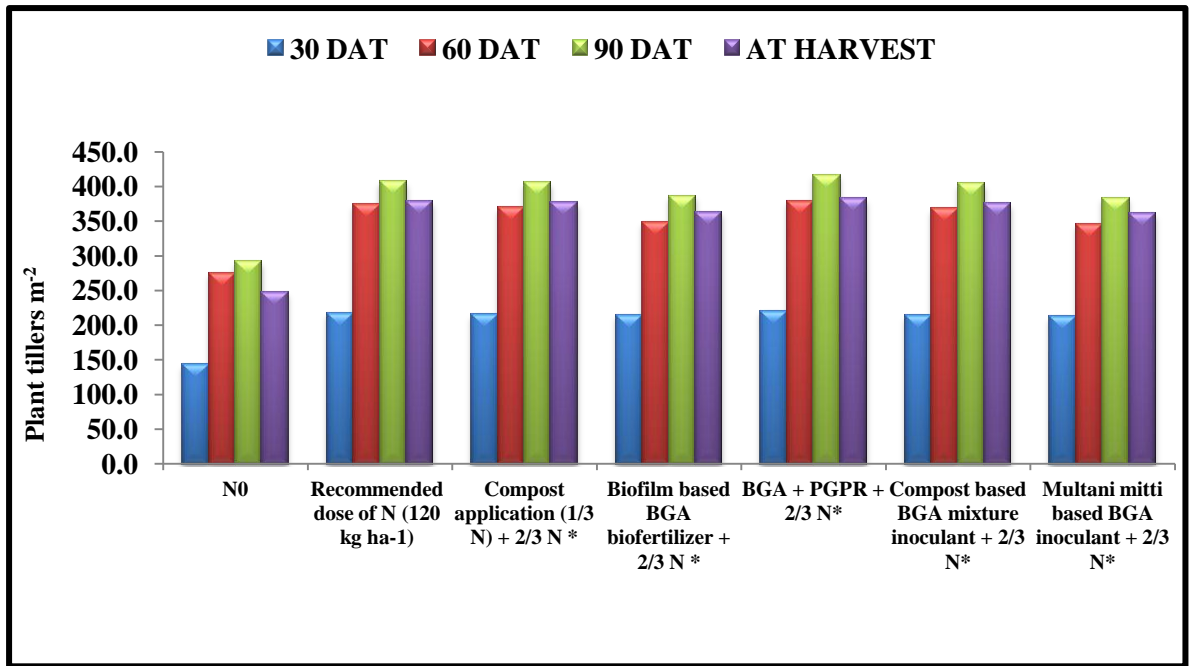


Fig. 4.3: Effect of cyanobacterial inoculants on number of tillers m⁻² over the crop growth

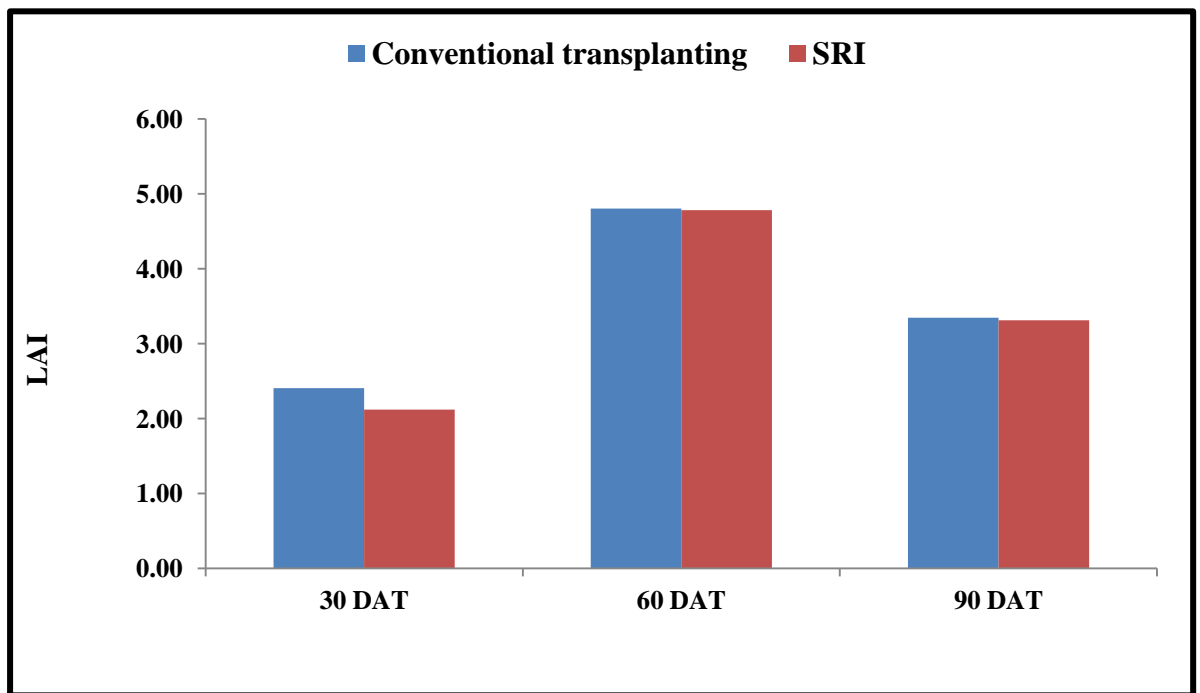


Fig. 4.4: Leaf area index (LAI) as affected by method of planting over crop growth period



Plate 3 Rice crop in SRI and Conventional method



Plate 4 Rice crop in no nitrogen (N_0)



Plate 5 Rice crop at physiological maturity

BGA+PGPR and 2/3 N through chemical fertilizer showed significantly higher dry matter accumulation in conventional method over same treatment in SRI of cultivation.

4.3.1.4 Leaf area index (LAI)

Leaf area index (LAI) indicates the photosynthetic capacity of plant. The change in LAI as influenced by methods of cultivation and cyanobacterial inoculants was measured at 30, 60, 90 DAT (Table 4.4; Fig. 4.4). LAI was found significantly higher in CT over SRI at 30 DAT. However, LAI did not show any significant difference between both the methods at 60 and 90 DAT. LAI was increased up to 60 DAT and declined thereafter. Highest LAI values recorded with CT and SRI were 4.80 and 4.78, respectively. Treatment containing BGA+PGPR and 2/3 N through fertilizer showed highest LAI which was 5.26 and 3.81 at 60 and 90 day after transplanting. But this treatment had non-significant LAI over other three treatment including RDN (120 kg ha⁻¹), Compost application (1/3 N) + 2/3 N through fertilizer and Compost based BGA mixture inoculant + 2/3 N through fertilizer at 60 and 90 DAT. All these four treatment were statistically superior over Biofilm based BGA biofertilizer + 2/3 N through fertilizer and Multani mitti based BGA inoculant + 2/3 N through fertilizer.

Interactions between planting methods and cyanobacterial inoculants on LAI were found significant at 30, 60 and 90 DAT (Table 4.4.1). At 60 DAT, within conventional method, BGA+PGPR and 2/3 N through chemical fertilizer showed LAI (5.17) which was statistically significant over treatment containing no nitrogen. This treatment also stands significant over no nitrogen treatment in SRI method. In SRI method, same treatment showed LAI (5.34) which was statistically significant over treatment containing no nitrogen.

4.3.2 Yield attributes

Study of yield attributes is a prerequisite for predicting yielding ability of crop. Differences in different yield attributes viz., panicle number m⁻², weight and length of panicles, number of grain panicle⁻¹ and test (1000- grain) weight of rice grain were non-significant between CT and SRI (Table 4.5). Numbers of panicle m⁻² were 356.5 and 353.1 in CT and SRI, respectively while respective values for grain per panicle were 157.2 and 158.2. Equal test weight of 21.4 g was recorded in both the methods. Among the sub-plot treatments, all yield attributing characters found significantly higher with cyanobacterial inoculants and RDN over N control. The treatment BGA+PGPR and 2/3 nitrogen through fertilizer had highest values in all the parameters. But this treatment was statistically at par over with three treatment including RDN, Compost application (1/3 N) + 2/3 N through fertilizer and Compost based BGA mixture inoculant + 2/3 N through fertilizer. All these four treatment were statistically superior over treatment having Biofilm based BGA biofertilizer + 2/3 N through fertilizer and Multani mitti based BGA inoculant + 2/3 N through fertilizer in respect to number,

Table 4.3 Effect of planting methods and cyanobacterial inoculants on dry matter accumulation

Treatment	Dry matter accumulation g m ⁻²		
	30 DAT	60 DAT	90 DAT
Planting Method			
Conventional transplanting	160.0	287.5	711.2
SRI	149.8	282.5	709.3
SEm±	0.32	1.83	2.07
LSD (P=0.05)	1.95	NS	NS
Cyanobacterial inoculants			
N ₀	141.1	221.7	473.1
Recommended dose of N (120 kg ha ⁻¹)	157.9	301.3	754.0
Compost application (1/3 N) + 2/3 N	157.3	297.6	751.3
Biofilm based BGA biofertilizer + 2/3 N	156.3	287.5	743.3
BGA + PGPR + 2/3 N	158.8	305.2	760.6
Compost based BGA mixture inoculant + 2/3 N	157.0	295.9	750.3
<i>Multani mitti</i> based BGA inoculant + 2/3 N	156.2	285.5	739.1
SEm±	0.99	5.04	5.06
LSD (P=0.05)	2.90	14.70	14.78
Cyanobacterial inoculants x Method			
SEm±	1.40	7.12	7.15
LSD (P=0.05)	Sig.	NS	NS

Table 4.4 Effect of planting methods and cyanobacterial inoculants on leaf area index

Treatment	Leaf area index (LAI)		
	30 DAT	60 DAT	90 DAT
Planting Method			
Conventional transplanting	2.41	4.80	3.35
SRI	2.12	4.78	3.31
SEm±	0.023	0.032	0.025
LSD (P=0.05)	0.137	NS	NS
Cyanobacterial inoculants			
N ₀	1.22	3.20	1.61
Recommended dose of N (120 kg ha ⁻¹)	2.48	5.23	3.73
Compost application (1/3 N) + 2/3 N	2.48	5.17	3.69
Biofilm based BGA biofertilizer + 2/3 N	2.38	4.82	3.43
BGA + PGPR + 2/3 N	2.50	5.26	3.81
Compost based BGA mixture inoculant + 2/3 N	2.41	5.12	3.66
<i>Multani mitti</i> based BGA inoculant + 2/3 N	2.37	4.75	3.39
SEm±	0.067	0.087	0.066
LSD (P=0.05)	0.196	0.253	0.194
Cyanobacterial inoculants x Method			
SEm±	0.095	0.122	0.094
LSD (P=0.05)	Sig.	Sig.	Sig.

Table 4. 3.1 Interaction between planting methods and cyanobacterial inoculants on dry matter accumulation m⁻² at 30 DAT

Planting methods	Bacterial inoculants						
	T ₁	T ₂	T ₃	T ₄	T ₅	T ₆	T ₇
Conventional	149.8	164.3	165.3	158.5	165.6	162	154.9
SRI	132.3	151.5	149.3	154	151.9	152.0	157.4
S. Em±				1.40			
LSD (P=0.05)				4.10			

Table 4. 4.1 Interaction between planting methods and cyanobacterial inoculants on leaf area index at 60 DAT

Planting methods	Bacterial inoculants						
	T ₁	T ₂	T ₃	T ₄	T ₅	T ₆	T ₇
leaf area index at 60 DAT							
Conventional	3.56	5.14	4.92	4.27	5.17	5.70	4.87
SRI	2.85	5.32	5.42	5.38	5.34	4.53	4.63
S. Em±				0.122			
LSD (P=0.05)				0.357			

weight and length of panicles and number of grain panicle⁻¹. Test weight difference was at par in all the treatments including treatment having no nitrogen application.

Interaction between planting method and cyanobacterial inoculants on the attributes like number, weight and length of panicles and number of grain panicle⁻¹ were found significant. Interaction was non-significant in case of 100 grain weight. Within conventional method, BGA+PGPR and 2/3 N through chemical fertilizer showed significantly higher number of grain panicle⁻¹ (171.5) which was statistically significant over treatment containing no nitrogen (Table 4.5.1). In SRI method, same treatment showed significantly higher number of grain panicle⁻¹ (171.7) which was statistically significant over treatment containing no nitrogen.

4.3.3 Grain and straw yield

Effect of conventional and SRI methods of planting on grain and straw yield and harvest index was found non-significant (Table 4.6; Fig. 4.5 a, b). Grain yield was 4.37 t ha⁻¹ and 4.34 t ha⁻¹ in conventional and SRI method of planting, respectively; While the respective values for straw yield were 8.21 and 8.11 t ha⁻¹. Harvest index for both the methods was same

(0.35). In sub-plots treatments, grain and straw yields were significantly higher with cyanobacterial inoculants and RDN over N control. Treatment containing BGA+PGPR and 2/3 nitrogen through chemical fertilizer showed highest grain and straw yield which was 4.69 t ha⁻¹ and 8.56 t ha⁻¹, respectively. But effect of this treatment was statistically at par with other three treatment including recommended dose of N (120 kg ha⁻¹), Compost application (1/3 N) + 2/3 N through fertilizer and Compost based BGA mixture inoculant + 2/3 N through fertilizer. All these four treatment were significantly better than the Biofilm based BGA biofertilizer + 2/3 N through fertilizer and *Multani mitti* based BGA inoculant + 2/3 N through fertilizer. N control had the lowest grain (3.18 t ha⁻¹) and straw (8.21 t ha⁻¹) yields.

Interaction between planting method and cyanobacterial inoculants on grain and straw yield were found significant (Table 4.6.1). Within conventional method, BGA+PGPR and 2/3 N through chemical fertilizer showed higher grain yield (4.62 t ha⁻¹) which was statistically significant over treatment containing no nitrogen. This treatment also stands significant over no nitrogen treatment in SRI method. In SRI method, same treatment showed higher grain yield (4.76 t ha⁻¹) which was statistically significant over treatment containing no nitrogen. BGA+PGPR and 2/3 N through chemical fertilizer are on par with each other in both conventional method and SRI method of cultivation.

4.3.4 Economics of rice cultivation

Analysis of economics of cost of cultivation and return of rice cultivation on the prevailing prices of input and output are shown in Table 4.7. Cost of cultivation was found lower in all treatment of SRI method as compared conventional method. This might be due to low cost on nursery, seed and irrigation. Cost of cultivation was higher in the treatments having compost application compared to other treatments. Net return and B : C ratio in all the treatments were higher under SRI method compared to conventional method. Treatment having BGA + PGPR + 2/3 N (through fertilizer) under SRI method gave the highest net return (₹ 74, 596) followed by the recommended dose of N (RDN) through fertilizer (₹ 74556) + 2/3 N through chemical fertilizer. B : C ratio was highest in the same treatments having ₹ 1.91 and ₹ 1.89. All treatment showed higher B : C ratio in SRI method.

4. 4 Discussion

Significantly higher plant height, tillers, dry matter accumulation and LAI was recorded in conventional transplanting (CT) method compared to system of rice intensification (SRI) at 30 days after transplanting (DAT). Higher plant growth in CT over SRI might be due to shorter/ younger (12 days old) seedling of rice planted in SRI, whereas in CT 21 days old seedlings were planted. Younger seedling in SRI took longer time to absorb transplanting shock and mortality of seedlings was 15 % higher in SRI compared to CT. Adverse climatic condition prevailing during 2011 wet season having almost no rainfall in

Table 4.5 Effect of planting methods and cyanobacterial inoculants on yield attributes of rice

Treatments	No. of panicle m ⁻²	Weight of panicle (g)	Length of panicle (cm)	Grains per panicle	Test weight (g)
Planting method					
Conventional transplanting	356.5	2.8	28.7	157.2	21.4
SRI	353.1	2.8	28.7	158.2	21.4
SEm±	1.75	0.02	0.08	0.70	0.11
LSD (P=0.05)	NS	NS	NS	NS	NS
Cyanobacterial inoculants					
N ₀	247.7	1.8	26.1	125.7	21.2
Recommended dose of N(120 kg ha ⁻¹)	378.5	3.1	29.6	168.0	21.5
Compost application (1/3 N) + 2/3 N	376.2	3.0	29.4	165.7	21.4
Biofilm based BGA biofertilizer + 2/3 N	362.8	2.7	28.4	154.2	21.3
BGA + PGPR + 2/3 N	382.8	3.2	30.0	171.6	21.7
Compost based BGA mixture inoculant + 2/3 N	375.0	2.9	29.4	164.9	21.5
<i>Multani mitti</i> based BGA inoculant + 2/3 N	360.8	2.7	28.3	154.0	21.3
SEm±	4.01	0.07	0.25	3.20	0.33
LSD (P=0.05)	11.71	0.20	0.74	9.35	0.95
Cyanobacterial inoculants x Method					
SEm±	5.67	0.10	0.35	4.53	0.46
LSD (P=0.05)	Sig.	Sig.	Sig.	Sig.	NS

Table 4.6 Effect of planting method and cyanobacterial inoculants on grain and straw yield and harvest index

Treatment	Grain yield (t ha ⁻¹)	Straw yield (t ha ⁻¹)	Harvest index (%)
Planting Method			
Conventional transplanting	4.37	8.21	0.35
SRI	4.34	8.11	0.35
SEm±	0.015	0.057	0.002
LSD (P=0.05)	NS	NS	NS
Cyanobacterial inoculants			
N ₀	3.18	6.96	0.31
Recommended dose of N (120 kg ha ⁻¹)	4.68	8.54	0.36
Compost application (1/3 N) + 2/3 N	4.64	8.48	0.36
Biofilm based BGA biofertilizer + 2/3 N	4.38	8.06	0.35
BGA + PGPR + 2/3 N	4.69	8.56	0.36
Compost based BGA mixture inoculant + 2/3 N	4.63	8.51	0.35
<i>Multani mitti</i> based BGA inoculant + 2/3 N	4.29	8.00	0.35
SEm±	0.07	0.12	0.01
LSD (P=0.05)	0.20	0.35	0.02
Cyanobacterial inoculants x Method			
SEm±	0.09	0.17	0.008
LSD (P=0.05)	Sig.	Sig.	Sig.

Table 4.5.1 Interaction between planting methods and cyanobacterial inoculants on grain number panicle⁻¹

Planting methods	Bacterial inoculants						
	T ₁	T ₂	T ₃	T ₄	T ₅	T ₆	T ₇
grain number panicle ⁻¹							
Conventional	112.4	161.9	181.3	149.0	171.5	176.4	147.9
SRI	139.0	174.1	150.0	159.3	171.7	153.3	160.0
S. Em±				4.5			
LSD (P=0.05)				13.2			

Table 4. 6.1 Interaction between planting methods and cyanobacterial inoculants on grain yield (t ha⁻¹)

Planting methods	Bacterial inoculants						
	T ₁	T ₂	T ₃	T ₄	T ₅	T ₆	T ₇
Grain yield (t/ha)							
Conventional	3.53	4.55	4.82	4.46	4.62	4.26	4.36
SRI	2.83	4.82	4.46	4.29	4.76	5.00	4.22
S. Em±				0.09			
LSD (P=0.05)				0.29			

July month resulting in continuous higher temperature caused mortality of tender seedling in early age in SRI. However, plant height under CT and SRI at 60 and 90 DAT and at harvest stage were statistically at par. Various types of reports are found about the effect of methods of planting on rice plant growth. Nissanka and Bandara (2004) found no variation in plant height between the SRI and conventional transplanting. However, Haque (2002) found the highest plant height from wider spacing as done in SRI. Shirame *et al.* (2000) reported that the number of functional leaves, leaf area and total number of tillers hill⁻¹ were higher at wider spacing which increased the photosynthetic rate leading to taller plants. Shortening of plants in tiller crop at harvest might be due to shortage of time for proper vegetative growth and development. Similar results were also reported by many workers (Rahman, 2001; Khisha, 2002; Sarker *et al.*, 2002). However, Akita and Tanaka (1992) reported that at maturity the tallest plants were found at low plant density. Sharma and Ghosh (1998) and Richharia *et al.* (1964) observed higher plant height in tiller plant than those in seed plants.

Table 4.7 Economics of rice cultivation as affected by methods of rice cultivation and cyanobacterial inoculants

Treatment	Cost of cultivation (₹ ha ⁻¹)	Gross return (₹ ha ⁻¹)	Net return (₹ ha ⁻¹)	B:C ratio (₹)
Conventional method				
N ₀	40685	84660	43985	1.08
Recommended dose of N (120 kg ha ⁻¹)	42065	109207	67142	1.60
Compost application (1/3 N) + 2/3 N	49605	110560	60955	1.23
Biofilm based BGA biofertilizer + 2/3 N	41685	103377	61692	1.48
BGA + PGPR + 2/3 N	41685	109810	68125	1.63
Compost based BGA mixture inoculant + 2/3 N	41685	101550	59865	1.44
<i>Multani mitti</i> based BGA inoculant + 2/3 N	41685	106450	64765	1.55
SRI				
N ₀	38084	76920	38836	1.02
Recommended dose of N (120 kg ha ⁻¹)	39646	114020	74556	1.89
Compost application (1/3 N) + 2/3 N	47004	102080	36556	1.35
Biofilm based BGA biofertilizer + 2/3 N	39084	102170	63084	1.61
BGA + PGPR + 2/3 N	39084	113680	74596	1.91
Compost based BGA mixture inoculant + 2/3 N	39084	101860	71286	1.82
<i>Multani mitti</i> based BGA inoculant + 2/3 N	39084	101370	62286	1.59

Tiller is the important factor which decides the numbers of panicle m⁻² and ultimately yield of rice. The difference in number of tillers in CT was significant over SRI at 30 DAT. However, this superiority of CT over SRI in tiller count was not recorded at 60, 90 DAT and harvest. Numbers of tillers in both the methods were statistically at par in all the observations at 60, 90 DAT and harvest. It was recorded that the tillers per hill were higher in SRI compared to CT. This might be due to higher spacing in SRI (25 x 25 cm) which makes more space available for growth of individual hill compared to CT. However, per m⁻² basis, both methods recorded same number of tillers. The practice of transplanting one young seedling hill⁻¹ with wider spacing (SRI) had advantage in reducing transplanting injury and increasing tiller (Horie, 2004) and minimizes the competition between plants (Rabenandrasana, 1999). Earlier

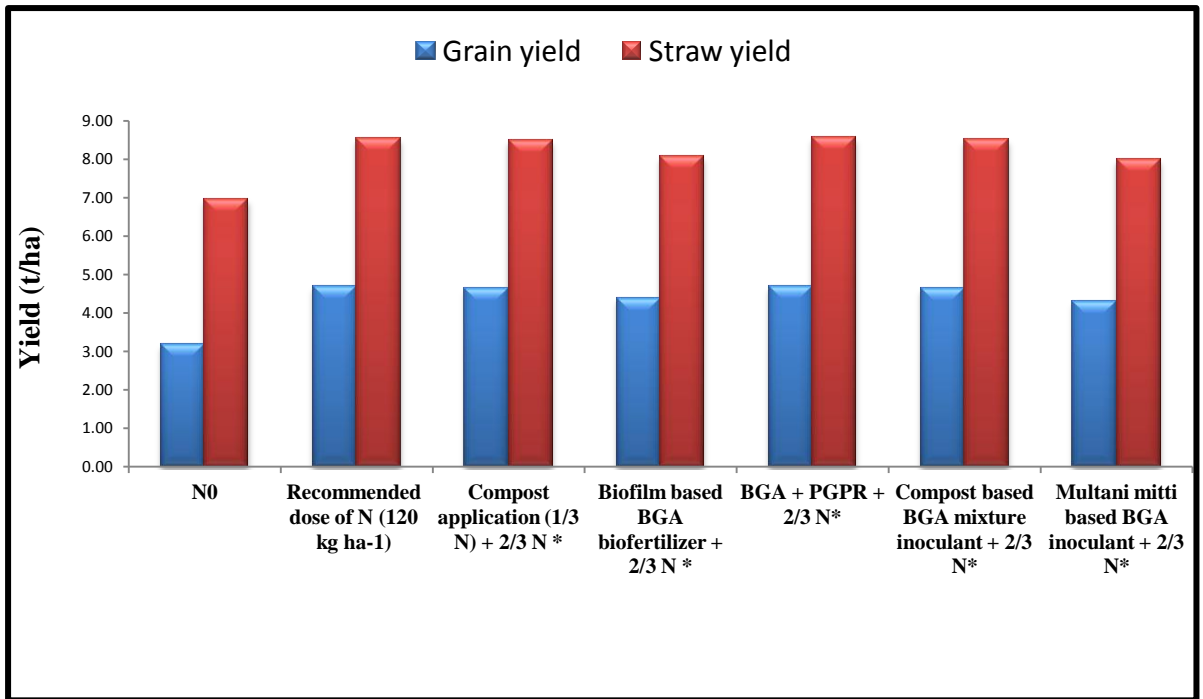


Fig. 4.5a: Grain and straw yield of rice as influenced by different cyanobacterial inoculants

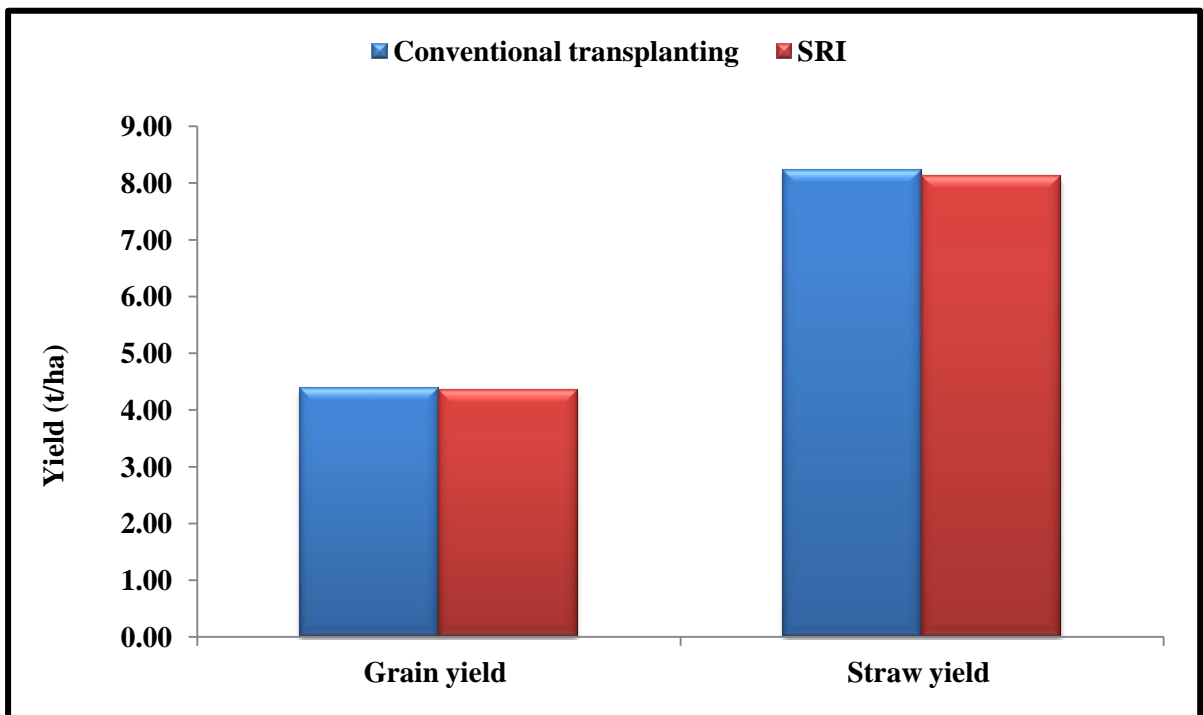


Fig. 4.5b: Grain and straw yield of rice as influenced by methods of planting

transplanting induced the transplanting shock at a more convenient point in the growth cycle when they could rebound faster and had little effect on tillers (Uphoff, 2002). Alternate wetting and drying maintaining a thin film of water. This might be responsible for opening the soil for both oxygen and nitrogen and promoted the root growth during initial growth stages which ultimately increased tiller density (Uphoff, 2001). Nissanka and Bandara (2004) observed that, the tiller number plant⁻¹ were higher in the SRI compared to conventional transplanting, conventional broadcasting and high density broadcasting, but this parameter, when expressed per unit area basis were not significantly different. However, Karmakar *et al.* (2004) and Lu *et al.* (2004) found number of tillers per unit area were higher in closer spacing that contributed to obtain higher yield.

Significantly higher dry matter accumulation was recorded in conventional transplanted rice as compared to SRI method at 30 DAT but at 60 and 90 DAT difference in dry matter accumulation was non-significant. Sharma and Ghosh (1998) and Sharma (1992) also observed highest dry weight in clonally propagated crop compared to nursery seedlings. Lu *et al.* (2004) reported that dense spacing increased the dry matter production of rice. Leaf area index (LAI) indicates the photosynthetic capacity of plant. LAI was found significantly higher in CT over SRI at 30 DAT. However, it was not significantly different between both the methods at 60 and 90 DAT. LAI was found increased up to 60 DAT and declined thereafter. Kim *et al.* (1999) and Ray *et al.* (2000) reported higher LAI in closer spacing, but Vijayakumar *et al.* (2006) observed maximum LAI in wider spacing.

Study of yield attributes is a prerequisite for predicting yielding ability of crop. Conventional and SRI method of planting did not show any significant effect on different yield attributes viz., panicle number m⁻², weight and length of panicles, number of grain panicle⁻¹ and test (1000- grain) weight of rice grain. However, Husain *et al.* (2004) observed higher number of effective tiller and number of grains per panicle under SRI compared to farmers' practice. Husain *et al.* (2003a) found more effective tillers and higher panicle length in the SRI than the conventional transplanting method. Sengthong (2002) noticed highest number of effective tillers when transplanted at spacing of 40 cm × 40 cm and Wang *et al.* (2002) reported higher number of effective tillers in the younger seedlings. Gasparillo *et al.* (2001) noted significant variation in panicle length between the SRI and non-SRI methods. Goel and Verma (2000) found higher panicle length from the transplanting method than the direct sowing method. Paul (1999) and Rahman (2001) found that nursery seedlings gave the longest panicles compared to the clonally propagated tillers. Ang *et al.* (2002) obtained maximum filled grains panicle⁻¹ from 40 cm × 40 cm spacing of the SRI. Like this experiment, Bari (2004) also observed that planting method had no significant influence on 1000-grains weight. But Husain *et al.* (2004) and Hossain *et al.* (2003) observed higher 1000-grains weight under SRI compared to the farmers' practice.

Grain and straw yield and harvest index in conventional and SRI method of planting were statistically at par. Nissanka and Bandara (2004) reported that grain yield was 7.6 t ha^{-1} in the SRI and it was 9%, 20% and 12% greater than the conventional transplanting, and normal and high density broadcasting and further suggested that the higher grain yield production in the SRI farming system might be attributed to the vigorous and healthy growth, development of more productive tillers and leaves ensuring greater resource utilization in the SRI compared to conventional transplanting and broadcasting systems. Husain *et al.* (2003a) found 39% higher straw yield in SRI compared to traditional methods. Suryavanshi *et al.* (2012) reported significantly higher grain yield in SRI compared to conventional transplanting. Stoop (2005) and Hossain *et al.* (2003) also found higher harvest index in SRI compared the conventional method, though, Barison (2003) found no difference for the same.

Free-living cyanobacteria are known to contribute an average of 20– 30 kg N ha^{-1} , whereas the value is up to 600 kg ha^{-1} for the *Azolla-Anabaena* system. Besides their N-enrichment potential, they are known to increase the water holding capacity, porosity and cation exchange capacity of soil. Problems associated with the establishment of inoculated strains (mainly due to competition with native strains in soil) and inconsistency in crop response as a result of environmental fluctuations has slowed down their adoption by farmers all over the world (Lee and Watanabe 1977; Whitton, 2000). A positive effect of bacterial inoculations on roots of maize and wheat has been reported (Arsac *et al.*, 1990; Creus *et al.*, 1996; Jacoud *et al.*, 1999). Similar beneficial effects of bacterial inoculations were also observed on plant biomass of rice varieties. In *Basmati* 385, an increase of 76 to 78% in plant biomass over control was noted. In Super *Basmati* maximum increase (83%) in plant biomass was shown. Four plant growth promoting rhizobacteria belonging to genera *Enterobacter* and *Aeromonas* showed beneficial effects on rice seedlings. These isolates could be proved useful for developing a bio-fertilizer for rice. Lucy *et al.* (2004) compiled results of large number of experiments on PGPR and concluded that PGPR inoculation had shown inconsistent results and found 50 to 70% increases in yields. It was reported that inconsistency in results was due to the quality of experimental design and analysis of results (Lucy *et al.*, 2004). Sakthivel *et al.* (1986) reported 12-27% greater plant height in response to PGPR inoculation than the non-inoculated control in rice.

Mehnaz *et al.* (2000) inoculated PGPR in seedlings of rice varieties *Basmati* 385 and Super *Basmati* and found beneficial effects of PGPR on root area of both rice varieties. PGPR like *Aeromonas* R8 and *Enterobacter* S1 and *Azospirillum brasilense* Wb3 showed maximum increase in root area of *Basmati* 385 where almost a doubling in root area over uninoculated plants was observed. In Super *Basmati*, all *Enterobacter* and *Aeromonas* strains proved to be more effective in increasing root area compared to *Azospirillum brasilense* strain. Sathia and Ramesh (2009) also reported positive influence on growth parameters like plant height, tillers

and dry matter production of aerobic rice were positively influenced by different nitrogen management practices. The usefulness of increased N application on tiller production was also observed. The PGPR isolates significantly increased the shoot length of rice seedlings (Ashrafuzzaman *et al.*, 2009). Chi *et al.* (1998) concluded from field trials that *Azospirillum* inoculated rice seedlings were taller and more vigorous than the non-inoculated controls.

The inoculated seedlings also showed a high survival rate in comparison to the non-inoculated ones. Elbadry *et al.* (1999) used phototrophic purple non-sulphur bacteria (PPNSB) as well as *Azospirillum*, *Azotobacter*, *Clostridium* and cyanobacteria for the inoculation of rice and their results established a positive response to rice to PPNSB, *Clostridium*, *Azotobacter* and *Azospirillum*, clearly reflecting the potential of PGPR to promote the growth and yield of rice. Hence, it can be concluded that many PGPR have a positive impact on growth of rice. The usefulness of increased N application on tiller production was also observed by Singh *et al.* (2006). Imtiyaj *et al.* (2011) found increased plant height, tillers and dry matter accumulation in rice due to the application of bio-fertilizers like BGA and *Azolla*. A PGPR *Pseudomonas fluorescens* B16 isolated from the roots of graminaceous plants has been shown to colonize the roots of various plants, and to increase the height, flower number, fruit number and total fruit weight of tomato plants (Minorsky, 2008). Prayitno *et al.* (1999) investigated the interaction between two groups of rice endophytic bacterial strains and several rice cultivars. Inoculation experiments showed that these rice-associating bacteria could promote, inhibit or have no influence on plant growth. Furthermore, the growth effects were greatly influenced by the environmental growth conditions. They suggested that some of these rice-associating bacteria possess important genes that influence their ability to intimately colonize on and/or within rice tissues promoting plant growth of rice. Baghel (2011) reported increased plant height, tillers, dry matter accumulation and LAI of rice due to the application of nitrogen by different sources.

The increasing LAI increases dry matter production, but net canopy photosynthesis could not increase indefinitely because of increased mutual shading of leaves. Overall, genetic, plant densities, spacing and fertilization are the major factors influencing the leaf area of plants grown under field conditions (Fageria *et al.*, 2006). Van *et al.* (2000) have reported an increase in 30% leaf area of rice due to inoculation of PGPR in pot and field experiments over no inoculation. A significant increase in shoot dry matter of rice seedlings was observed in response to PGPR isolates (Ashrafuzzaman *et al.*, 2009).

The treatment BGA+PGPR and 2/3 nitrogen through fertilizer had highest values in all the yield attributing characters. But this treatment was statistically at par over with three treatment including RDN, compost application (1/3 N) + 2/3 N through fertilizer and Compost based BGA mixture inoculant + 2/3 N through fertilizer. All these four treatment were statistically superior over treatment having Biofilm based BGA biofertilizer + 2/3 N

through fertilizer and *Multani mitti* based BGA inoculant + 2/3 N through fertilizer in respect to number, weight and length of panicles and number of grain panicle⁻¹. Test weight difference was at par in all the treatments including treatment having no nitrogen application. Interaction between planting method and cyanobacterial inoculants on the attributes like number, weight and length of panicles and number of grain panicle⁻¹ were found significant. Chaudhary (2008) found positive and significant impact of PGPR on number of panicles hill⁻¹ and filled grains hill⁻¹, the major yield attributes of rice. Biswas *et al.* (2000a) reported that yield increase of PGPR-inoculated rice was obtained due to significant increase in number of panicles and filled grains panicle⁻¹, and also the total number of spikelet's plant⁻¹ as compared to un-inoculated plants. Similarly, Elbadry *et al.* (1999b) reported that inoculation with PGPR significantly increased plant dry weight and number of productive tillers when compared to the non-inoculated control. Hence, use of PGPR help in improving the major yield attributes of rice. The N fertilization increased 1,000-grain weight of rice over control due to application of N through PU or ZEU (Meena *et al.*, 2002; Naik and Das, 2007). Sathiyia and Ramesh (2009) reported that various nitrogen management practices showed significant difference on yield attributes of aerobic rice. Imtiyaj *et al.* (2011) reported increased yield attributes like effective tillers, panicle length and no. of grains per panicle in rice due to the application of bio-fertilizers like BGA and *Azolla*.

Grain and straw yields were significantly higher with cyanobacterial inoculants and RDN over N control. Treatment containing BGA+PGPR and 2/3 nitrogen through chemical fertilizer showed highest grain and straw yield which was 4.69 t ha⁻¹ and 8.56 t ha⁻¹, respectively. But effect of this treatment was statistically at par with other three treatment including recommended dose of N (120 kg ha⁻¹), Compost application (1/3 N) + 2/3 N through fertilizer and Compost based BGA mixture inoculant + 2/3 N through fertilizer. All these four treatment were significantly better than the treatments containing Biofilm based BGA biofertilizer + 2/3 N through fertilizer and *Multani mitti* based BGA inoculant + 2/3 N through fertilizer. N control had the lowest grain (3.18 t ha⁻¹) and straw (8.21 t ha⁻¹) yields. Interaction between planting method and cyanobacterial inoculants on grain and straw yield was found significant.

Enhancement of rice seed germination, root and shoot growth, weight of rice grains and their protein content and the fertilizing action of N₂-fixing cyanobacteria has been generally attributed to the release of synthesized nitrogenous compounds either by decomposition of the cells or excretion (Nayak *et al.*, 2004). Wetland rice field can provides an ideal condition for the growth of cyanobacteria, which accumulate 19-38 kg N ha⁻¹ per crop and reduce the use of urea fertilizer in rice culture by 25-33 % (Hashem, 2001). Mule *et al.* (1999) reported positive effect of cyanobacterial inoculation on rice seedling, dry weight and shoot length as compared to control. They also reported that post-harvest soil showed best

quality when inoculated with *Telipothrix tenuis* or *T. tenuisturea* because, a) soil inoculated with these treatments recorded the highest increase in oxidisable carbon and soluble carbon compared with control and enhanced phosphorus content of soil compared with application of urea alone. b) The percentage of aggregate having >50 micron was highest with the addition of this cyanobacterium alone.

The use of bio-fertilizer and bioenhancer such as N₂ fixing bacteria and beneficial micro-organism can reduce chemical fertilizer applications and consequently lower production cost. Utilization of PGPR in order to increase the productivity may be a viable alternative to organic fertilizers which also helps in reducing the pollution and preserving the environment in the spirit of an ecological agriculture (Stefan *et al.*, 2008). Mehnaz *et al.* (2000) also found the usefulness of PGPR in increasing rice yield and those isolates could be proved useful for developing a bio-fertilizer for rice. Chaudhary (2008) did not find significant increase in grain yield over control due to the sole application of PGPR like *Bacillus subtilis* inoculation, but inoculation with *Azospirillum brasilense* increased the grain yield of rice significantly over both control (non-inoculated) and *Bacillus subtilis* inoculation. He concluded that *Azospirillum* was more suitable to rice. However, Imtiyaj *et al.* (2011) found increased grain and straw yield of rice due to the application of bio-fertilizers like BGA and *Azolla*. Das and Saha (2003) reported that inoculation of *Azotobacter* and *Azospirillum* either alone or in combination, not only increased the populations of both the diazotrophs and their activity in rice rhizosphere but also highly stimulated the availability of inorganic and organic fractions of nitrogen in rhizosphere soils of rice, which resulted in greater yield of the crop.

Lucy *et al.* (2004) reviewed reports on effects of PGPR from large number of publications around the world and concluded that results from different experiment were not harmonized and were often inconsistent and up to 50-70 % yield increases were reported. Omar *et al.* (1989) reported 15-20% increase in rice yield due to the application of PGPR while Kloepper *et al.* (1989) found 4.9 to 15.5% increase in yield. In the same report he reported 3 to 160% increased rice yield due to the application of *Pseudomonas sp.* in field experiment. Alam *et al.* (2001) also reported increase in rice dry matter and yield and N accumulation by 6 to 24% and concluded that yield increase was due to increased root length, leaf area and chlorophyll content. Tran Van *et al.* (2000) observed the rice grain yield by 13 to 22% due to application of PGPR. Several studies (Garcia and Dobereiner, 1996; Smith *et al.*, 1984) showed that an inappropriate combination of bacteria and crop plant often resulted in a negative effect on the nitrogen accumulation and growth of the host plant. Also, a number of experiments showed that the extent of the positive effect of the bacteria on nitrogen accumulation and crop growth varied with the species or variety of the host plant (Bouton and Brooks, 1982). Hence, bacterial inoculation may not always result in persistent response,

because of varying ecological factors and environmental conditions (Lynch, 1990). Results of a study on *Azospirillum brasilense* inoculation in rice showed that grain yield and 1000-grain weight increased over control (Omar *et al.*, 1989).

The highest straw yield of rice due to inoculation of PGPR, BGA and compost could be attributed to the higher supply of N and other micronutrients into soil (Bisht *et al.*, 2006; Khan *et al.*, 2008). The increased availability of Fe and other micronutrients in soil with inoculation of PGPR, BGA and compost in rice was responsible for higher yields as compared to control plots (Nayyar and Chhibba, 2000). When compost was added to the clay soil along with SRI practices the *Azospirillum* count rose to $1400 \times 10^3 \text{ Mg}^{-1}$ and yield to 10.5 t ha^{-1} (Randriamiharisoa, 2001). Hence, the variation in partitioning of photosynthates in grain and vegetative organs of different treatments possibly caused a significant variation in HI.

Cost of cultivation was found lower in all treatment of SRI method as compared conventional method. This might be due to low cost on nursery, seed and irrigation. Cost of cultivation was higher in the treatments having compost application compared to other treatments. Net return and B : C ratio in all the treatments were higher under SRI method compared to conventional method. Treatment having BGA + PGPR + 2/3 N (through fertilizer) under SRI method gave the highest net return (₹ 74, 596) followed by the recommended dose of N (RDN) through fertilizer (₹ 74556) + 2/3 N through chemical fertilizer. B : C ratio was highest in the same treatments having ₹ 1.91 and ₹ 1.89.

4.5 Conclusion

The growth and yield of rice was same in conventional and SRI planting method. However, net return and B:C ratio was higher in SRI method. Cyanobacterial inoculant can be used to replace approximately one-third of recommended dose of N fertilizer. Combination of plant growth promoting rhizobacteria (PGPR) along with BGA and 2/3 dose of chemical N fertilizer gave highest crop growth and yield so this treatment can be used in integrated nutrient management of rice.

Effect of cyanobacterial inoculants and planting methods on nutrient uptake and soil chemical parameters

Abstract

A field experiment was undertaken during the rainy (*Khariif*) season of 2011 to investigate the influence of methods of crop establishment and cyanobacterial inoculants on soil chemical properties and nutrient uptake. The experiment was laid out in split plot design with fourteen treatments combinations, comprising two methods of crop establishment viz., conventional transplanting (CT) and system of rice intensification (SRI) and seven cyanobacterial inoculants viz., N₀; Recommended dose of N (120 kg ha⁻¹); Compost application (1/3 N) + 2/3 N; Biofilm based BGA bio-fertilizer + 2/3 N; BGA + PGPR + 2/3 N; Compost based BGA mixture inoculant + 2/3 N and *Multani mitti* based BGA inoculant + 2/3 N. 2/3 N (80 kg ha⁻¹) and recommended dose of N (120 kg ha⁻¹) was applied through urea and applied in three splits. *Basmati* rice variety 'Pusa *Basmati* 1401' was taken in the experiment and these treatments were replicated three times. In CT and SRI, 21 and 12 days old seedlings, respectively were transplanted in puddled fields. Results indicated that conventional and SRI method of planting showed statistically at par concentrations and uptake of N, Fe, Zn, Mn and Cu in grain and straw. Protein content in grains was also same in both the methods (7.7 %). Treatment having cyanobacterial inoculants + PGPR + 2/3 N as fertilizer showed highest concentration and uptake of N, Fe, Zn, Mn and Cu in grain and straw; While treatment containing no nitrogen showed lowest values for concentration and uptake of these nutrient. Four treatments including BGA+PGPR+2/3 N through fertilizer, RDN, compost application (1/3 N) + 2/3 N through fertilizer and compost based BGA mixture inoculant + 2/3 N through fertilizer had significantly higher uptake of N, Fe, Zn, Mn and Cu in grain and straw than two treatments including Biofilm based BGA biofertilizer + 2/3 N through fertilizer and *Multani mitti* based BGA inoculant + 2/3 N through fertilizer. All cyanobacteria inoculated treatment and RDN had at par concentration of N, Fe, Mn and Cu in grain and straw. Status of available nitrogen, phosphorus, potassium and iron at crop harvest was same in both the methods of rice cultivation. Available nitrogen content of soil at crop harvest stage showed that compost application (1/3 N) + 2/3 N through chemical fertilizer recorded the highest available nitrogen content followed by compost based BGA mixture inoculant + 2/3 N through fertilizer and RDN. However available P, K and Fe in soil at crop harvest were statistically at par in all the treatments.

Keywords: Micronutrient uptake, N uptake, soil chemical properties, soil nutrient status, system of rice intensification (SRI).

5.1 Introduction

Rice is a staple food in south east and east Asia where about 90% of world's rice is grown and consumed (Prasad, 2009). Rice is the one of the most prominent food crop of India cultivated over 41.8 million ha area with 95.3 million tonnes production and represent staple food of India. By the year 2025, it is estimated that it will be necessary to produce about 60% more rice than what is currently produced to meet the food needs of a growing world population. In addition, the land available for crop production is decreasing steadily due to urban growth and land degradation. Hence, increases in rice production will have to come from the same or an even less amount of land. This means appropriate rice production practices should be adopted to improve rice yield per unit area. Among the production practices, water and nitrogen (N) have especial importance in increasing rice yield. Ladha and Reddy (2003) compared the rice grain yields and plant N requirements as they have increased through the years. Grain yields before the first green revolution were around 3 t ha⁻¹, with rice requiring 60 kg N ha⁻¹. During the first green revolution, grain yields reached 8 t ha⁻¹, with rice requiring 160 kg N ha⁻¹. The second green revolution is expected to produce grain yields of 12 t ha⁻¹ and require 240 kg N ha⁻¹ (Samonte *et al.*, 2006).

The significant role of human activities, particularly increasing cost of fossil fuels and the changes in land use and management have led to increased atmospheric carbon dioxide (CO₂) concentrations. One of the options currently being explored is the use of cyanobacteria as CO₂ sinks. Cyanobacteria are attractive candidates in this respect as they are fast growing and easier to manipulate in open fields. They possess an essential bio-physical mechanism (carbon concentrating mechanism, CCM), which concentrates CO₂ at the site of photosynthetic carboxylation (Kaplan *et al.*, 1994). However, nitrogen limitation is a key factor and in this context, “green options” such as biological nitrogen fixation through microbial inoculants, including cyanobacteria are promising resources.

The system of rice intensification (SRI) technique has stimulated a substantial amount of debate about the principles of irrigated rice cropping. However, little research following common scientific standards has been conducted to allow a thorough evaluation of SRI, especially in relation to cyanobacterial inoculation and its effects on nutrient uptake and soil nutrient status. Farmers in a number of countries have been able to increase the yields from their current rice varieties with available resources by utilizing SRI (Kabir and Uphoff, 2007). Higher productivity in SRI was achieved by making certain changes in the management of rice plants and the resources upon which these depend i.e., soil nutrients, air, water, soil biota, and solar energy (Thakur *et al.*, 2010).

The paddy field ecosystem represents a unique aquatic-terrestrial habitat, which provides a favorable environment for the growth and nitrogen fixation by cyanobacteria meeting their requirements for light, water, elevated temperature and nutrient availability

(Prasanna *et al.*, 2009). Enhancement of rice seed germination, root and shoot growth, weight of rice grains and their protein content and the fertilizing action of N₂-fixing cyanobacteria has been generally attributed to the release of synthesized nitrogenous compounds either by decomposition of the cells or excretion (Nayak *et al.*, 2004). Wetland rice field can provide an ideal condition for the growth of cyanobacteria, which accumulate 19-38 kg N ha⁻¹ crop⁻¹ and reduce the use of urea fertilizer in rice culture by 25-33 % (Hashem, 2001).

Along with cyanobacteria there are certain rhizobacteria that exert beneficial effect on plant growth and development are referred as plant growth promoting rhizobacteria (PGPR). PGPR affect the plant growth by, a) production and release of certain secondary metabolites (plant growth regulator/phytohormones/biologically active substances), b) preventing deleterious effect of phytopathogenic organism in the rhizosphere and, c) facilitating the availability and uptake of certain nutrients from the root environment (Zahir *et al.*, 2004). One of the most often reported PGPR *Penibacillus polymyxa* has a range of reported properties including nitrogen fixation, phosphorus solubilization, production of antibiotic and cytokinin and increased root and shoot growth of crops (Timmusk *et al.*, 1999). The biofilms based on cyanobacteria could be beneficial for rice cultivation. The beneficial effects of PGPR have been well demonstrated in both greenhouse and field conditions (Yanni *et al.*, 1997; Biswas *et al.*, 2000a, 2000b). With this background, a field experiment was conducted to evaluate the influence of cyanobacteria based inoculants and cultivation methods on plant nutrient uptake and soil nutrient status.

5.2 Materials and methods

The experiment was conducted during rainy (*kharif*) season (June- October) of 2011 at the Research Farm of Indian Agricultural Research Institute, New Delhi, situated at a latitude of 28°40' N and longitude of 77°12' E, altitude of 228.6 meters above the mean sea level (Arabian Sea). The mean annual rainfall of Delhi is 650 mm and more than 80% generally occurs during the south-west monsoon season (July-September) with mean annual evaporation 850 mm. During rice cropping season (June to October) maximum and minimum temperatures ranged between 43.6°C (8 June) to 29.4°C (30 October) and 11.9°C (28 October) to 29°C (7 June), respectively. The year 2011, had less than normal rainfall (539.4 mm during June to October) out of which 43.8, 226.8 and 163.6 mm rainfall which comprised 80.5 % of the total rainfall was received during July, August and September months, respectively. There were 6, 9 and 11 numbers of rainy days during July, August and September months, respectively. There was no rainfall in October. There was late onset of monsoon and July month had much less rainfall than normal. Daily evaporation was also higher during rainy season. Difference in maximum and minimum daily relative humidity was higher in June and October months. In other weather parameters many fluctuations were not recorded over the normal conditions of Delhi. The soils of experimental field had 134.9 kg ha⁻¹ alkaline

permanganate oxidizable N (Subbiah and Asija, 1956), 15.90 kg ha⁻¹ available P (Olsen *et al.*, 2006), 260.83 kg ha⁻¹ 1 N ammonium acetate exchangeable K (Prasad *et al.*, 2006) and 0.53 % organic carbon (Walkley and Black, 1934). The pH of soil was 8.1 (1: 2.5 soil and water ratio).

The experiment was laid out in split plot design with fourteen treatments combinations comprising of two methods of crop establishment viz., conventional transplanting (CT) and system of rice intensification (SRI) and seven cyanobacterial inoculants viz., N₀ no nitrogen; Recommended dose of N (120 kg ha⁻¹); Compost application (1/3 N) + 2/3 N; Biofilm based BGA bio-fertilizer + 2/3 N; BGA + PGPR + 2/3 N; Compost based BGA mixture inoculant + 2/3 N and *Multani mitti* based BGA inoculant + 2/3 N. Basmati rice variety ‘Pusa Basmati 1401’ was taken in the experiment and these treatments were replicated three times. In CT and SRI 21 and 12 days old seedlings, respectively were transplanted in puddled fields.

The experimental field was disc-ploughed twice and leveled and after puddling twenty-one days old seedlings of rice varieties were transplanted under conventional transplanting at 20 cm × 10 cm spacing keeping 2 seedlings hill⁻¹. For SRI, 12 days old seedlings were transplanted in main field. In conventional transplanting method 3-5 cm cropping water was maintained from transplanting to grain filling stage of crop and later only moist soil conditions were maintained. However in SRI, alternate wetting and drying conditions were maintained throughout the cropping season. In the experiment equal dose of inorganic fertilizers 60 kg P₂O₅ and 60 kg K₂O ha⁻¹ was applied in all the treatments. 120 kg ha⁻¹ Nitrogen was applied in treatment containing recommended dose of N and 80 kg ha⁻¹ in other treatment except treatment containing no nitrogen (N₀). The gross plot size was 6.2 m × 2.46 m for each treatment. Rotary weeder was used in SRI for weed management and interculture while in CT manual hand weeding was done. Crop was grown as per recommended practices and was harvested in the last fortnight of October.

N concentration in grain and straw of rice samples were determined by modified Kjeldahl method; while available N in soil after rice harvest was measured by Alkaline permanganate oxidizable N (Subbiah and Asija., 1956). Available phosphorus (P) content of soil was measured by Olsen’s method (Jackson, 1973) and available K content of soil was measured by Flame photometer method (Jackson, 1973). Soil iron (Fe) content of soil was measured by DTPA extraction method (Lindsey and Norwell, 1973). The Zn, Mn, Cu and Fe in grain and straw of rice crop were determined by rainy-digestion (di-acid digestion) procedure (Prasad *et al.*, 2006). The uptake/accumulation of various major and micronutrients in grain of rice were calculated by multiplying the grain yield of rice with their respective concentrations and expressed in kg ha⁻¹ and g ha⁻¹ for macro and micro nutrient.

All the data obtained from the experiment conducted under split plot design were statistically analyzed using the *F*-test as per the procedure given by Gomez and Gomez (1984). LSD values at *P* = 0.05 were used to determine the significance of difference between treatment means. Interaction effect was also statistically analysed in the same design.

5.3 Results

5.3.1 Concentration and uptake of nutrients in grain and straw

5.3.1.1 Concentration and uptake of nitrogen and protein in grain

Concentrations and uptake of nitrogen in grain and straw of rice were not significantly influenced by the conventional and SRI method of planting (Table 5.1). Protein content was also same (7.7 %). In sub-plots treatments, N concentrations in grain and straw were statistically at par in all treatments having cyanobacteria inoculation and recommended dose of nitrogen (RDN). N concentrations in grain and straw ranged between 1.14% and 0.38 % in N control (no nitrogen) to 1.33 % and 0.56 % in treatment containing BGA + PGPR + 2/3 N through chemical fertilizer respectively. However, N concentrations in cyanobacteria inoculated treatment and RDN were significantly higher than N control (no nitrogen). Recommended dose of nitrogen recorded highest protein content followed by BGA+PGPR+2/3 nitrogen through chemical fertilizer. But all cyanobacteria inoculation treatments and RDN had at par protein content. But cyanobacteria inoculated treatments and RDN had significant higher protein than N control.

Uptake of nitrogen in grain and straw was non-significant in conventional and SRI method of planting. Treatment containing BGA+PGPR+2/3 N through fertilizer showed highest uptake (62.39 kg ha⁻¹) closely followed by RDN (62.36 kg ha⁻¹). But this treatment had statistically at par in uptake with compost application (1/3 N) + 2/3 N through fertilizer (61.70 kg ha⁻¹) and compost based BGA mixture inoculant + 2/3 N through fertilizer (61.37 kg ha⁻¹). All these four treatments significantly higher N uptake over Biofilm based BGA biofertilizer + 2/3 N through fertilizer and *Multani mitti* based BGA inoculant + 2/3 N through fertilizer. In straw, N uptake was highest in BGA+PGPR+2/3 N through fertilizer (47.43 kg ha⁻¹) followed by RDN (46.95 kg ha⁻¹), compost application (1/3 N) + 2/3 N through fertilizer (61.70 kg ha⁻¹) and compost based BGA mixture inoculant + 2/3 N through fertilizer (61.37 kg ha⁻¹). N control showed lowest total N uptake 62.8 kg ha⁻¹ while BGA+PGPR+2/3 N through fertilizer had highest N uptake (109.8 kg ha⁻¹).

5.3.1.2 Concentration and uptake of iron (Fe) in grain and straw

Concentration of Fe in grain and straw in conventional and SRI method was found to be statistically at par having the concentration of 78.20 mg kg⁻¹ and 77.98 mg kg⁻¹ in grain and 421.80 mg kg⁻¹ and 419.86 mg kg⁻¹ in straw under conventional and SRI method of planting, respectively (Table 5.2 Fig. 5.2). Same trend was observed in uptake of Fe in grain

Table 5.1 Effect of planting method and cyanobacterial inoculants on nitrogen content, protein content and N uptake in grain and straw of rice

Treatment	N conc. in grain (%)	Protein content (%)	N conc. in straw (%)	N uptake in grain (kg ha ⁻¹)	N uptake in straw (kg ha ⁻¹)	Total N uptake (kg ha ⁻¹)
Planting method						
Conventional transplanting	1.30	7.7	0.52	57.1	43.3	100.42
SRI	1.30	7.7	0.52	56.7	42.8	99.56
SEm±	0.004	0.023	0.004	0.101	0.47	0.427
LSD (P=0.05)	NS	NS	NS	NS	NS	NS
Cyanobacterial inoculants						
N ₀	1.14	6.8	0.38	36.3	26.5	62.8
Recommended dose of N	1.33	7.92	0.55	62.3	46.9	109.3
Compost application (1/3 N) + 2/3 N	1.33	7.90	0.55	61.7	46.0	107.7
Biofilm based BGA bioferti.+2/3 N	1.32	7.86	0.55	57.9	44.3	102.1
BGA + PGPR + 2/3 N	1.33	7.91	0.56	62.3	47.4	109.8
Compost based BGA mix.ino..+2/3 N	1.32	7.87	0.55	61.4	46.6	107.9
<i>Multani mitti</i> based BGA inocu. +2/3 N	1.32	7.85	0.54	56.6	43.6	100.2
SEm±	0.01	0.04	0.01	0.96	1.02	1.47
LSD (P=0.05)	0.02	0.11	0.03	2.80	2.98	4.28
Cyanobacterial inoculants x Method						
SEm±	0.009	0.5	0.01	1.35	1.44	2.07
LSD (P=0.05)	Sig.	Sig.	Sig.	Sig.	Sig.	Sig.

and straw under both the methods of planting. In sub-plot treatments, all cyanobacteria inoculated plots and RDN showed statistically on par in their Fe concentrations in grain and straw but these concentrations were statistically higher than N control. Highest Fe uptake was recorded with BGA+PGPR+2/3 N through fertilizer being 397.12 g ha⁻¹ in grain and 3827.81 g ha⁻¹ in straw which is statistically on par in uptake with RDN, compost application (1/3 N) + 2/3 N through fertilizer (61.70 kg ha⁻¹) and compost based BGA mixture inoculant + 2/3 N through fertilizer (61.37 kg ha⁻¹). All these four treatments significantly higher N uptake over Biofilm based BGA biofertilizer + 2/3 N through fertilizer and *Multani mitti* based BGA inoculant + 2/3 N through fertilizer.

5.3.1.3 Concentration and uptake of zinc (Zn) in grain and straw

Concentrations and uptake of Zn in grain and straw in conventional and SRI method of planting were statistically at par (Table 5.3 Fig. 5.3). Concentration of 21.66 mg kg⁻¹ and 21.37 mg kg⁻¹ in grain and 147.87 mg kg⁻¹ and 147.71 mg kg⁻¹ in straw for conventional and SRI method of planting, respectively were recorded. Highest Zn concentration in grain (22.78 mg kg⁻¹) and straw (152.85 mg kg⁻¹) were observed in treatment BGA + PGPR + 2/3 N through fertilizer and these were statistically at par with RDN but these treatments had statistically higher Zn concentration and uptake compared to N control. Highest Zn uptake was recorded with BGA+PGPR+2/3 N through fertilizer being 106.84 g ha⁻¹ in grain and

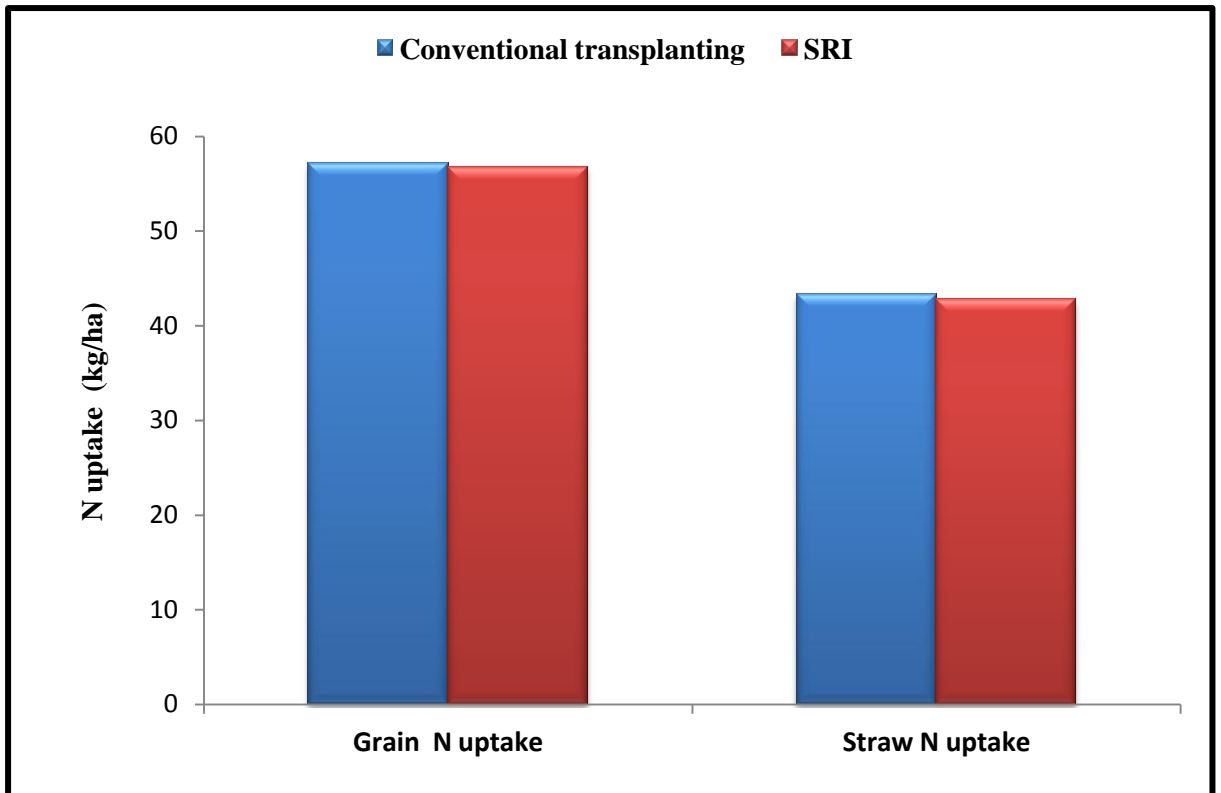


Fig. 5.1a: Nitrogen (N) uptake in grain and straw as affected by method of planting

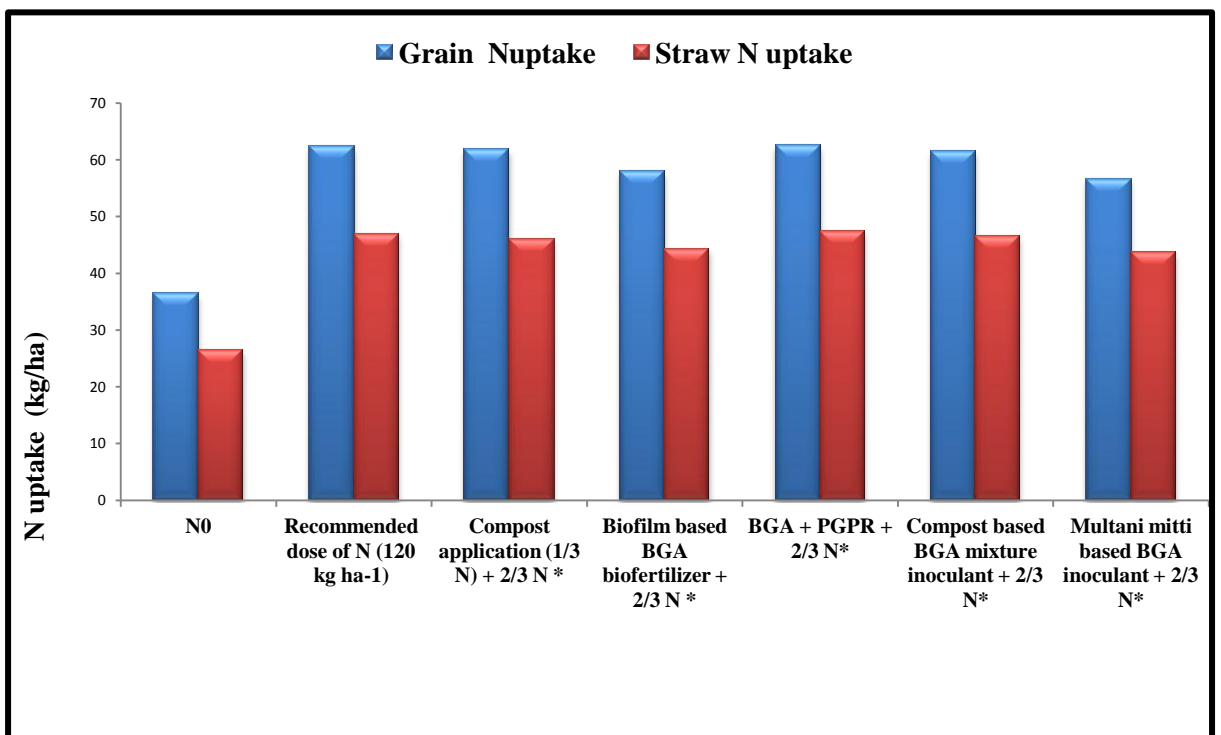


Fig. 5.1b: Nitrogen (N) uptake in grain and straw as affected by different cyanobacterial inoculants

Table 5.2 Effect of planting method and cyanobacterial inoculants on iron (Fe) content and uptake in grain and straw of rice

Treatment	Fe conc. in grain (mg kg ⁻¹)	Fe conc. in straw (mg kg ⁻¹)	Fe uptake in grain (g ha ⁻¹)	Fe uptake in straw (g ha ⁻¹)
Planting method				
Conventional transplanting	78.20	421.80	362.60	3492.33
SRI	77.98	419.86	359.88	3436.61
SEm±	0.215	0.848	1.808	31.527
LSD (P=0.05)	NS	NS	NS	NS
Cyanobacterial inoculants				
N ₀	73.34	282.57	238.48	1966.04
Recommended dose of N (120 kg ha ⁻¹)	78.77	443.00	392.3	3793.9
Compost application (1/3 N) + 2/3 N	79.19	444.26	389.67	3786.78
Biofilm based BGA biofertilizer + 2/3 N	78.50	442.93	357.65	3522.98
BGA + PGPR + 2/3 N	79.67	446.95	397.12	3827.81
Compost based BGA mix. inoculant + 2/3 N	78.75	443.86	387.89	3796.83
Multani mitti based BGA inoculant + 2/3 N	78.43	442.25	365.63	3556.98
SEm±	0.49	2.09	6.69	54.47
LSD (P=0.05)	1.42	6.09	19.52	158.99
Cyanobacterial inoculants x Method				
SEm±	0.68	2.95	9.45	77.03
LSD (P=0.05)	NS	Sig.	Sig.	Sig.

1308.98g ha⁻¹ in straw. Interaction between planting method and cyanobacterial inoculants on Zinc (Zn) content and uptake in grain and straw of rice was found significant.

5.3.1.4 Concentration and uptake of manganese (Mn) in grain and straw

Concentration and uptake of Mn in grain and straw in conventional and SRI method were non-significantly different (Table 5.4). Concentrations of 18.86 mg kg⁻¹ and 18.28 mg kg⁻¹ in grain and 57.44 mg kg⁻¹ and 57.13 mg kg⁻¹ in straw for conventional and SRI method, respectively were recorded. Highest Mn concentration grain (19.83 mg kg⁻¹) and straw (58.17 mg kg⁻¹) were observed in treatment BGA + PGPR + 2/3 N through fertilizer and these were statistically at par with RDN but these treatments had statistically higher Mn concentration and uptake compared to N control. Highest Mn uptake was recorded with BGA+PGPR+2/3 N through fertilizer both in grain (93.08 g ha⁻¹) in and in straw (498.07g ha⁻¹).

5.3.1.5 Concentration and uptake of copper (Cu) in grain and straw

Conventional and SRI method of planting showed statistically at par concentration of Cu in grain and straw (Table 5.5). Concentrations of 9.97 mg kg⁻¹ and 9.95 mg kg⁻¹ in grain and 35.7 mg kg⁻¹ and 35.67 mg kg⁻¹ in straw for conventional and SRI method, respectively

Table 5.3 Effect of planting method and cyanobacterial inoculants on Zinc (Zn) content and uptake in grain and straw of rice

Treatment	Zn conc. in grain (mg kg ⁻¹)	Zn conc. in straw (mg kg ⁻¹)	Zn uptake in grain (g ha ⁻¹)	Zn uptake in straw (g ha ⁻¹)
Planting method				
Conventional transplanting	21.66	147.87	94.9	1219.1
SRI	21.37	147.71	93.0	1202.9
SEm±	0.121	0.288	0.584	10.780
LSD (P=0.05)	NS	NS	NS	NS
Cyanobacterial inoculants				
N ₀	20.43	129.42	65.0	900.6
Recommended dose of N (120 kg ha ⁻¹)	21.38	150.88	99.9	1291.0
Compost application (1/3 N) + 2/3 N	21.87	150.97	101.4	1282.0
Biofilm based BGA biofertilizer + 2/3 N	21.41	150.03	93.7	1210.8
BGA + PGPR + 2/3 N	22.78	152.85	106.8	1309.0
Compost based BGA mix. inoculant + 2/3 N	21.33	150.52	99.1	1283.5
<i>Multani mitti</i> based BGA inoculant + 2/3 N	21.38	149.87	91.7	1200.1
SEm±	0.28	0.60	1.71	19.99
LSD (P=0.05)	0.82	1.75	5.00	58.35
Cyanobacterial inoculants x Method				
SEm±	0.39	0.84	2.42	28.27
LSD (P=0.05)	Sig.	Sig.	Sig.	Sig.

Table 5.4 Effect of planting method and cyanobacterial inoculants on manganese (Mn) content and uptake in grain and straw of rice

Treatment	Mn conc. in grain (mg kg ⁻¹)	Mn conc. in straw (mg kg ⁻¹)	Mn uptake in grain (g ha ⁻¹)	Mn uptake in straw (g ha ⁻¹)
Planting method				
Conventional transplanting	18.86	57.44	82.9	471.5
SRI	18.28	57.13	80.0	463.4
SEm±	0.178	0.225	1.056	4.283
LSD (P=0.05)	NS	NS	NS	NS
Cyanobacterial inoculants				
N ₀	16.17	55.63	51.5	387.2
Recommended dose of N (120 kg ha ⁻¹)	19.00	58.02	88.8	495.9
Compost application (1/3 N) + 2/3 N	18.72	57.28	86.9	485.0
Biofilm based BGA biofertilizer + 2/3 N	18.88	57.32	82.7	461.6
BGA + PGPR + 2/3 N	19.83	58.17	93.1	498.1
Compost based BGA mix. Inocu. + 2/3 N	18.68	57.68	86.8	490.0
<i>Multani mitti</i> based BGA inocu. + 2/3 N	18.72	56.88	80.3	454.6
SEm±	0.46	0.39	2.41	7.23
LSD (P=0.05)	1.33	1.14	7.03	21.09
Cyanoacterial inoculants x Method				
SEm±	0.64	0.55	3.40	10.21
LSD (P=0.05)	NS	Sig	Sig	Sig

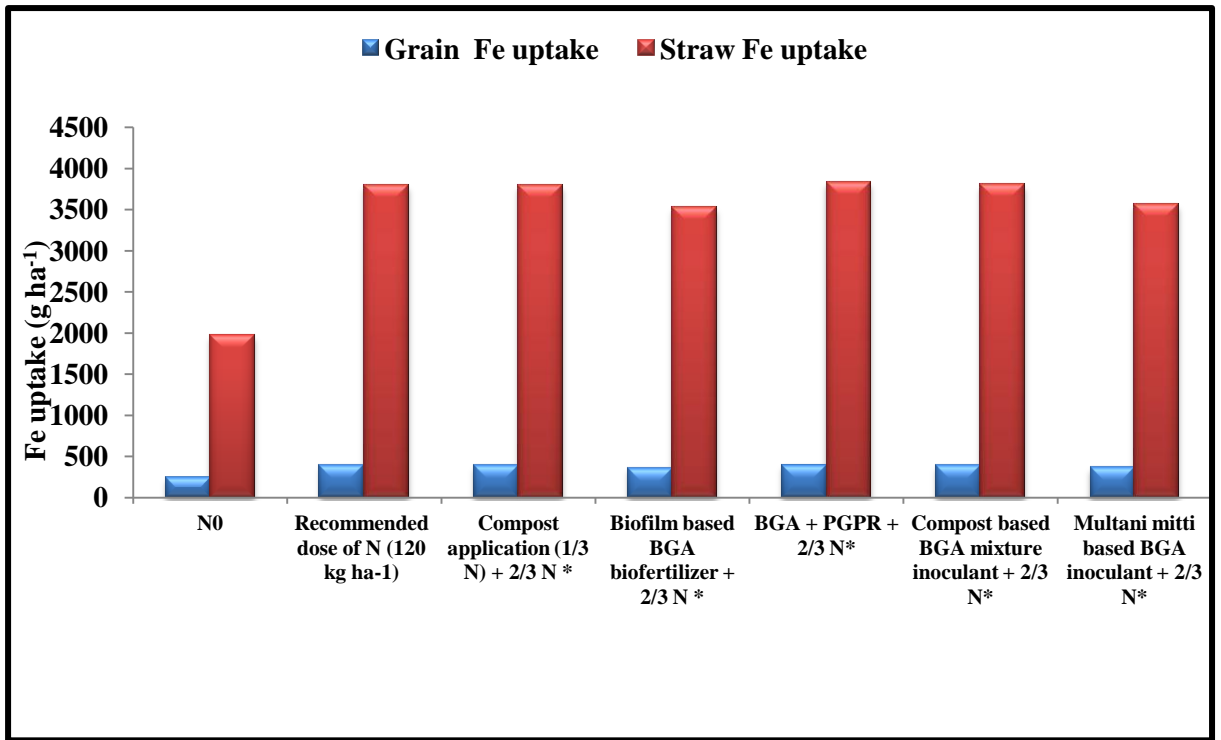


Fig. 5.2: Iron (Fe) uptake in grain and straw as affected by different cyanobacterial inoculants

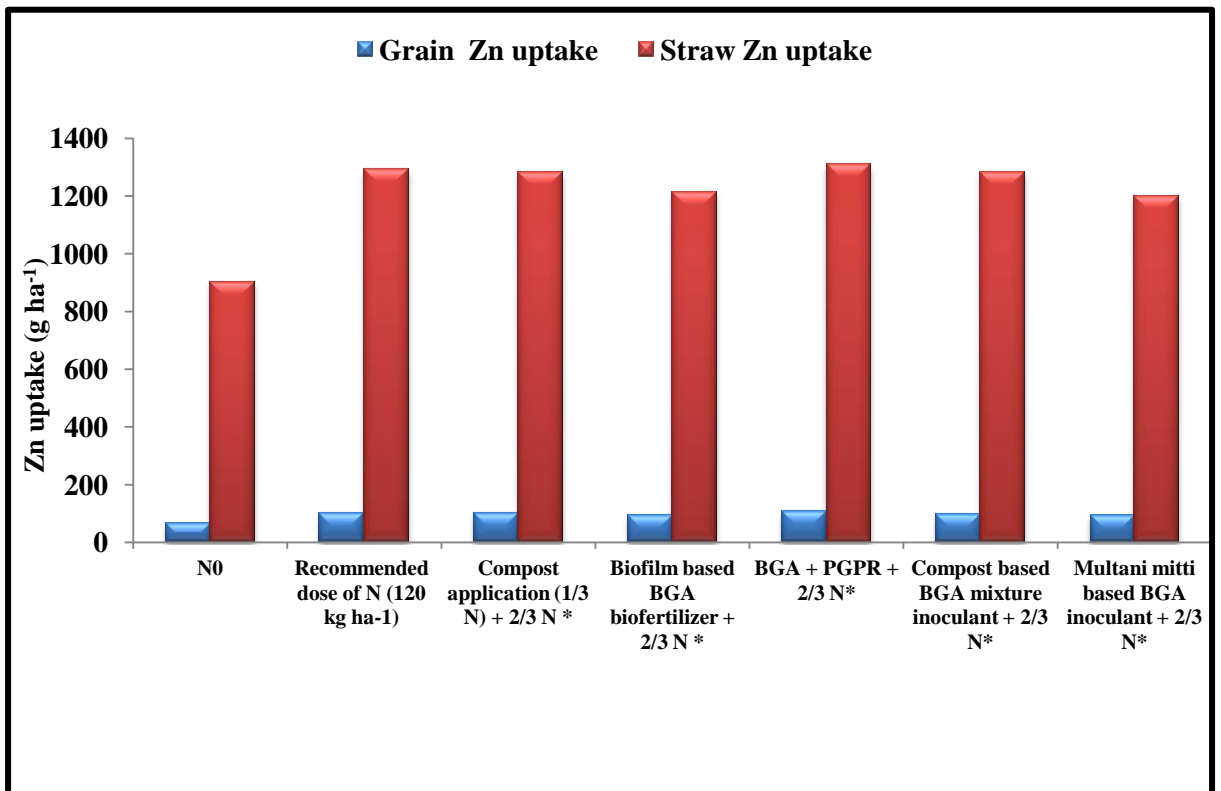


Fig. 5.3: Zinc (Zn) uptake in grain and straw as affected by different cyanobacterial inoculants

were recorded. Same trend was observed in uptake of Cu in grain and straw in both methods of planting. Lowest concentration in grain (9.07 mg kg⁻¹) and straw (34.50 mg kg⁻¹) was observed in N control (N₀). Highest concentration grain (10.29 mg kg⁻¹) and straw (36.32 mg kg⁻¹) was observed in treatment BGA + PGPR + 2/3 N through fertilizer and it was significantly higher than N control. Highest Cu uptake was recorded with BGA+ PGPR+ 2/3 N through fertilizer (48.23 g ha⁻¹) in grain and 310.79 g ha⁻¹ in straw.

Table 5.5 Effect of planting method and cyanobacterial inoculants on copper (Cu) content and uptake in grain and straw of rice

Treatment	Cu conc. in grain (mg kg ⁻¹)	Cu conc. in straw (mg kg ⁻¹)	Cu uptake in grain (g ha ⁻¹)	Cu uptake in straw (g ha ⁻¹)
Planting method				
Conventional transplanting	9.97	35.77	43.7	293.7
SRI	9.95	35.67	43.4	289.4
SEm±	0.009	0.024	0.103	2.291
LSD (P=0.05)	NS	NS	NS	NS
Cyanobacterial inoculants				
N ₀	9.07	34.50	28.9	240.1
Recommended dose of N (120 kg ha ⁻¹)	10.08	35.87	47.1	306.2
Compost application (1/3 N) + 2/3 N	10.22	36.05	47.4	305.3
Biofilm based BGA biofertilizer + 2/3 N	9.91	35.85	43.4	289.3
BGA + PGPR + 2/3 N	10.29	36.32	48.2	310.8
Compost based BGA mix. Inocu. + 2/3 N	10.24	35.65	47.3	303.2
<i>Multani mitti</i> based BGA inoculant + 2/3 N	9.90	35.82	42.4	286.1
SEm±	0.15	0.29	0.90	5.42
LSD (P=0.05)	0.43	0.85	2.62	15.81
Cyanobacterial inoculants x Method				
SEm±	0.20	0.41	1.27	7.66
LSD (P=0.05)	Sig	NS	Sig	Sig

5.3.2 Nutrient status of soil after crop harvest

Conventional and SRI method of planting showed statistically at par in available N, P, K and Fe in soil at crop harvest stage (Table 5.6). N control (N₀) showed lowest available N (85.7 kg ha⁻¹) and all other treatment having cyanobacterial inoculants and recommended dose of N had significantly higher available N than N control. Treatment having compost application (1/3 N) + 2/3 N through chemical fertilizer recorded the highest value of available N (158.62 kg ha⁻¹) followed by compost based BGA mixture + 2/3 N through fertilizer (147.65 kg ha⁻¹), recommended dose of fertilizer (144.5 kg ha⁻¹) and biofilm based BGA biofertilizer (142.9 kg ha⁻¹). Since phosphorous (P) and potassium (K) fertilizers were applied in equal quantity in all treatment including treatment containing no nitrogen, there was no significant difference in available P and K contents. Available iron content was also statistically at par in all the treatments.

Table 5.6 Effect of planting method and cyanobacterial inoculants on available N, P, K and Fe in soil at crop harvest

Treatment	Available N (kg ha ⁻¹)	Available P (kg ha ⁻¹)	Available K (kg ha ⁻¹)	Available Fe (kg ha ⁻¹)
Planting method				
Conventional transplanting	134.13	16.36	263.0	14.7
SRI	139.44	16.17	261.4	14.5
SEm±	2.561	0.161	0.375	0.069
LSD (P=0.05)	NS	NS	NS	NS
Cynobacterial inoculants				
N ₀	85.70	15.97	260.8	14.1
Recommended dose of N (120 kg ha ⁻¹)	144.51	16.05	262.4	14.4
Compost application (1/3 N) + 2/3 N	158.62	16.68	263.0	14.9
Biofilm based BGA biofertilizer + 2/3 N	142.94	16.03	261.6	14.5
BGA + PGPR + 2/3 N	136.67	16.93	263.5	14.8
Compost based BGA mixture inoculant + 2/3 N	147.65	16.13	262.6	14.6
Multani mitti based BGA inoculant + 2/3 N	141.38	16.05	261.9	14.8
SEm±	5.63	0.38	1.07	0.20
LSD (P=0.05)	16.42	NS	NS	NS
cyanobacterial inoculants x Method				
SEm±	7.95	0.54	1.50	0.28
LSD (P=0.05)	NS	Sig	Sig	NS

5.4 Discussion

Concentrations and uptake of nitrogen, iron, zinc, manganese and copper in grain and straw of rice were not significantly influenced by the conventional and SRI method of planting. Protein content was also same (7.7%) in both methods of planting. N concentrations in grain and straw were statistically at par in all treatments having cyanobacteria inoculation and recommended dose of nitrogen (RDN). However, N concentrations in cyanobacteria inoculated treatment and RDN were significantly higher than N control. PGPR are directly involved in increased concentration and uptake of nitrogen, synthesis of phytohormones, solubilization of minerals such as phosphorus, and production of siderophores that chelate iron and make it available to the plant root (Lalande *et al.*, 1989; Glick, 1995; Bowen and Rovira, 1999). It has also been reported that PGPR is able to solubilize inorganic and/or organic phosphates in soil (Liu *et al.*, 1992). Native and applied plant growth promoting rhizobacteria (PGPR) play an important role in better crop growth, yield, nutrient uptake and

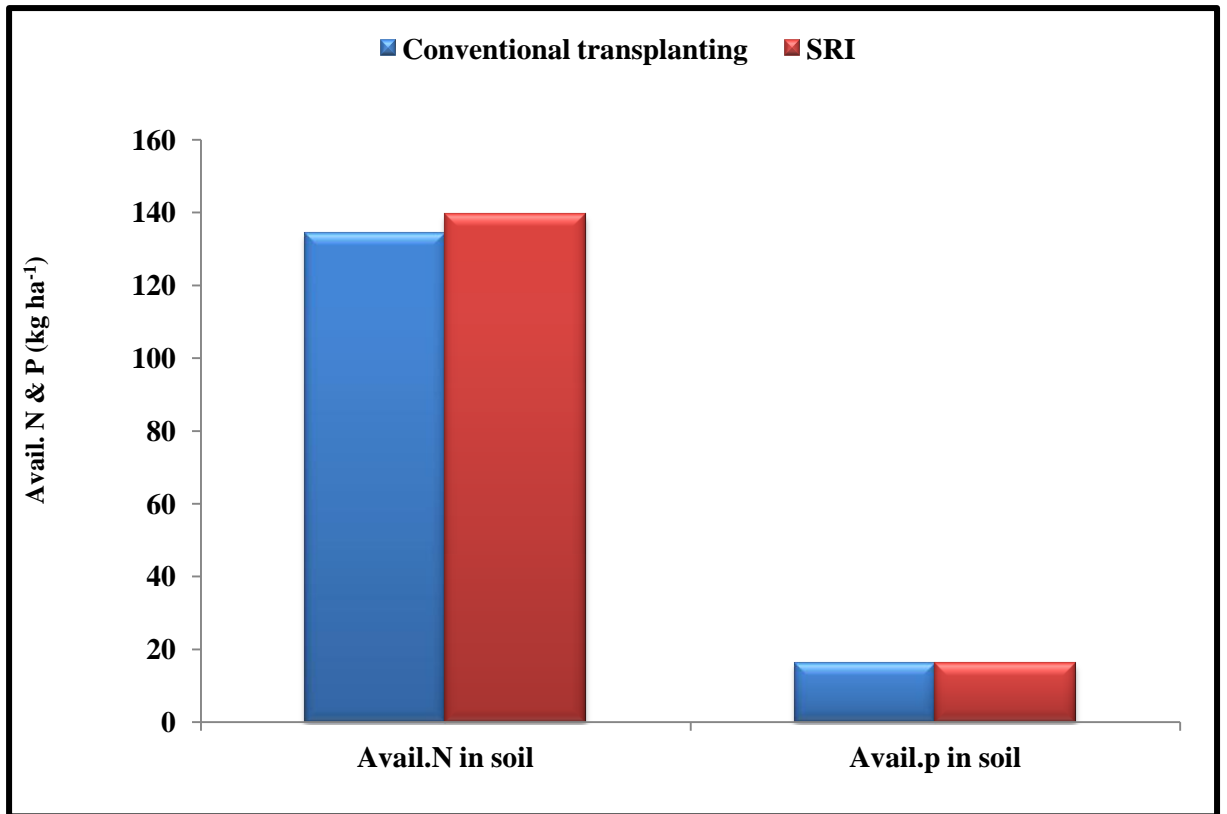


Fig. 5.4: Available N and P content of soil after crop harvest as affected by method of planting

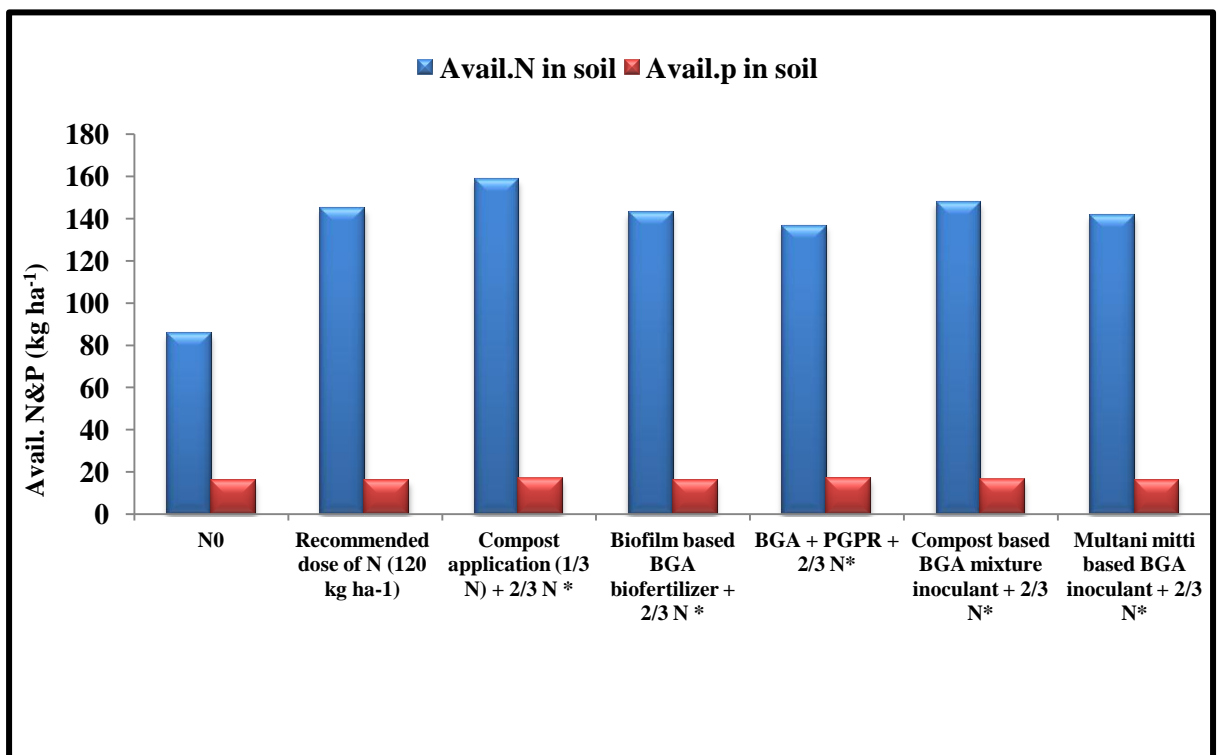


Fig. 5.5: Available N and P content of soil after crop harvest as affected by different cyanobacterial inoculants

soil chemical and biological parameters in crop however their responses varied from 50 to 70 % increase in yield (Lucy *et al.*, 2004). Choudhary (2008) found significant positive effect of PGPR on N, P and K concentrations in grain and straw of rice. The highest N, P and K concentration in rice grain was recorded with *Azospirillum* inoculation, being significantly higher than control and in *Bacillus* inoculation. Singh *et al.* (2011) also found significant positive effect of BGA application on N concentration and uptake in grain and straw of rice. The role of *Anabaena* in fixation of atmospheric N₂ is well established (Singh and Bisoyi, 1989; Nayak *et al.*, 2004) for rice crop. Therefore, increased availability of N in the rhizosphere resulted in its increased uptake due to *Anabaena* inoculation. Imtiyaj *et al.* (2011) reported significantly increased N, P and K concentration and uptake in grain and straw of rice due to the application of BGA, *Azolla* and other bio-fertilizers. Toro *et al.* (1997) reported that co-inoculation of *Glomus intraradices* and *Bacillus subtilis* significantly increased the vegetative biomass and N and P accumulation in plant tissues. Alam *et al.* (2001) reported that amount of total N increased by the inoculation of rhizobacteria in soil, and they have further suggested that free-living rhizobacteria may have some effects on N accumulation in the inoculated plots. Similarly, Lin *et al.* (1983) have also reported that plant nutrient uptake can be enhanced by the inoculation with free-living bacteria.

Continuous incorporation of summer green manuring over the years increased OC content of soil and which might also be responsible for higher nutrient availability (Swarup, 1987; Chatterjee *et al.*, 1979; Nayyar and Chhibba, 2000; Mandal *et al.*, 2003, and Dwivedi *et al.*, 2005; Bana, 2009). Naklang *et al.* (2006) reported that for production of same total above ground biomass N, P, and K uptake in a system with modern-type varieties, many more nutrients are accumulated in the rice straw. If only the grain yield is removed from the field, nutrient “mining” is relatively less. K concentration in rice grain was reported to increase with increasing levels of N and influenced favourably by sources of N (Duhan *et al.*, 2001; Shivay *et al.*, 2001) and bacterial and cyanobacterial PGPR and BGA also increased the availability of nutrients especially N in soil. Singh *et al.* (2011) reported significant effect of BGA application on K concentration and uptake in grain and straw of rice. Imtiyaj *et al.* (2011) also found significantly increased K concentration and uptake in grain and straw of rice due to the application of BGA, *Azolla* and other bio-fertilizers.

PGPR facilitates plant growth and development, both directly and indirectly (Glick, 1995). Direct stimulation may include providing plants with fixed nitrogen, phyto-hormones or iron (that has been sequestered by bacterial siderophores) and solubilized phosphate, while indirect stimulation of plant growth includes preventing phyto-pathogens (bio-control) through production of antibiotics/ siderophores and hydrogen cyanide and thus promoting plant growth and development (Kloepper *et al.*, 1993). Green manuring residue incorporation significantly influenced the micronutrients concentration in plant system (Chahal *et al.*, 1999)

and PGPR and BGA also worked similarly. Singh *et al.* (2011) also found significant positive effect of BGA application on Fe, Zn and Cu concentration and uptake in grain and straw of rice. Intiyaj *et al.* (2011) reported significantly increased Fe, Zn and Cu concentration and uptake in grain and straw of rice due to the application of BGA, *Azolla* and other bio-fertilizers. Under integrated nutrient management (INM) under the presence of combined nitrogen the BGA not only continue to grow but show concentration dependent increase in the uptake of added chemical nitrogen. This metabolized nitrogen which is prevented from being lost through denitrification, percolation and run off, is slowly released through excretion and autolysis (Suseela and Goyal, 1995; Singh and Mandal, 2000) and thus help in better crop growth, yield and higher nutrient uptake.

Conventional and SRI method of planting showed statistically at par available N, P, K and Fe in soil at crop harvest stage. N control (N_0) showed lowest available N (85.7 kg ha^{-1}) and all other treatment having cyanobacterial inoculants and recommended dose of N had significantly higher available N than N control. Suryavanshi (2011) also did not find any significant change in available N and available Fe under conventional and SRI planting. Treatment having compost application ($1/3 \text{ N}$) + $2/3 \text{ N}$ through chemical fertilizer recorded the highest value of available N ($158.62 \text{ kg ha}^{-1}$) followed by compost based BGA mixture + $2/3 \text{ N}$ through fertilizer ($147.65 \text{ kg ha}^{-1}$), recommended dose of fertilizer (144.5 kg ha^{-1}) and biofilm based BGA bio-fertilizer (142.9 kg ha^{-1}). Higher available N in soil at harvest stage due to compost application might be due to slow release of N from compost. Since phosphorous (P) and potassium (K) fertilizers were applied in equal quantity in all treatment including treatment containing no nitrogen, there was no significant difference in available P and K contents.

5.5 Conclusion

Conventional and SRI methods of rice cultivation did not influence the concentration and uptake of nutrients like N, Fe, Zn, Mn and Cu in grain and straw. BGA+ PGPR+ $2/3 \text{ N}$ through fertilizer showed highest concentrations and uptake of these nutrients in grain and straw though these were at par with recommended dose of N fertilizer. Available nitrogen content at crop harvest was higher when treatment included compost and it was at par with recommended dose of N fertilizer. However, available P, K and Fe in soil at harvest stage were same in all the treatments.

6. RESEARCH PAPER - III

Influence of cyanobacterial inoculants and planting methods of rice on soil microbial parameters, aggregation and carbon content

Abstract

A field experiment was conducted during the rainy (*khariif*) season of 2011 at the research farm of the IARI, New Delhi to study the influence of methods of crop establishment and cyanobacterial inoculants on soil microbial parameters, aggregation and carbon content in basmati rice (variety 'Pusa *Basmati* 1401'). The experiment was laid out in split plot design with two methods of crop establishment viz., conventional transplanting (CT) and system of rice intensification (SRI) as main plots and seven cyanobacterial inoculants viz., N₀; Recommended dose of N (120 kg ha⁻¹); Compost application (1/3 N) + 2/3 N; Biofilm based BGA bio-fertilizer + 2/3 N; BGA + PGPR + 2/3 N; Compost based BGA mixture inoculant + 2/3 N and *Multani mitti* based BGA inoculant + 2/3 N. 2/3 N (80 kg ha⁻¹) and recommended dose of N (120 kg ha⁻¹) was applied through urea in three splits in the experiment. These treatments were replicated three times. Results showed that all the microbial parameters including acetylene reductase activity (ARA), soil chlorophyll, dehydrogenase activity and microbial biomass carbon (MBC) content of soil were higher at 60 days after transplanting (DAT) compared to those at crop harvest. However, no significant difference was recorded in acetylene reductase activity (ARA), soil chlorophyll, dehydrogenase activity and MBC content under conventional and SRI methods either at 60 DAT or at crop maturity. Treatment containing BGA + PGPR + 2/3 N (fertilizer) recorded highest ARA, soil chlorophyll and dehydrogenase activity MBC at 60 DAT and at crop maturity. Application of compost (1/3 N) + 2/3 N through (fertilizer) showed higher carbon content in all the three fractions viz., very labile, labile and less labile. Treatment containing no nitrogen (N₀) recorded lowest C in all fractions at both 0-7.5 cm and 7.5-15 cm depths. Conventional and SRI methods did not show any significant difference labile and less labile carbon fraction at both depths. Highest mean weight diameter (MWD) indicating better soil aggregation in treatment having compost application (1/3 N) + 2/3 N (fertilizer), which was significantly higher than *Multani mitti* based BGA inoculant + 2/3 N (fertilizer) and biofilm based BGA bio-fertilizer 2/3 N (fertilizer) at 0-7.5 cm depth.

Keywords: Cyanobacterial inoculants, labile carbon, organic carbon fractions, soil chlorophyll, soil microbial biomass, system of rice intensification (SRI).

6.1 Introduction

Rice cultivation requires large amounts of water and due to growing scarcity of water farmers are shifting to cultivation of less water-demanding crops (Thiyagarajan, 2001). Increasing prices of agricultural inputs prevent poor farmers from adopting modern production technologies. Excessive use of agrochemicals (chemical fertilizers and pesticides/insecticides) damages soil biota (Stoop *et al.*, 2002). In such a situation, the system of rice intensification (SRI), which is a low-cost and high yielding system, might be a sustainable alternative to conventional paddy production (Batuvitage, 2002).

Diazotrophic cyanobacteria are natural renewable biological nitrogen sources that are being used as bio-fertilizer for rice (Sinha *et al.*, 2002). At least one third of Indian rice fields are amenable to cyanobacterial bio-fertilizer application. Besides bio-nitrogen, they contribute plant growth-promoting substances (Misra and Kaushik, 1989), prevent the growth of weeds (Subramanyan *et al.*, 1965), add soil organic matter (Roychoudhary *et al.*, 1980), increase phosphorus availability and decrease sulphide injury to rice seedlings (Arora, 1969). The effects on rice yield of soil inoculation by blue-green algae were first reported by Watanabe *et al.* (1951), with a 25% increase in yield after inoculation of poorly drained paddy with *Tolypothrix tenuis*. But, despite such natural inherent quality of cyanobacteria, use is restricted to a very limited hectareage. Adoption at the farmer's level is negligible due to erratic response. This is mostly due to technological problems governing inoculum quality, establishment under field situation and poor shelf-life (Jha and Prasad, 2006).

Information on organic C stocks in agricultural soils is important because of the effects of soil organic carbon (SOC) on crop production and on climate change. The SOC stock at any time reflects the long term balance between additions of organic C from different sources and its losses through different pathways. Following the adoption of large-scale intensive cropping, this long-term balances are modified since intensive cropping encourages oxidative losses of C due to continued soil disturbance, while it also leads to a large-scale addition of C to the soil through crop residues. This may cause either a net build up or a net depletion of SOC stock (Rasmussen *et al.*, 1980; Cole *et al.*, 1993; Kong *et al.*, 2005). To better understand the mechanisms by which C is lost or stabilized in soil, the SOC stock is separated into a labile or actively cycling pool, a slow pool, and a stable or passive, recalcitrant pool with varying residence times (Parton and Rasmussen, 1994). The labile C pool is the fraction of SOC with the most rapid turnover rates. This pool is important from the point of view of crop production. It fuels the soil food web and therefore greatly influences nutrient cycling for maintaining soil quality and its productivity (Chan *et al.*, 2001; Majumder, 2007). This pool is also sensitive to land management changes. The highly recalcitrant or passive pool is, on the other hand, altered only very slowly by microbial

activities and hence hardly serves as a good indicator for assessing soil quality and productivity (Weil *et al.*, 2003; Sherrod *et al.*, 2005; Majumder, 2007).

Only a very few studies have been done so far on the changes in labile pools of SOC in tropical and subtropical regions of the world (Rudrappa *et al.*, 2006), although these studies are of special importance in the region since the organic C stock of soils here is inherently low. This low C stock poses a serious threat to soil health and thus long term sustainable crop production in the region (Mandal, 2005; Sharma *et al.*, 2005). Farmers strive to increase productivity of rice-wheat cropping system in Indo-Gangetic Plains in India through the use of improved technologies but in the last decade or so, per unit productivity growth of rice-wheat cropping system is stagnating/ declining in this region (Duxbury, 2002), often resulting in food deficits in the country. This decline in productivity is attributed to the loss of soil organic matter, mineral nutrients, soil aggregates, and structural stability (Hobbs and Morris, 1996), which results in low soil fertility and productivity of the system. Several field researches in this region (Gami *et al.*, 2001; Regmi *et al.*, 2002; Shrestha *et al.*, 2006) urged the need of alternative ways for rebuilding soil organic matter on Indo-Gangetic Plains. With this background an experiment was conducted to determine the effect of methods of rice cultivation and inoculation of cyanobacterial inoculants on soil microbial parameters and soil carbon fractions and aggregation.

6.2 Materials and methods

The experiment was conducted during rainy (*khariif*) season (June-October) of 2011 at the Research Farm of Indian Agricultural Research Institute, New Delhi. The mean annual rainfall of Delhi is 650 mm and more than 80% generally occurs during the monsoon season (July-September) with mean annual evaporation 850 mm. The year 2011, had less than normal rainfall (539.4 mm during June to October) out of which 43.8, 226.8 and 163.6 mm rainfall which comprised 80.5 % of the total rainfall was received during July, August and September months, respectively. There was late onset of monsoon and July month had much less rainfall than normal. The soils of experimental field had 134.9 kg ha⁻¹ alkaline permanganate oxidizable N (Subbiah and Asija, 1956), 15.90 kg ha⁻¹ available P (Olsen's method, Jackson, 1973), 260.83 kg 1 N ammonium acetate exchangeable K (Prasad *et al.*, 2006) and 0.53 % organic carbon (Walkley and Black, 1934). The pH of soil was 8.1.

The experiment was laid out in split plot design with two methods of crop establishment viz., conventional transplanting (CT) and system of rice intensification (SRI) as main plot and subplot comprised of seven cyanobacterial inoculants viz., N₀; Recommended dose of N (120 kg ha⁻¹); Compost application (1/3 N) + 2/3 N; Biofilm based BGA bio-fertilizer + 2/3 N; BGA + PGPR + 2/3 N; Compost based BGA mixture inoculant + 2/3 N and *multani mitti* based BGA inoculant + 2/3 N. 2/3 N (80 kg ha⁻¹) and 120 kg ha⁻¹ was applied through urea in three splits and common dose of P₂O₅ (60 kg ha⁻¹) and K₂O (60 kg ha⁻¹)

¹) were applied in all the plots. *Basmati* rice variety 'Pusa *Basmati* 1401' was taken in the experiment and these treatments were replicated three times. In CT and SRI, 21 and 12 days old seedlings, respectively were transplanted in puddled fields.

The experimental field was disc-ploughed twice and levelled and after puddling 21 days old seedlings of rice varieties were transplanted under conventional transplanting at 20 cm × 10 cm spacing keeping 2 seedlings hill⁻¹. For SRI, 12 days old seedlings were transplanted in main field. In conventional transplanting method 3-5 cm cropping water was maintained from transplanting to grain filling stage of crop and later only moist soil conditions were maintained. However in SRI, alternate wetting and drying conditions were maintained throughout the cropping season. Crop was harvested in the last fortnight of October.

Observations on soil microbial parameters were taken before rice planting, at mid-season stage (60 DAT) and harvest stage of rice crop. The methodology standardized by Prasanna *et al.* (2003) regarding depth of soil cores, time of sampling was adopted. Soil microbial parameters like microbial biomass C (Nunan *et al.*, 1998), acetylene reductase activity (ARA) (Prasanna *et al.* 2003), dehydrogenase activity (Casida *et al.*, 1964) and soil chlorophyll (Nayak *et al.* 2004) were studied.

Estimation of soil carbon and aggregation

All the soil samples taken before planting and 60 days after transplanting (DAT) were brought to the laboratory, air dried and ground to pass a 2-mm sieve before analyses. The soil samples were analyzed for pH (1:2 soil: water suspension) with a glass electrode, electrical conductivity (EC, 1:2 soil: water supernatant) using a conductivity meter, particle size distribution by pipette method and bulk density by core method. The soils were non-saline, alkaline in reaction and sandy loam in texture.

The determination of oxidizable carbon was repeated using 5 and 10 ml of concentrated sulphuric acid instead of the 20 ml specified by Walkley and Black (1934). The resulting three acid-aqueous solution ratios of 0.5:1, 1:1 and 2:1 (which corresponded respectively to 12 N, 18 N, and 24 N of H₂SO₄) allowed comparison of oxidizable organic carbon extracted under increasing oxidizing conditions. The amount of oxidizable organic carbon determined using 5, 10, and 20 ml of concentrated sulphuric acid when compared with total carbon concentration allowed separation of total organic carbon into three fractions of decreasing oxidizability: Fraction 1 (12 N H₂SO₄)—organic carbon oxidizable under 12 N H₂SO₄, Fraction 2 (18 N-12 N H₂SO₄)—the difference in oxidizable organic carbon extracted between 18N and 12N H₂SO₄ and Fraction 3 (24N-18N H₂SO₄)—the difference in oxidizable organic carbon extracted between 24N and 18N H₂SO₄.

For soil aggregation, soil samples were collected from field at 60 DAT when soil moisture was near field capacity and then samples were air dried. Air-dried samples were

passed through 8 mm sieves. Where ever, clods were bigger and the amount of soil passing through was small, larger clods are gently broken apart at their natural cleavages. A known amount of soil sample (50 g) is immersed in water for short period of time for wetting. The wetted sample was sieved through the nest of sieves in Yoder's apparatus (Yoder, 1936) and mean weight diameter (MWD) was determined as:

$$\text{MWD} = \sum X_i W_i$$

Where, W_i is the proportion of each aggregate class i in relation to the weight of soil sample taken for analysis, X_i the mean diameter of the class (mm).

All the data obtained from the experiment were statistically analyzed using the F -test as per the procedure given by Gomez and Gomez (1984). LSD values at $P = 0.05$ were used to determine the significance of difference between treatment means. Interaction effect was also statistically analysed in the same design.

6. 3 Results

6. 3.1 Soil microbial parameters

6. 3.1.1 Acetylene reductase activity (ARA)

Soil acetylene reductase activity (ARA) was higher at 60 DAT and decreased to ward maturity (Table 6.1 and 6.2). ARA activity both at 60 DAT and at crop maturity in both conventional and SRI method was statistically on par with each other. Among sub-plot treatments, treatment containing BGA+PGPR and 2/3 nitrogen (fertilizer) showed highest ARA activity. But ARA at this treatment was statistically at par with other three treatment viz., Recommended dose of N (120 kg ha⁻¹), Compost application (1/3 N) + 2/3 N (fertilizer) and Compost based BGA mixture inoculant + 2/3 N (fertilizer). ARA in these four treatments were statistically higher than treatments having Biofilm based BGA biofertilizer + 2/3 N (fertilizer) and *Multani mitti* based BGA inoculant + 2/3 N (fertilizer). Similar trend was observed in the observation recorded at crop maturity. Treatment having no nitrogen recorded lowest ARA value being 0.74 and 0.70 mole ethylene g⁻¹ hr⁻¹ at 60 DAT and at crop maturity, respectively. Where as, treatment having BGA+PGPR and 2/3 N through urea recorded highest ARA value being 2.13 and 1.57 mole ethylene g⁻¹ hr⁻¹ at 60 DAT, and at crop maturity, respectively.

Interaction effect between planting method and cyanobacterial inoculants on soil ARA activity was found significant both at 60 DAT and at crop harvest (Table 3.1 a). Treatment containing BGA+PGPR and 2/3 N through chemical fertilizer showed highest value (2.31 mole ethylene g⁻¹ hr⁻¹) in SRI which was superior over same treatment in conventional method. This treatment was statistically significant over treatment containing no nitrogen in both SRI and conventional method of planting.



Plate 6 Extraction of chlorophyll from soil



Plate 7 Determination of dehydrogenase activity in soil



Plate 8 Soil aggregation as affected by method of planting and cyanobacterial inoculants



Plate 9 Taking optical density on spectrophotometer



Plate 10 Determination of soil aggregation

6.3.1.2 Soil chlorophyll

Soil chlorophyll content was measured before sowing, 60 DAT and at crop maturity (Table 6.1 and 6.2). It was observed that, chlorophyll content was higher at 60 DAT and decreased to ward maturity in all the treatments. Soil chlorophyll content both at 60 DAT and at crop maturity in both conventional and SRI methods was statistically non-significant. Among sub-plot treatments, treatment containing BGA+PGPR and 2/3 nitrogen (fertilizer) showed highest chlorophyll content which was 2.14 mg ml^{-1} at 60 days of crop growth. Other three treatment including recommended dose of N (120 kg ha^{-1}) (2.03 mg ml^{-1}), compost application ($1/3 \text{ N}$) + $2/3 \text{ N}$ (fertilizer) (1.96 mg ml^{-1}) and compost based BGA mixture inoculant + $2/3 \text{ N}$ (fertilizer) (1.96 mg ml^{-1}) are on par with BGA+PGPR and 2/3 nitrogen. All these four treatments are statistically significant over treatment containing Biofilm based BGA bio-fertilizer + $2/3 \text{ N}$ (fertilizer) and *Multani mitti* based BGA inoculant + $2/3 \text{ N}$ (fertilizer). Similar trend was observed at crop maturity. Highest soil chlorophyll values were recorded with treatment BGA+PGPR and $2/3 \text{ N}$ (fertilizer) being 2.14 and 1.75 mg ml^{-1} at 60 DAT, and at crop maturity, respectively. Treatment having no nitrogen application recorded lowest soil chlorophyll.

Interaction effect between planting method and cyanobacterial inoculants on soil chlorophyll content was found significant (Table 3.1 a). Within conventional method, BGA+PGPR and $2/3 \text{ N}$ through chemical fertilizer was statistically significant over treatment containing no nitrogen. This treatment also stands significant over no nitrogen treatment in SRI method. In SRI method, same treatment showed significantly superior over treatment containing no nitrogen in both SRI and conventional method. But BGA+PGPR and $2/3 \text{ N}$ through chemical fertilizer stands non-significant in both conventional and SRI method of cultivation.

6.3.1.3 Soil dehydrogenase activity

Soil dehydrogenase activity was measured before sowing, 60 DAT and at crop maturity. It was observed that the dehydrogenase activity was higher at 60 DAT and decreased at crop harvest stage (Table 6.1 and 6.2). Dehydrogenase enzyme activity both at 60 DAT and at crop maturity in both conventional and SRI method was statistically non-significant. Slightly higher value of dehydrogenase enzyme activity was observed in conventional method compared to SRI.

Among the treatments containing cyanobacterial inoculants, BGA+PGPR and 2/3 nitrogen (fertilizer) showed highest dehydrogenase activity which was $15.17 \mu\text{g TPF g}^{-1} \text{ soil day}^{-1}$ after 60 days of crop growth. This treatment and three other treatment including recommended dose of N (120 kg ha^{-1}) ($14.87 \mu\text{g TPF g}^{-1} \text{ soil day}^{-1}$), compost application ($1/3 \text{ N}$) + $2/3 \text{ N}$ (fertilizer) ($14.80 \mu\text{g TPF g}^{-1} \text{ soil day}^{-1}$) and compost based BGA mixture

inoculant + 2/3 N (fertilizer) ($14.58 \mu\text{g TPF g}^{-1} \text{ soil day}^{-1}$) had statistically significant values over the Biofilm based BGA biofertilizer + 2/3 N (fertilizer) and *Multani mitti* based BGA inoculant + 2/3 N (fertilizer). Similar trend was observed at crop maturity. Treatment having no nitrogen application recorded lowest value being at 5.02 and $2.69 \mu\text{g TPF g}^{-1} \text{ soil day}^{-1}$ at 60 DAT and at crop maturity, respectively. Treatment BGA+PGPR + 2/3 N (fertilizer) recorded highest value being 15.17 and $9.39 \mu\text{g TPF g}^{-1} \text{ soil day}^{-1}$ at 60 DAT and at crop maturity, respectively.

Interaction effect between planting method and cyanobacterial inoculants on soil dehydrogenase activity was found significant at 60 DAT and non-significant at harvest (Table 6.2.1). Within conventional method, BGA+PGPR and 2/3 N through chemical fertilizer was statistically significant over treatment containing no nitrogen. This treatment also stands significant over no nitrogen treatment in SRI method. In SRI method, same treatment showed significantly superior over treatment containing no nitrogen in both SRI and conventional method. But BGA+PGPR and 2/3 N through chemical fertilizer stands non-significant in both conventional and SRI method of cultivation.

6.3.1.4 Soil microbial biomass carbon (MBC)

Effect of two planting methods and different microbial inoculants on soil microbial biomass carbon (MBC) was recorded before transplanting, 60 DAT and at crop harvest (Table 6.2). Results showed that MBC was higher at 60 DAT and decreased at crop maturity stage. MBC both at 60DAT and at crop maturity in both conventional and SRI method was statistically at par. Among sub-plots, treatment containing BGA+PGPR and 2/3 N (fertilizer) showed highest MBC ($285.83 \mu\text{g MBC g}^{-1} \text{ soil}$) after 60 days of crop growth. Along with this treatment, three other treatment including recommended dose of N (120 kg ha^{-1}) ($280.8 \mu\text{g MBC g}^{-1} \text{ soil}$), compost application (1/3 N) + 2/3 N (fertilizer) ($268.67 \mu\text{g MBC g}^{-1} \text{ soil}$) and compost based BGA mixture inoculant + 2/3 N (fertilizer) ($264.17 \mu\text{g MBC g}^{-1} \text{ soil}$) showed significantly higher MBC over treatments having Biofilm based BGA biofertilizer + 2/3 N (fertilizer) and *Multani mitti* based BGA inoculant + 2/3 N (fertilizer). Similar trend in MBC was observed at crop maturity. Treatment having no nitrogen recorded lowest value. Highest MBC values were recorded with treatment BGA+PGPR and 2/3 N (fertilizer).

Interaction effect between planting method and cyanobacterial inoculants on soil microbial biomass carbon (MBC) was found significant both at 60 DAT and at crop harvest (Table 6.2.2). Within conventional method, BGA+PGPR and 2/3 N through chemical fertilizer was statistically significant over treatment containing no nitrogen. This treatment also stands significant over no nitrogen treatment in SRI method. In SRI method, same treatment showed significantly superior over treatment containing no nitrogen in both SRI and conventional method. But BGA+PGPR and 2/3 N through chemical fertilizer stands non-

significant in both conventional (289.33 $\mu\text{g MBC g}^{-1}\text{soil}$) and SRI method (282.33 $\mu\text{g MBC g}^{-1}\text{soil}$) of cultivation.

Table 6.1 Effect of planting methods and cyanobacterial inoculants on soil microbial parameters at 60 DAT

Treatments	ARA (n mole ethylene $\text{g}^{-1}\text{hr}^{-1}$)	Soil chlorophyll (mg ml^{-1})	Dehydroge -nase ($\mu\text{g TPF g}^{-1}$ soil day^{-1})	Microbial biomass carbon (μg MBC g^{-1} soil)
Planting Method				
Conventional transplanting	1.71	1.788	12.78	232.10
SRI	1.73	1.769	12.87	235.29
SEm \pm	0.007	0.007	0.061	0.838
LSD (P=0.05)	NS	NS	NS	NS
Bacterial inoculants				
N ₀	0.74	0.947	5.02	84.00
Recomm. dose of N (120 kg ha ⁻¹)	1.92	2.032	14.87	280.83
Compost application (1/3 N) + 2/3 N	1.98	1.969	14.80	268.67
Biofilm based BGA bioferti. + 2/3 N	1.67	1.715	12.78	229.67
BGA + PGPR + 2/3 N	2.13	2.140	15.17	285.83
Compost bas. BGA mix. Inoc.+ 2/3 N	2.01	1.962	14.58	264.17
Multani mitti based BGA inoc.+ 2/3 N	1.62	1.683	12.57	222.67
SEm \pm	0.08	0.064	0.45	10.43
LSD (P=0.05)	0.23	0.188	1.32	30.43
Bacterial inoculants x planting Method				
SEm \pm	0.11	0.09	0.63	14.73
LSD (P=0.05)	Sig.	Sig.	Sig.	Sig.

Table 6.1.1 Interaction effect of planting method and cyanobacterial inoculants on soil ARA and soil chlorophyll

Planting methods	Bacterial inoculants						
	T ₁	T ₂	T ₃	T ₄	T ₅	T ₆	T ₇
ARA (n mole ethylene g ⁻¹ hr ⁻¹) at 60 DAT							
Conventional	0.89	1.63	2.17	2.09	1.95	1.86	1.42
SRI	0.59	2.20	1.80	1.24	2.31	2.17	1.82
S. Em±				0.111			
LSD (P=0.05)				0.32			
Soil chlorophyll (mg ml ⁻¹) at 60 DAT							
Conventional	1.11	2.37	1.84	1.54	2.21	1.68	1.77
SRI	0.78	1.70	2.10	1.89	2.07	2.24	1.60
S. Em±				0.091			
LSD (P=0.05)				0.27			

6.3.2 Soil carbon content

Carbon content of soil was analysed before transplanting and at 60 DAT at two different depths viz., 0-7.5 cm and 7.5-15 cm by Chan *et al.*(2001) modified Walkley and Black method and three fractions of carbon viz., very labile, labile and less labile was determined (Table 6.3). Very labile carbon fraction at 0-7.5 cm depth layer was marginally higher (0.78%) in SRI method of cultivation compared to conventional method (0.76%). This subtle difference might be due to higher microbial activity and better soil physical condition in top 0-7.5 cm depth under SRI compared to conventional method of cultivation. Very labile fraction of C content between these methods was also stand on par to each other at 7.5-15 cm layer.

Among the sub-plot treatments, compost (1/3 N) + 2/3 N (fertilizer) showed highest C content (0.83%) after 60 days of crop growth. Carbon content in other treatments including RDN (120 kg ha⁻¹) (0.81%), BGA+PGPR + 2/3 nitrogen (fertilizer) (0.81%) and compost based BGA mixture inoculant + 2/3 N (fertilizer) (0.80%) were statistically at par. However, all these four treatments had statistically higher C content over the treatment with Biofilm based BGA biofertilizer + 2/3 N (fertilizer) and *Multani mitti* based BGA inoculant + 2/3 N (fertilizer). But differences in C content were non-significant among all these treatments at 7.5-15 cm depth. Labile fraction of C was similar between the method of cultivation at both the depth. In sub-plots no significant difference in labile C fraction between cyanobacterial

Table 6.2 Effect of planting method and cyanobacterial inoculants on soil microbial properties at crop harvest

Treatments	ARA (n mole ethylene g ⁻¹ hr ⁻¹)	Soil chlorophyll (µg g ⁻¹ soil)	Dehydrogenase (µg TPF g ⁻¹ soil day ⁻¹)	Microbial biomass carbon (µg MBC g ⁻¹ soil)
Planting Method				
Conventional transplanting	1.32	1.39	7.93	142.81
SRI	1.36	1.39	8.03	154.71
SEm±	0.011	0.004	0.044	5.150
LSD (P=0.05)	NS	NS	NS	NS
Bacterial inoculants				
N ₀	0.70	0.514	2.69	57.00
Recommended dose of N (120 kg ha ⁻¹)	1.46	1.686	9.29	183.67
Compost application (1/3 N) + 2/3 N	1.49	1.613	9.24	178.67
Biofilm based BGA biofert. + 2/3 N	1.31	1.323	8.12	131.83
BGA + PGPR + 2/3 N	1.57	1.754	9.39	186.50
Compost bas.BGA mixt. Inocu. + 2/3 N	1.51	1.572	9.14	170.67
Multani mitti based BGA inoc. + 2/3 N	1.26	1.284	7.98	133.00
SEm±	0.05	0.072	0.25	9.76
LSD (P=0.05)	0.14	0.209	0.73	28.49
Bacterial inoculants x Methods				
SEm±	0.067	0.10	0.35	13.80
LSD (P=0.05)	Sig.	Sig.	NS	Sig.

Table 6.2.1 Interaction effect of planting method and cyanobacterial inoculants on soil dehydrogenase activity and Soil MBC

Planting methods	Bacterial inoculants						
	T ₁	T ₂	T ₃	T ₄	T ₅	T ₆	T ₇
Dehydrogenase ($\mu\text{g TPF g}^{-1}$ soil day⁻¹) activity at 60 DAT							
Conventional	5.89	14.20	16.23	12.16	15.83	11.85	13.32
SRI	4.16	15.54	13.38	13.41	14.50	17.30	11.82
S. Em±				0.64			
LSD (P=0.05)				1.86			
Microbial biomass carbon ($\mu\text{g MBC g}^{-1}$ soil) at 60 DAT							
Conventional	76.00	281.00	342.00	190.00	289.33	133.67	312.67
SRI	92.00	280.67	195.33	269.33	282.33	394.67	132.67
S. Em±				14.74			
LSD (P=0.05)				43.03			

Inoculant treatment with recommended dose of N (fertilizer). Same trend was observed in case of less labile C fraction in both main and sub-plot treatments. Treatment receiving no N, recorded the least content of all C fractions at both the soil depth. Carbon content was higher in 0-7.5 cm depth compared to 7.5-15 cm depth. Very labile C was greater as compared to labile or less labile C fractions.

Interaction between planting method and cyanobacterial inoculants on C content was found significant in all the three fractions (Table 6.3.1). Treatment containing (1/3 N) + 2/3 N (fertilizer) showed statistically significant carbon in very labile fraction over no nitrogen in 0-7.5 cm depth in SRI method; While the same treatment shows significantly higher value at 7.5-15 cm depth over no nitrogen in conventional method of planting.

6.3.3 Soil aggregation

Aggregation property of soil was determined by taking soil sample at 60 DAT at two different viz., 0-7.5 cm and 7.5-15 cm. Aggregation properties were expressed in terms of mean weight diameter (MWD) of aggregate. Soil aggregations at both the depths were statistically at par in both the methods of rice cultivation. In sub-plot treatments it was found that at 0-7.5 cm depth treatment containing compost application (1/3 N) + 2/3 N (fertilizer) showed highest mean weight diameter (MWD) of aggregates as 1.12 mm after 60 DAT. Other three treatments viz., recommended dose of N (120 kg ha⁻¹) (1.11 mm), BGA+ PGPR and 2/3 nitrogen (fertilizer) (1.09 mm) and compost based BGA mixture inoculant + 2/3 N (fertilizer) (1.08 mm) were similar in MWD values.

Table 6.3 Effect of planting methods and cyanobacterial inoculants on soil C content at 60 days after transplanting

Treatments	Labile carbon fraction (cm)					
	Very labile		Labile		Less labile	
	0-7.5	7.5-15	0-7.5	7.5-15	0-7.5	7.5-15
Planting method						
Conventional	0.76	0.61	0.67	0.55	0.45	0.28
SRI	0.78	0.61	0.67	0.56	0.45	0.29
SEm±	0.005	0.003	0.003	0.004	0.002	0.001
LSD (P=0.05)	S	NS	NS	NS	NS	NS
Bacterial inoculants						
N ₀	0.64	0.55	0.58	0.48	0.32	0.22
Recommended dose of N (120 kg ha ⁻¹)	0.81	0.63	0.68	0.58	0.47	0.30
Compost application (1/3 N) + 2/3 N	0.83	0.63	0.70	0.60	0.48	0.30
Biofilm based BGA biofertilizer + 2/3 N	0.74	0.62	0.68	0.55	0.47	0.29
BGA + PGPR + 2/3 N	0.81	0.62	0.68	0.56	0.47	0.29
Compost based BGA mixture inoculant + 2/3 N	0.80	0.62	0.68	0.56	0.47	0.29
Multani mitti based BGA inoculant + 2/3 N	0.73	0.61	0.68	0.55	0.47	0.29
SEm±	0.01	0.012	0.009	0.022	0.006	0.004
LSD (P=0.05)	0.04	0.03	0.03	0.06	0.02	0.01
Bacterial inoculants x Method						
SEm±	0.01	0.016	0.012	0.031	0.008	0.006
LSD (P=0.05)	Sig.	Sig.	Sig.	Sig.	Sig.	Sig.

All these four treatments had significantly higher MWD of aggregates compared to Biofilm based BGA biofertilizer + 2/3 N through chemical fertiliser and *Multani mitti* based BGA inoculant + 2/3 N (fertilizer). This significant difference disappeared at 7.5-15 cm depth. Treatments with no nitrogen application recorded lowest MWD of aggregates at both the soil depth (0.58 and 0.26 mm at 0-7.5 cm and 7.5-15 cm depth, respectively). MWD were higher in 0-7.5 cm depth compared to 7.5-15 cm depth. This difference may be due to higher C content in top 7.5 cm soil compared to 7.5-15 cm depth.

Table 6.4 Effect of planting method and cyanobacterial inoculants on soil aggregation at 60 DAT

Treatments	Mean weight diameter (MWD) (mm)	
	Depth (cm)	
	0-7.5 cm	7.5-15 cm
Planting method		
Conventional	0.99	0.59
SRI	1.00	0.60
SEm±	0.004	0.005
LSD (P=0.05)	NS	NS
Bacterial inoculants		
N ₀	0.58	0.26
Recommended dose of N (120 kg ha ⁻¹)	1.11	0.67
Compost application (1/3 N) + 2/3 N	1.12	0.68
Biofilm based BGA biofertilizer + 2/3 N	1.01	0.64
BGA + PGPR + 2/3 N	1.09	0.65
Compost based BGA mixture inocu.+ 2/3 N	1.08	0.65
Multani mitti based BGA inoculant + 2/3 N	0.99	0.64
SEm±	0.018	0.016
LSD (P=0.05)	0.05	0.048
Bacterial inoculants x Planting method		
SEm±	0.025	0.023
LSD (P=0.05)	Sig.	Sig.

Interaction between planting methods and cyanobacterial inoculants on mean weight diameter (MWD) was found significant at both the soil depths (Table 6.4.1). Within conventional method, treatment containing (1/3 N) + 2/3 N (fertilizer) was statistically significant over treatment containing no nitrogen. This treatment also stands significant over no nitrogen treatment in SRI method. In SRI method, same treatment showed significantly superior over treatment containing no nitrogen in both SRI and conventional method.

Table 6.3.1 Interaction effect of planting method and cyanobacterial inoculants on soil carbon content

Planting methods	Bacterial inoculants						
	T ₁	T ₂	T ₃	T ₄	T ₅	T ₆	T ₇
Very labile carbon (0-7.5 cm) at 60 DAT (%)							
Conventional	0.64	0.74	0.68	0.28	0.49	1.31	1.16
SRI	0.64	0.88	0.98	1.21	1.14	0.30	0.30
S. Em±				0.018			
LSD (P=0.05)				0.05			
Very labile carbon (7.5-15 cm) at 60 DAT (%)							
Conventional	0.51	0.62	0.68	0.62	0.60	0.62	0.61
SRI	0.58	0.65	0.58	0.62	0.64	0.62	0.62
S. Em±				0.016			
LSD (P=0.05)				0.05			

6.4 Discussion

Soil ARA is the indicator of nitrogen fixation ability of the cyanobacteria. The principle in determination of Soil ARA is that, cyanobacteria have ability to reduce acetylene to ethylene. In this experiment, ARA activity was higher at 60 DAT and decreased to ward crop maturity. ARA activity both at 60 DAT and at crop maturity in both conventional and SRI method was statistically at par. Among sub-plot treatments, treatment containing BGA+PGPR and 2/3 N (fertilizer) showed highest ARA activity. Treatment having no nitrogen recorded lowest ARA value. Addition of N fertilizer is known to have a considerable influence on ARA (Singh and Bisoyi, 1989) and deep placement of N fertilizer in the soil is recommended for the dual benefit of N fertilizer and biological N₂ fixation. Higher levels of N are known to inhibit N fixation by BGA (Rowell *et al.*, 1977; Mekonnen *et al.*, 2002; Prasanna *et al.*, 2003). However, Nayak *et al.* (2004) found maximum ARA in N₃₀ treatments at 45 DAT while at 90 DAT the maximum values were recorded in plots receiving the N₉₀ level of urea application, indicative of tolerance of BGA/*Azolla* up to 90 kg N ha⁻¹. This study demonstrated that the fertility of rice fields depends not only on surface colonizing/floating BGA and *Azolla* but also on the direct and indirect interactions of BGA with the microflora in the below-surface soil environment.

Soil chlorophyll content is the measure of growth of cyanobacteria in soil. In present study, it was observed that soil chlorophyll content both at 60 DAT and at crop maturity in

Table 6.4.1 Interaction effect of planting method and cyanobacterial inoculants on mean weight diameter (mm)

Planting methods	Bacterial inoculants						
	T ₁	T ₂	T ₃	T ₄	T ₅	T ₆	T ₇
Mean weight diameter (MWD) (mm) (0-7.5 cm) at 60 DAT							
Conventional	0.59	1.20	1.18	1.23	1.31	0.71	0.74
SRI	0.57	1.02	1.05	0.80	0.87	1.46	1.25
S. Em±				0.023			
LSD (P=0.05)				0.07			
Mean weight diameter (MWD) (mm) (7.5-15 cm) at 60 DAT							
Conventional	0.23	0.69	0.58	0.59	0.56	0.74	0.74
SRI	0.30	0.64	0.79	0.69	0.74	0.56	0.53
S. Em±				0.025			
LSD (P=0.05)				0.07			

both conventional and SRI methods was statistically non-significant. Highest soil chlorophyll was recorded with treatment BGA+PGPR at 60 DAT, and at crop maturity. Treatment having no nitrogen application recorded lowest soil chlorophyll. Effectiveness of BGA in enhancing soil chlorophyll content was also increased due to the application of compost with BGA. But, Nayak *et al.* (2004) did not find any significant effect on soil chlorophyll content due to different nitrogen levels. However, at 45 DAT the overall effect of biofertilizers (BGA + *Azolla*, and *Azolla* alone) on soil chlorophyll content was significantly higher than the control or application of BGA biofertilizers alone, although individual combinations of biofertilizers and nitrogenous fertilizers did not exhibit any significant differences. In all treatments involving biofertilizers (BGA, *Azolla* and BGA + *Azolla*), a distinct increase in chlorophyll (% over control/ uninoculated) was observed with maximum values (up to a fourfold increase over the control) being observed in N₁₂₀ + BGA + *Azolla* treated plots. However, no definite pattern of increase or decrease in soil chlorophyll was observed at 90 DAT. The N₆₀ level of N fertilization and control plots (no bio-fertilizers) exhibited the highest soil chlorophyll at 90 DAT. Soil cores from N₉₀ + BGA + *Azolla* and N₁₂₀ + BGA and BGA + *Azolla* treatments exhibited increased soil chlorophyll over the control. This clearly indicated that inoculated BGA and *Azolla* bio-fertilizers bring about an enhancement in biomass, in terms of soil chlorophyll, in contrast with numerous reports

available on the growth inhibition of BGA at high levels of N fertilization (Mekonnen *et al.*, 2002).

Dehydrogenase enzyme activity was at par at 60 DAT and at crop maturity in conventional and SRI method BGA+PGPR and 2/3 nitrogen (fertilizer) showed highest dehydrogenase activity after 60 days of crop growth. Dehydrogenase is a respiratory chain enzyme of soil microorganisms, which is an indicator of biological redox system and microbial activity. Dehydrogenase activities were significantly higher in case of SRI as compared to other conventional and double transplanting (Suryavanshi, 2011). Microbial biomass and soil enzymes are considered as potential indicator of soil quality due to their relationship to soil biology, ease of measurement, rapid responses to changes in soil management and high sensitivity to changes originated by management and environment factor (Karlen *et al.*, 1997; Marx, 2001; Jimenez *et al.*, 2002). The rationale for the use of microbial and biochemical characteristics as soil quality indicators is their central role in cycling of C and N and their sensitivity to change (Nannipieri *et al.*, 1990). Although, it is now well established that many soil microorganisms can produce plant growth regulating substances (Phytohormones), little has been done to exploit the influence of microbially-produced phytohormones on plant growth and development, partly because our knowledge is still so incomplete, but even more so because the processes involved are so complex. Influence on dehydrogenase enzyme activity was further enhanced when PGPR and BGA were applied with compost. Dehydrogenase and alkaline phosphatase activities in soil increased with the application of farmyard manure and *S. aculeata* green manure (Dhull *et al.*, 2004). Application of green manures *S. aculeata* significantly increased the activity of urease, dehydrogenase, amidase and phosphatase over prilled urea (50-150 kg N ha⁻¹) and control (Sriramachandrasckharan, 2002). The increase in dehydrogenase and alkaline phosphatase activities with increasing dose of fertilizer as well as organic amendments is reflections of organic matter build up by incorporation of green manuring and which led to increase in microbial activities (Tarafdar *et al.*, 1989; Pascual *et al.*, 2002). Punia (2010) also reported significantly higher microbial biomass C and alkaline dehydrogenase, phosphatase and FDA enzymatic activity in *Basmati* rice soils when *Basmati* rice was grown after incorporation of *S. aculeata* and lowest was recorded with summer fallow. *S. aculeata* green manuring crop supplied readily mineralizable C, N and improved soil organic matter status, which led to the more microbial biomass C (Chander *et al.*, 1997; Tilak, 2004). The size of soil microbial biomass C is governed by various management practices such as crop rotation, organic amendments, crop residue management and green manuring. The supply of readily metabolizable C by organic manure is likely to have been one of the most influential factors contributing to the biomass increases. This suggestion is consistent with the observations of Hopkins and Shiel (1996). Soil microbial biomass C is a small component

of soil organic matter and which is directly related with soil organic matter status (Dhull *et al.*, 2004; Jedidi *et al.*, 2004). Fertilizer application particularly N and increasing inputs of organic residue increase the soil organic matter and microbial biomass carbon (Mahmood *et al.*, 1997; Graham *et al.*, 2002).

MBC both at 60DAT and at crop maturity in both conventional and SRI method was statistically at par. Suryavanshi (2011) recorded higher MBC in soil under SRI compared to conventional and double transplanting of rice. Treatment containing BGA+PGPR and 2/3 nitrogen (fertilizer) showed highest MBC; While treatment having no nitrogen recorded lowest MBC value. Soil microbial biomass is fundamental to maintaining soil functions as it represents the main source of soil enzymes that regulate transformation processes of elements in soils. It also controls the build-up and breakdown of organic matter, the decomposition of organic residues, and serves as early indicator of changes in soil management. Rupela *et al.* (2006) reported that MBC was higher in SRI plots than those of conventional flood rice. Microbial activity in soil is dictated by availability of nutrients in an environment and the increased activity reflects the increased availability of nutrients. Rice planting methods did not influence soil chlorophyll content significantly. All the three methods showed at par soil chlorophyll content, but CT recorded higher chlorophyll than the other two methods. This may be due to a greater population of cyanobacteria, commonly found as floating assemblages in flooded rice fields (similar to CT). Cyanobacteria have been widely employed as inoculants for enhancing soil fertility and improving soil structure, besides increasing crop yields, especially in rice crop (Nayak *et al.*, 2004; Prasanna *et al.*, 2009a,b).

However, the role of cyanobacteria under different crop establishment modes has not been evaluated. The SRI methodology may be favourable for the growth of these organisms, as aerobic environment, followed by micro-aerobic environment may prevail alternatively. Analyses of cyanobacteria from several rice based agro-ecologies of India have revealed that ecology/ecotype of the region or carbon/moisture levels does not alter the ratio of the dominant cyanobacterial members (Nayak *et al.*, 2001; Prasanna and Nayak, 2007) however, their abundance and diversity can be governed by the rice varieties (Prasanna *et al.*, 2009a). The size and activity of the soil microbial biomass depend on quantity and quality of soil organic matter (Weigand *et al.*, 1995) and is related to soil properties (Kaiser *et al.*, 1992), climatic conditions (Grisi *et al.*, 1998), and crop management (Heisler and Kaiser, 1995; Kandeler *et al.*, 1999; Nieder *et al.*, 2008).

Three fractions of carbon viz., very labile, labile and less labile were determined in carbon content of soil before transplanting and at 60 days after transplanting (DAT) at two different depths viz., 0-7.5 cm and 7.5-15 cm. Very labile fraction of C content between these methods was also similar at 7.5-15 cm layer. Treatments with compost (1/3 N) + 2/3 N (fertilizer) showed highest C content (0.83%) after 60 days of crop growth. Labile fraction of

C was on par between the method of cultivation at both the depth. Sub-plots showed no significant difference in labile C fraction between cyanobacterial inoculant treatment with recommended dose of N (fertilizer). Same trend was observed in case of less labile C fraction in both main and sub-plot treatments. Treatment receiving no N, recorded the least content of all C fractions at both the soil depth. Carbon content was higher in 0-7.5 cm depth compared to 7.5-15 cm depth. Positive effect of manure application on SOC build-up under maize-wheat system in semi-arid India has earlier been reported by Benbi *et al.* (1998).

Barreto *et al.* (2011) also showed that labile C fraction constitutes a larger fraction, 54–59%, of the TOC in soils under agroforestry systems in Brazil. The results suggested that the organic carbon in soils under maize-wheat and agroforestry systems is less stable and could be easily lost through organic matter decomposition if the current land-use is discontinued. On the contrary, soils under rice-wheat contained greater proportion (63%) of organic C in less labile and recalcitrant fractions. Our values are similar to those reported by Majumder *et al.* (2008) for rice based cropping systems in hot humid tropics of India, where in rice-berseem cropping system, the less labile and recalcitrant fractions (fraction 3 and fraction 4) together constituted about 55% of the total organic carbon. The greater proportion of organic C existing in less labile and recalcitrant forms in rice-wheat system may be attributed to the retarded rate of C oxidation in these soils that are flooded for 3 months during the rice season (Jenkinson, 1988; Watanabe, 1984). As a result of slower decomposition of C substrates in anaerobic soils than in aerated soils, plant molecules that are more resistant to microbial degradation such as lignins might gradually accumulate in the organic matter of paddy soils (Colberg, 1988; Tate, 1979). Several studies provided indirect evidence for the accumulation of lignin-derived substances in submerged soils (Mitsuchi, 1974; Tsutsuki and Kuwatsuka, 1979; Ye and Wen, 1991). Slower lignin decomposition in submerged soils leads to incorporation of phenolic moieties into young SOM fractions (Olk *et al.*, 1996), imparting considerable recalcitrance to soil organic matter.

Aggregation properties were expressed in terms of mean weight diameter of aggregate. Soil aggregations at both the depths were statistically at par in both the methods of rice cultivation. In sub-plot treatments it was found that at 0-7.5 cm depth treatment containing compost application (1/3 N) + 2/3 N (fertilizer) showed significantly higher mean weight diameter (MWD) of aggregates compared Biofilm based BGA biofertilizer + 2/3 N through fertilizser and *Multani mitti* based BGA inoculant + 2/3 N (fertilizer). MWD were higher in 0-7.5 cm depth compared to 7.5-15 cm depth. This difference may be due to higher C content in top 7.5 cm soil compared to 7.5-15 cm depth. Cyanobacteria are known to contribute to macro-aggregation and result in improved resistance to soil erosion, because as primary producers, they contribute to the enrichment of soil with soil organic matter (SOM) and to the improvement of biological activity (Acea *et al.*, 2003). Cyanobacteria are known to

secrete extracellular polymeric substances (EPS) dominated by polysaccharides which can bind soil particles (Issa *et al.*, 2001). Prasanna *et al.* (2009) reported the significant contribution of cyanobacterial biomass in enhancing microbial activity and plant growth promotion, emphasizing their significance in sustainable management of rice ecosystem. Issa *et al.* (2007) observed improvement in the aggregation of cyanobacteria inoculated soils which could be related to increase in soil carbon and EPS that cause change in the soil micro-morphological characteristics of the aggregates. Maqubela *et al.* (2009) reported that inoculation of degraded soil with strain of *Nostoc* could improve the fertility and structural stability of degraded soil.

6.5 Conclusion

Microbial parameters including acetylene reductase activity (ARA), soil chlorophyll, dehydrogenase activity and microbial biomass carbon (MBC) content of soil were at par under conventional and SRI methods at 60 DAT or at crop maturity. ARA, soil chlorophyll, dehydrogenase activity and MBC were higher at 60 DAT compared to those at crop harvest. Treatment with BGA + PGPR + 2/3 N (fertilizer) showed highest ARA, soil chlorophyll and dehydrogenase activity at 60 DAT and at crop maturity. Application of compost (1/3 N) + 2/3 N (fertilizer) showed higher carbon content in all the three fractions, very labile, labile and less labile, although very labile C at 0-7.5 cm depth was marginally different except in *Multani mitti* based BGA inoculant + 2/3 N (fertilizer) and biofilm based BGA bio-fertilizer 2/3 N (fertilizer). Highest mean weight diameter (MWD) indicating better soil aggregation was recorded with compost application (1/3 N) + 2/3 N through fertilizer.

7. GENERAL DISCUSSION

System of Rice Intensification (SRI) method of cultivation is slowly gaining momentum all over the world including India. At least one third of Indian rice fields are amenable to cyanobacterial bio-fertilizer application. The SRI can help in water saving and use of cyanobacterial inoculants in rice can replace chemical fertilizer to some extent. The results of the investigation entitled “**Influence of cyanobacteria based inoculants and cultivation methods on rice (*Oryza sativa* L.) yield, soil carbon content and aggregation**” carried out at IARI, New Delhi are discussed in this chapter.

Significantly higher plant height, tillers, dry matter accumulation and LAI were recorded in conventional transplanting (CT) method compared to system of rice intensification (SRI) at 30 days after transplanting (DAT). Higher plant growth in CT over SRI might be due to shorter/ younger (12 days old) seedling of rice planted in SRI, whereas in CT 21 days old seedlings were planted. Younger seedling in SRI took longer time to absorb transplanting shock and mortality of seedlings was 15 % higher in SRI compared to CT. Adverse climatic condition prevailing during 2011 wet season (Fig. 3.1) having almost no rainfall in July month resulting in continuous higher temperature caused mortality of tender seedling in early age in SRI. However, plant height under CT and SRI at 60 and 90 DAT and at harvest stage were statistically at par. Contradictory reports are found about the effect of methods of planting on rice plant growth. Nissanka and Bandara (2004) found no variation in plant height between the SRI and conventional transplanting. Haque (2002) found the highest plant height from wider spacing as done in SRI. Whereas, Akita and Tanaka (1992) reported that at maturity the tallest plants were found at low plant density. Younger seedlings had more vigour, root growth and lesser transplant shock because of lesser leaf area during initial growth stages which stimulate increased cell division causing more stem elongation which might increased plant height of SRI (Rahman, 2001 and Sangsu *et al.*, 1999). Shrirame *et al.* (2000) reported that the number of functional leaves, leaf area and total number of tillers hill⁻¹ were higher at wider spacing which increased the photosynthetic rate leading to taller plants. Shortening of plants in tiller crop at harvest might be due to shortage of time for proper vegetative growth and development. Similar results were also reported by many workers (Rahman, 2001; Khisha, 2002; Sarker *et al.*, 2002). The practice of transplanting one young seedling hill⁻¹ with wider spacing (SRI) had advantages in reducing transplanting injury and increasing tillers (Horie, 2004) and minimizes the competition between plants (Rabenandrasana, 1999). Earlier transplanting induced the transplanting shock at a more convenient point in the growth cycle when they could rebound faster and had little effect on tillerage (Uphoff, 2002). Alternate wetting and drying maintaining a thin film of water that

might open the soil for both oxygen and nitrogen and promoted the root growth during initial growth stages which ultimately increased tiller density (Uphoff, 2001). Nissanka and Bandara (2004) observed that the tiller number plant⁻¹ were higher in the SRI compared to conventional transplanting, conventional broadcasting and high density broadcasting, but this parameter, when expressed per unit area basis were not significantly different. However, Karmakar *et al.* (2004) and Lu *et al.* (2004) found higher tillers in closer spacing that contributed to obtain higher yield. Kim *et al.* (1999) and Ray *et al.* (2000) reported higher LAI in closer spacing, but Vijayakumar *et al.* (2006) observed maximum LAI in wider spacing.

Contradictory reports are available about the effect of SRI on yield attributes of rice. In this study no difference was seen on yield attributes viz., panicle number m⁻², weight and length of panicles, number of grain panicle⁻¹ and test weight of rice grain under conventional and SRI method of planting. However, Husain *et al.* (2004) observed higher number of effective tiller and number of grains per panicle under SRI compared to farmers practice. Sengthong (2002) noticed highest number of effective tillers when transplanted at spacing of 40 cm × 40 cm and Wang *et al.* (2002) reported higher number of effective tillers in the younger seedlings. Gasparillo *et al.* (2001) noted significant variation of panicle length between the SRI and non-SRI methods. Ang *et al.* (2002) obtained maximum filled grains panicle⁻¹ from 40 cm × 40 cm spacing of the SRI. Like this experiment, Bari (2004) also observed that planting method had no significant influence on 1000-grains weight. But Husain *et al.* (2004) and Hossain *et al.* (2003) observed higher 1000-grains weight under SRI compared to the farmers practice.

Grain and straw yield and harvest index were statistically at par in conventional and SRI method of planting. Different types of report are found about the effect of planting on rice yield. Nissanka and Bandara (2004) reported that grain yield was 7.6 t ha⁻¹ in the SRI and it was 9% greater than the CT which might be attributed to the vigorous and healthy growth, development of more productive tillers and leaves ensuring greater resource utilization in the SRI compared to CT. Husain *et al.* (2003a) found 39% higher straw yield in SRI compared to traditional methods. Suryavanshi *et al.* (2012) reported significantly higher grain yield in SRI compared to conventional transplanting. Stoop (2005) and Hossain *et al.* (2003) also found higher harvest index in SRI compared the CT, though, Barison (2003) found no difference for the same.

Inoculation of cyanobacterial inoculants and recommended dose of nitrogen showed significantly higher plant height, tillers m⁻², dry matter accumulation and leaf area index (LAI) over N control; However, difference between the effect of cyanobacterial inoculants along with 2/3 N and recommended dose of N was non-significant at 30, 60 and 90 DAT and at harvest. Free-living cyanobacteria are known to contribute an average of 20-30 kg N ha⁻¹,

whereas the value is up to 600 kg ha⁻¹ for the *Azolla-Anabaena* system. Besides their N-enrichment potential, they are known to increase the water holding capacity, porosity and cation exchange capacity of soil. Problems associated with the establishment of inoculated strains (mainly due to competition with native strains in soil) and inconsistency in crop response as a result of environmental fluctuations has slowed down their adoption by farmers all over the world (Lee and Watanabe, 1977; Whitton, 2000). A positive effect of bacterial inoculations on roots of maize and wheat has been reported (Creus *et al.*, 1996; Jacoud *et al.*, 1999). Similar beneficial effects of bacterial inoculations were also observed on plant biomass and root growth of rice varieties (Mehnaz *et al.*, 2000).

Sakthivel *et al.* (1986) reported 12-27% greater plant height in response to PGPR inoculation than the non-inoculated control in rice. Plant height increased with increasing level of N either through PU and ZEU (Jaiswal and Singh, 2001; Ghatak *et al.*, 2005). Sathia and Ramesh (2009) also reported positive influence on growth parameters like plant height, tillers and dry matter production of aerobic rice were positively influenced by different nitrogen management practices. The usefulness of increased N application on tiller production was also observed by Singh *et al.* (2006). The PGPR isolates significantly increased the shoot length of rice seedlings (Ashrafuzzaman *et al.*, 2009). Van *et al.* (2000) reported that inoculation had a significant positive effect on tillers hill⁻¹ (13%) and shoot weight (33%). The usefulness of increased N application on tiller production was also observed by Singh *et al.* (2006). Van *et al.* (2000) have reported an increase in 30% leaf area of rice due to inoculation of PGPR in pot and field experiments over no inoculation. Ashrafuzzaman *et al.* (2009) found a significant increase in shoot dry matter of rice seedlings was observed in response to PGPR isolates.

Significantly higher yield attributes like panicle m⁻², weight and no. of grains panicle⁻¹ were found with cyanobacterial inoculants and RDN (120 kg ha⁻¹) over N control. The treatment BGA+PGPR and 2/3 nitrogen (fertilizer) had highest values in all the parameters. But this treatment was statistically at par over with RDN, compost application (1/3 N) + 2/3 N (fertilizer) and Compost based BGA mixture inoculant + 2/3 N (fertilizer). Test weight difference was at par in all the treatments including treatment having N₀. Biswas *et al.* (2000a) reported significantly increase in number of panicles and filled grains panicle⁻¹, and also the total number of spikelets plant⁻¹ in PGPR-inoculated rice compared to un-inoculated plants. Similarly, Elbadry *et al.* (1999) reported that inoculation with PGPR significantly increased plant dry weight and number of productive tillers when compared to the non-inoculated control. Hence, use of PGPR help in improving the major yield attributes of rice. Sathiya and Ramesh (2009) reported that various nitrogen management practices showed significant difference on yield attributes of aerobic rice. Grain and straw yields were significantly higher with cyanobacterial inoculants and RDN over N control. Treatment

BGA+PGPR and 2/3 nitrogen (fertilizer) showed highest grain (4.69 t ha⁻¹) and straw yield (8.56 t ha⁻¹). But this treatment was statistically at par with other three treatment including RDN, Compost application (1/3 N) + 2/3 N (fertilizer) and Compost based BGA mixture inoculant + 2/3 N (fertilizer).

Enhancement of rice seed germination, root and shoot growth, weight of rice grains and their protein content and the fertilizing action of N₂-fixing cyanobacteria has been generally attributed to the release of synthesized nitrogenous compounds either by decomposition of the cells or excretion (Nayak *et al.*, 2004). Wetland rice field can provides an ideal condition for the growth of cyanobacteria, which accumulate 19-38 kg N ha⁻¹ per crop and reduce the use of urea fertilizer in rice culture by 25-33 % (Hashem, 2001). The use of bio-fertilizer and bioenhancer such as N₂ fixing bacteria and beneficial micro-organism can reduce chemical fertilizer applications and consequently lower production cost. Utilization of PGPR in order to increase the productivity may be a viable alternative to organic fertilizers which also helps in reducing the pollution and preserving the environment in the spirit of an ecological agriculture (Stefan *et al.*, 2008).

Lucy *et al.* (2004) reviewed reports on effects of PGPR from large number of publications around the world and concluded that results from different experiment were not harmonized and were often inconsistent and up to 50-70 % yield increases were reported. Omar *et al.* (1989) reported 15-20% increase in rice yield due to the application of PGPR while Kloepper *et al.* (1989) found 4.9 to 15.5% increase in yield. In the same report he reported 3 to 160% increased rice yield due to the application of *Pseudomonas sp.* in field experiment. Imtiyaj *et al.* (2011) found increased grain and straw yield of rice due to the application of bio-fertilizers like BGA and *Azolla*. Alam *et al.* (2001) also reported increase in rice dry matter and yield and N accumulation by 6 to 24% and concluded that yield increase was due to increased root length, leaf area and chlorophyll content. Tran Van *et al.* (2000) observed the rice grain yield by 13 to 22% due to application of PGPR. Several studies (Garcia and Dobereiner, 1996; Smith *et al.*, 1984) showed that an inappropriate combination of bacteria and crop plant often resulted in a negative effect on the nitrogen accumulation and growth of the host plant. Also, a number of experiments showed that the extent of the positive effect of the bacteria on nitrogen accumulation and crop growth varied with the species or variety of the host plant (Bouton and Brooks, 1982; Chanway *et al.*, 1988). Hence, bacterial inoculation may not always result in persistent response, because of varying ecological factors and environmental conditions (Lynch, 1990).

Mehnaz *et al.* (1998) concluded from a field trial that inoculation with PGPR significantly increased the grain yield (11.7%) of rice over non-inoculated control. Whereas, Sherchand, (2000) found 15-20% increase in rice yield due to the inoculation of bacterial PGPR over non-inoculated control. The increased straw yield of rice due to inoculation of

PGPR, BGA and compost could be attributed to the higher supply of N and other micronutrients into soil (Bisht *et al.*, 2006; Khan *et al.*, 2008). The increased availability of Fe and other micronutrients in soil with inoculation of PGPR, BGA and compost in rice was responsible for higher yields as compared control plots (Nayyar and Chhibba, 2000). When compost was added to the clay soil along with SRI practices the *Azospirillum* count rose to $1400 \times 10^3 \text{ t}^{-1}$ and yield to 10.5 t ha^{-1} (Randriamiharisoa, 2001). Hence, the variation in partitioning of photosynthates in grain and vegetative organs of different treatments possibly caused a significant variation in harvest index (HI). The similar results have also been reported by several research workers (Imtiaz *et al.*, 2003; Ozkutlu *et al.*, 2006). Tran Van *et al.* (2000) reported increased shoot weight of rice due to the inoculation of PGPR. Cost of cultivation was found lower in all treatment of SRI method as compared conventional method. This might be due to low cost on nursery, seed and irrigation. Cost of cultivation was higher in the treatments having compost application compared to other treatments. Net return and B:C ratio in all the treatments were higher under SRI method compared to conventional method. BGA + PGPR + 2/3 N (fertilizer) under SRI method gave the highest net return (₹ 74, 596) followed by RDN(₹ 74556).

Concentrations and uptake of nitrogen, iron, zinc, manganese and copper in grain and straw of rice were not significantly influenced by the conventional and SRI method of planting. N concentrations in grain and straw were statistically at par in all treatments having cyanobacteria inoculation and RDN. However, N concentrations in cyanobacteria inoculated treatment and RDN were significantly higher than N control. BGA+PGPR+2/3 N (fertilizer) showed at par N, Fe, Zn, Mn and Cu uptake with RDN, compost application (1/3 N) + 2/3 N (fertilizer) and compost based BGA mixture inoculant + 2/3 N (fertilizer). PGPR are directly involved in increased concentration and uptake of nitrogen, synthesis of phytohormones, solubilization of minerals such as phosphorus, and production of siderophores that chelate iron and make it available to the plant root (Lalande *et al.*, 1989; Glick, 1995; Bowen and Rovira, 1999). It has also been reported that PGPR is able to solubilize inorganic and/or organic phosphates in soil (Liu *et al.*, 1992).

Chemical fertilizer is the most important input required for rice cultivation. Native and applied PGPR play an important role in better crop growth, yield, nutrient uptake and soil chemical and biological parameters in crop however their responses varied from 50 to 70 % increase in yield (Lucy *et al.*, 2004). The role of *Anabaena* in fixation of atmospheric N_2 is well established (Singh and Bisoyi, 1989; Nayak *et al.*, 2004) for rice crop. Therefore, increased availability of N in the rhizosphere resulted in its increased uptake due to *Anabaena* inoculation. Imtiyaj *et al.* (2011) reported significantly increased N, P and K concentration and uptake in grain and straw of rice due to the application of BGA, *Azolla* and other bio-fertilizers. Lin *et al.* (1983) reported that plant nutrient uptake can be enhanced by the

inoculation with free-living bacteria. K concentration in rice grain was reported to increase with increasing levels of N and influenced favourably by sources of N (Duhan *et al.*, 2001; Shivay *et al.*, 2001) and bacterial and cyanobacterial PGPR and BGA also increased the availability of nutrients especially N in soil.

PGPR facilitates plant growth and development, both directly and indirectly (Glick, 1995). Direct stimulation may include providing plants with fixed nitrogen, phyto-hormones or iron (that has been sequestered by bacterial siderophores) and solubilized phosphate, while indirect stimulation of plant growth includes preventing phyto-pathogens (bio-control) through production of antibiotics/ siderophores and hydrogen cyanide and thus promoting plant growth and development (Kloepper *et al.*, 1993). Under integrated nutrient management (INM) under the presence of combined nitrogen the BGA not only continue to grow but show concentration dependent increase in the uptake of added chemical nitrogen. This metabolized nitrogen which is prevented from being lost through denitrification, percolation and run off, is slowly released through excretion and autolysis (Suseela and Goyal, 1995; Singh and Mandal, 2000) and thus help in better crop growth, yield and higher nutrient uptake. Singh *et al.* (2011) and Imtiyaj *et al.* (2011) also found significantly increased N, P, K, Fe, Zn, Mn and Cu concentration and uptake in grain and straw of rice due to the application of BGA, *Azolla* and other bio-fertilizers.

Status of available N, P, K and Fe in soil at crop harvest stage was at par in conventional and SRI method of planting. Suryavanshi, (2011) also did not find any significant change in available N and available Fe under conventional and SRI planting. Treatment with compost application (1/3 N) + 2/3 N (fertilizer) recorded the highest value of available N followed by compost based BGA mixture + 2/3 N (fertilizer). Since phosphatic and potassic fertilizers were applied in equal quantity in all treatment including treatment containing no nitrogen, there was no significant difference in available P and K contents.

The principle in determination of Soil ARA is that cyanobacteria have ability to reduce acetylene to ethylene. ARA activity both at 60 DAT and at crop maturity in both conventional and SRI method was statistically at par. Treatment containing BGA+PGPR and 2/3 nitrogen through chemical fertilizer showed highest ARA activity. Lowest ARA was with N₀ 60 DAT and at crop maturity. Addition of N fertilizer is known to have a considerable influence on ARA (Singh and Bisoyi, 1989) and higher N levels are known to inhibit N fixation by BGA (Mekonnen *et al.*, 2002; Prasanna *et al.*, 2003). Nayak *et al.* (2004) found maximum ARA in N₃₀ treatments at 45 DAT while at 90 DAT the maximum values were recorded in plots receiving the N₉₀ level of urea application, indicative of tolerance of BGA/*Azolla* up to 90 kg N ha⁻¹. This study demonstrated that the fertility of rice fields depends not only on surface colonizing/floating BGA and *Azolla* but also on the direct and indirect interactions of BGA with the microflora in the below-surface soil environment.

Soil chlorophyll content is the measure of growth of cyanobacteria in soil. Soil chlorophyll content was higher at 60 DAT and decreased to ward crop maturity in all the treatments. Treatment BGA+PGPR and 2/3 nitrogen (fertilizer) showed highest chlorophyll content. Effectiveness of BGA in enhancing soil chlorophyll content was also increased due to the application of compost with BGA. But, Nayak *et al.* (2004) did not find any significant effect on soil chlorophyll content due to different nitrogen levels. However, at 45 DAT the overall effect of bio-fertilizers (BGA + *Azolla*, and *Azolla* alone) on soil chlorophyll content was significantly higher than the control or application of BGA bio-fertilizers alone, although individual bio-fertilizers and nitrogenous fertilizers did not exhibit any significant differences. The N₆₀ level of N fertilization and control plots (no bio-fertilizers) exhibited the highest soil chlorophyll at 90 DAT. Soil cores from N₉₀ + BGA + *Azolla* and N₁₂₀ + BGA and BGA + *Azolla* exhibited increased soil chlorophyll over the control. This indicated that inoculated BGA and *Azolla* bio-fertilizers bring about an enhancement in biomass, in terms of soil chlorophyll, in contrast with numerous reports available on the growth inhibition of BGA at high levels of N fertilization (Mekonnen *et al.*, 2002).

Soil dehydrogenase activity was higher at 60 DAT and decreased at crop harvest stage. BGA+PGPR and 2/3 nitrogen through chemical fertilizer showed highest dehydrogenase activity after 60 days of crop growth. Dehydrogenase is a respiratory chain enzyme of soil microorganisms, which is an indicator of biological redox system and microbial activity. Dehydrogenase activity was significantly higher in case of SRI as compared to conventional transplanting (Suryavanshi, 2011). Microbial biomass and soil enzymes are considered as potential indicator of soil quality due to their relationship to soil biology, ease of measurement, rapid responses to changes in soil management and high sensitivity to changes originated by management and environment factor (Karlen *et al.*, 1997; Marx., 2001; Jimenez *et al.*, 2002). The rationale for the use of microbial and biochemical characteristics as soil quality indicators is their central role in cycling of C and N and their sensitivity to change (Nannipieri *et al.*, 1990). Application of green manures *S. aculeata* significantly increased the activity of urease, dehydrogenase, amidase and phosphatase over prilled urea (50-150 kg N ha⁻¹) and control (Sriramachandrasckharan, 2002). The increase in dehydrogenase and alkaline phosphatase activities with increasing dose of fertilizer as well as organic amendments is reflections of organic matter build up by incorporation of green manuring and which led to increase in microbial activities (Tarafdar *et al.*, 1989; Pascual *et al.*, 2002). Soil microbial biomass C is a small component of soil organic matter and which is directly related with soil organic matter status (Dhull *et al.*, 2004; Jedidi *et al.*, 2004). Fertilizer application particularly N and increasing inputs of organic residue increase the soil organic matter and microbial biomass carbon (Graham *et al.*, 2002).

Slightly higher value of microbial biomass carbon (MBC) was observed in SRI method compared to conventional transplanting. This might be due to better soil condition of soil for microbes. Suryavanshi, (2011) recorded higher MBC in soil under SRI compared to conventional transplanting of rice. Rupela *et al.* (2006) reported that MBC was higher in SRI plots than those of conventional flood rice. Microbial activity in soil is dictated by availability of nutrients in an environment and the increased activity reflects the increased availability of nutrients. Rice planting methods did not influence soil chlorophyll content significantly. This may be due to a greater population of cyanobacteria, commonly found as floating assemblages in flooded rice fields (similar to CT). Treatment containing BGA+PGPR and 2/3 nitrogen through chemical fertilizer showed highest MBC after 60 days of crop growth. Treatment having no nitrogen recorded lowest MBC value. Soil microbial biomass is fundamental to maintaining soil functions as it represents the main source of soil enzymes that regulate transformation processes of elements in soils. It also controls the build-up and breakdown of organic matter, the decomposition of organic residues, and serves as early indicator of changes in soil management.

Cyanobacteria have been widely employed as inoculants for enhancing soil fertility and improving soil structure, besides increasing crop yields, especially in rice crop (Nayak *et al.*, 2004; Prasanna *et al.*, 2009a, b). However, the role of cyanobacteria under different crop establishment modes has not been evaluated. The SRI methodology may be favorable for the growth of these organisms, as aerobic environment, followed by micro-aerobic environment may prevail alternatively. Analyses of cyanobacteria from several rice based agro-ecologies of India have revealed that ecology/ecotype of the region or carbon/moisture levels does not alter the ratio of the dominant cyanobacterial members (Nayak *et al.*, 2001; Prasanna and Nayak, 2007); However, their abundance and diversity can be governed by the rice varieties (Prasanna *et al.*, 2009a). The size and activity of the soil microbial biomass depend on quantity and quality of soil organic matter (Weigand *et al.*, 1995) and is related to soil properties (Kaiser *et al.*, 1992), climatic conditions (Grisi *et al.*, 1998) and crop management (Kandeler *et al.*, 1999; Nieder *et al.*, 2008).

Among the three fractions of carbon viz., very labile, labile and less labile, very labile carbon fraction was marginally higher (0.78%) in SRI method of cultivation compared to conventional method at 0-7.5 cm depth. This subtle difference might be due to higher microbial activity and better soil physical condition in top 0-7.5 cm depth under SRI compared to conventional method of cultivation. Very labile fraction of C content between these methods was also on par at 7.5-15 cm layer. Treatments with compost (1/3 N) + 2/3 N (fertilizer) showed highest C content (0.83%) after 60 DAT. Carbon content in other treatments including RDN, BGA+PGPR + 2/3 nitrogen (fertilizer) and compost based BGA mixture inoculant + 2/3 N (fertilizer) (0.80%) were statistically at par. Labile fraction of C

was non-significant between the method of cultivation at both the depth. Cyanobacterial treatment showed no significant difference in labile C fraction between cyanobacterial inoculant treatment with RDN (fertilizer). Treatment receiving no N, recorded the least content of all C fractions at both the soil depths. Carbon content was higher in 0-7.5 cm depth compared to 7.5-15 cm depth. Very labile C was greater compared to labile or less labile C fractions.

Positive effect of manure application on SOC build-up under maize-wheat system in semi-arid India has earlier been reported by Benbi *et al.* (1998). Barreto *et al.* (2011) also showed that labile C fraction constitutes a larger fraction, 54–59%, of the TOC in soils under agroforestry systems in Brazil. The results suggested that the organic carbon in soils under maize-wheat and agroforestry systems is less stable and could be easily lost through organic matter decomposition if the current land-use is discontinued. On the contrary, soils under rice-wheat contained greater proportion (63%) of organic C in less labile and recalcitrant fractions. Our values are similar to those reported by Majumder *et al.* (2008) for rice based cropping systems in hot humid tropics of India, where in rice-berseem cropping system, the less labile and recalcitrant fractions together constituted about 55% of the total organic carbon. The greater proportion of organic C existing in less labile and recalcitrant forms in rice-wheat system may be attributed to the retarded rate of C oxidation in these soils that are flooded for 3 months during the rice season (Jenkinson, 1988; Watanabe, 1984). As a result of slower decomposition of C substrates in anaerobic soils than in aerated soils, plant molecules that are more resistant to microbial degradation such as lignins might gradually accumulate in the organic matter of paddy soils (Colberg, 1988; Tate, 1979). Several studies provided indirect evidence for the accumulation of lignin-derived substances in submerged soils (Ye and Wen, 1991). Slower lignin decomposition in submerged soils leads to incorporation of phenolic moieties into young SOM fractions (Olk *et al.*, 1996), imparting considerable recalcitrance to soil organic matter. While continuous rice-wheat cropping without balanced fertilization (N–P–K) caused a significant depletion of SOC, the same with balanced fertilization maintained the level and, in combination with organics, significantly improved it. A major fraction (~67%) of the C supplemented in the system through organic amendments (FYM, PS, and GM) was lost, however, and a small fraction was left to be stabilized into SOC (Majumdar *et al.*, 2008).

Soil aggregations at both the depths were statistically at par in both the methods of rice cultivation. In cyanobacterial inoculations it was found that at 0-7.5 cm depth treatment containing compost application (1/3 N) + 2/3 N (fertilizer) showed highest mean weight diameter (MWD) of aggregates as 1.12 mm after 60 DAT. Treatments with N₀ recorded lowest MWD of aggregates at both the soil depths. MWD were higher in 0-7.5 cm depth compared to 7.5-15 cm depth. This difference may be due to higher C content in top 7.5 cm

soil compared to 7.5-15 cm depth. Cyanobacteria are known to contribute to macro-aggregation and result in improved resistance to soil erosion, because as primary producers, they contribute to the enrichment of soil with soil organic matter (SOM) and to the improvement of biological activity (Acea *et al.*, 2003). Cyanobacteria are known to secrete extracellular polymeric substances (EPS) dominated by polysaccharides which can bind soil particles (Issa *et al.*, 2001). Prasanna *et al.* (2009) reported the significant contribution of cyanobacterial biomass in enhancing microbial activity and plant growth promotion, emphasizing their significance in sustainable management of rice ecosystem. Issa *et al.* (2007) observed improvement in the aggregation of cyanobacteria inoculated soils which could be related to increase in soil carbon and EPS that cause change in the soil micro-morphological characteristics of the aggregates. Maqubela *et al.* (2009) reported that inoculation of degraded soil with strain of *Nostoc* could improve the fertility and structural stability of degraded soil.

Conclusion

The growth and yield of rice and nutrient uptake were same in conventional and SRI planting method. Microbial, chemical and physical properties also were alike in both methods. However, net return and B: C ratio was higher in SRI method. Available nitrogen content at crop harvest was higher when treatment included compost and it was at par with recommended dose of N fertilizer. Microbial parameters like acetylene reductase activity (ARA), soil chlorophyll, dehydrogenase activity and microbial biomass carbon (MBC) content of soil were higher at 60 DAT compared to those at crop harvest. Treatment with BGA + PGPR + 2/3 N through chemical fertilizer showed highest ARA, soil chlorophyll and dehydrogenase activity at 60 DAT and at crop maturity. Application of compost (1/3 N) + 2/3 N through chemical fertilizer showed higher carbon content in all the three fractions, very labile, labile and less labile. Highest mean weight diameter (MWD) indicating better soil aggregation was recorded with compost application (1/3 N) + 2/3 N through c fertilizer. Treatment containing no (N₀) nitrogen recorded lowest carbon and mean weight diameter.

8. SUMMARY AND CONCLUSION

A field experiment was conducted at the Genetics D block of Indian Agricultural Research Institute, New Delhi, India during wet (*khariif*) season of 2011 to study the “**Influence of cyanobacteria based inoculants and cultivation methods on rice (*Oryza sativa* L.) yield, soil carbon content and aggregation**”. The salient findings of the experiment are summarized here.

- Conventional and SRI method of planting did not have significant effect on plant growth parameters, yield attributes and grain and straw yield of rice. Nutrient concentrations and uptake by grain and straw and soil chemical and microbial properties were same under both methods. Soil carbon and aggregation also remained unaffected due to the methods of planting.
- Inoculation of cyanobacterial inoculants and recommended dose of nitrogen (RDN) showed significantly higher plant height, tillers, dry matter accumulation and LAI over N control.
- Yield attributes like panicle m^{-2} , weight and no. of grains panicle $^{-1}$ were found significantly higher with cyanobacterial inoculants and RDN over N control. The treatment BGA+PGPR and 2/3 nitrogen through fertilizer had highest values.
- Highest grain and straw yield was recorded with BGA+PGPR and 2/3 nitrogen through fertilizer but these were statistically at par with recommended dose of N, compost application (1/3 N) + 2/3 N through fertilizer and compost based BGA inoculant + 2/3 N through fertilizer.
- Highest net return and B : C ratio was given by SRI method. Treatment BGA + PGPR + 2/3 N (through fertilizer) under SRI method gave the highest net return (₹ 74, 596 ha $^{-1}$) followed by the RDN through fertilizer (₹ 74556 ha $^{-1}$).
- Treatment with cyanobacterial inoculants + PGPR + 2/3 N as fertilizer showed highest concentration and uptake of N, Fe, Zn, Mn and Cu in grain and straw but these were statistically at par with concentration and uptake at RDN.
- Available nitrogen content of soil at crop harvest stage was highest with compost application (1/3 N) + 2/3 N through fertilizer followed by compost based BGA inoculant + 2/3 N through fertilizer and RDN. However available P, K and Fe in soil at crop harvest were statistically at par in all the treatments.
- Microbial parameters including acetylene reductase activity (ARA), soil chlorophyll, dehydrogenase activity and microbial biomass carbon (MBC) content of soil were higher at 60 days after transplanting (DAT) compared to those at crop harvest.

- Treatment having BGA + PGPR + 2/3 N through chemical fertilizer showed higher values of the microbial parameter at both 60 DAT and at harvest.
- Application of compost (1/3 N) + 2/3 N through chemical fertilizer showed higher carbon content in all the three fractions i.e., very labile, labile and less labile. Very labile C at 0-7.5 cm depth was significantly lower in *Multani mitti* based BGA inoculant + 2/3 N through chemical fertilizer and biofilm based BGA bio-fertilizer + 2/3 N through chemical fertilizer compared to other cyanobacterial inoculants and RDF.
- Treatment containing no nitrogen (N₀) recorded lowest C in all fractions at both 0-7.5 cm and 7.5-15 cm depths.
- Highest mean weight diameter (MWD) was recorded in treatment having compost application (1/3 N) + 2/3 N through chemical fertilizer, which was significantly higher than *Multani mitti* based BGA inoculant + 2/3 N through chemical fertilizer and biofilm based BGA bio-fertilizer + 2/3 N through chemical fertilizer at 0-7.5 cm depth.

Conclusions

- ❖ Cyanobacterial inoculants can be used to replace approximately one-third (40 kg ha⁻¹) of recommended dose of N fertilizer. Combination of PGPR with BGA and 2/3 dose of chemical N fertilizer gave highest grain yield and net return so this treatment can be used in integrated nutrient management of rice.
- ❖ Microbial parameters like acetylene reductase activity (ARA), soil chlorophyll, dehydrogenase activity and microbial biomass carbon (MBC) content of soil were higher with BGA + PGPR + 2/3 N (fertilizer) at 60 DAT and at crop maturity.
- ❖ Higher carbon content in all the three fractions, very labile, labile and less labile was found with application of compost (1/3 N) + 2/3 N through fertilizer. Highest mean weight diameter (MWD) indicating better soil aggregation was recorded with compost application (1/3 N) + 2/3 N through fertilizer.
- ❖ Conventional and SRI method of planting were same in all the parameters studied on plant growth, yield and nutrient uptake by crop, chemical, microbial and physical properties of soil. However, net return and B: C ratio was higher in SRI method.
- ❖ It was recommended that BGA+PGPR and 2/3 N from fertilizer can be applied to contribute 1/3 N (40 kg ha⁻¹) in integrated rice nutrition and for getting better crop growth, yield and return. With this treatment physical, chemical and microbial quality of soil and nutrient concentration in grain can be improved. Both the methods of planting were same in most of the parameters studied so, SRI should be preferred due to eminent water saving in this method.

Future scope of research

- ❖ The experiment on plant cyabobacteria based growth promoting rhizobacteria (PGPR) should be conducted for at least two years and their residual effects also may be studied on different crops.
- ❖ Field experiments may be conducted on biofilm and liquid biofertilizers.
- ❖ Interaction between PGPR, plant enzymes and nutrients should be studied in different cropping systems.

**धान (ओरायज़ा सटायवा एल.) की उपज, मृदा-कार्बन अंश एवं समुच्चयन पर सयानोबक्टेरिया-
आधारित संरोप्यो तथा सस्य विधियों का प्रभाव**

सारांश

सन २०११ की वर्षा (खरीफ) ऋतू के दौरान, धान (पूसा बासमती १४०१) की फसल में कृषि विधियों एवं सयानोबक्टेरिया संबंधी संरोप्यो का पादप वृद्धि, उपज, पोषकतत्व उद्ग्रहण, मृदा के रासायनिक एवं सूक्ष्मजीवी सम्बन्धी गुणों, मृदा कार्बन एवं समुच्चयन पर प्रभाव जात करने के लिए खेत में एक प्रयोग किया गया। यह प्रयोग विभक्त भूखंड डिजाईन में, चौदह उपचार संयोजन के साथ किया गया जिसमें फसल-स्थापना की दो विधियों का उपयोग किया गया- पारंपरिक रोपण (सी टी) विधि एवं धान सधानिकरण तंत्र (एस आर आय) तथा सात सयानोबक्टेरिया संबंधी संरोप्य यथा, एन. ; एन की संस्तुत खुराक (१२० की ग्रा/ हे) ; कम्पोस्ट अनुप्रयोग (१/३ एन) + २/३ एन ; बायोफिल्म एन एवं मुल्तानी मिटटी आधारित बी जी ए संरोप्य + २/३ एन ; २/३ एन (१८० की ग्रा / हे) एवं यूरिया के माध्यम से अनुप्रयुक्त एन की संस्तुत खुराक (१२० की ग्रा / हे) तथा ये उपचार तीन प्रतिकृतियों में किये गए। सी टी एवं एस आर आय में क्रमशः २१ एवं १२ दिन आयु की पौध की रोपाई की गयी। परिणामों ने दर्शाया की पारंपरिक एवं एस आर आय विधि का धान की पादप वृद्धि प्राचलों, उपज गुणों तथा दोनों एवं ऋसा की उपज पर कोई महत्वपूर्ण प्रभाव नहीं था। रोपाई की विधियों का पोषक तत्व सांद्रताओं तथा उनका दाने द्वारा उद्ग्रहण और मृदा के रासायनिक एवं सूक्ष्मजीव संबंधी गुणों पर कोई प्रभाव नहीं था। मृदा कार्बन एवं समुच्चय गुण भी रोपण विधियों से अप्रभावित रहे। बी जी ए + पि जी पि आर, एवं उर्वरक के माध्यम से २/३ नाइट्रोजन से उच्चतम पादप वृद्धि हुई की यह आर डी एन, कम्पोस्ट अनुप्रयोग (१/३ एन) + उर्वरक के माध्यम से २/३ एन तथा कम्पोस्ट आधारित बी जी ए मिश्रण + उर्वरक के माध्यम से २/३ एन के समकक्ष थी। ए कंट्रोल की तुलना में, बी जी ए + पी जी पी आर एवं २/३ नाइट्रोजन तथा आर डी एन के साथ उपज संबंधी गुणों यथा पुष्पगुच्छ एम^{-३}, प्रति पुष्पगुच्छ दानों की संख्या एवं भार तथा दाना एवं भूसा उपज महत्वपूर्ण रूप से अधिक पाए गए। वैसे २/३ एन सहित सयानोबक्टेरिया संरोप्यो के साथ तथा आर डी एन के बीच भिन्नता नगण्य थी। उपचार बी जी ए + पी जी पी आर एवं रासायनिक उर्वरक के साथ २/३ नाइट्रोजन ने अधिकतम उपज दर्शायी किन्तु यह नाइट्रोजन की संस्तुस मात्रा, कम्पोस्ट अनुप्रयोग (१/३ एन) + उर्वरक के माध्यम से २/३ एन तथा कम्पोस्ट आधारित बी जी ए मिश्रण संरोप्य + उर्वरक के माध्यम से २/३ एन के साथ संखिकीय रूप से समकक्ष थी। पारंपरिक विधि की तुलना में एस आर आय में उच्चतर शुद्ध लाभ एवं बी : सी अनुपात देखा गया। एस आर आय विधि के अंतर्गत बी जी ए + पी जी पी आर + २/३ एन (उर्वरक के माध्यम से) द्वारा उच्चतम शुद्ध लाभ (रु ७४,५९६ प्रति हे) हुई जिसके उर्वरक के माध्यम से संस्तुत एन की संस्तुत खुराक + उर्वरक के माध्यम से २/३ एन (रु ७४,५९६ प्रति हे) का स्थान रहा। बी जी ए + पी जी पी आर + उर्वरक के माध्यम से २/३ एन, आर डी एन, कम्पोस्ट अनुप्रयोग (१/३ एन) + उर्वरक द्वारा २/३ एन तथा कम्पोस्ट आधारित बी जी ए मिश्रण संरोप्य + उर्वरक द्वारा २/३ एन के अनुप्रयोग से दाने एवं भूसे में एन, एफड, जेडएन, एमएन, एवं सीयु की अधिक संद्रताये और अधिक उद्ग्रहण देखा गया। फसल की कटाई अवस्था

पर कम्पोस्ट अनुप्रयोग (१/३ एन) + रासायनिक उर्वरक के माध्यम से २/३ एन के साथ मृदा का उपलब्ध एन अंश अधिकतम था, इसके बाद कम्पोस्ट आधारित बी.जी.ए. संरोप्य + उर्वरक के माध्यमसे २/३ एँ का स्थान रहा ! वैसे, सभी उपचारों में, फसल की कटाई अवस्था पर उपलब्ध पी. के. एवं एफ इ. संखिकीय रूप से सामान थे! सूक्ष्मजीव सम्बन्धी सभी प्राचल तथा, ऐसिटिलिन रिडाक्टेज सक्रियता (ऐ. आर. ऐ.), मृदा हरीतावर्ण ,डीहायड्रोजिनेज सक्रियता तथा मृदा का सूक्ष्मजीव सम्बन्धी जैवमात्रा कार्बन (एम्. बी.सी.) अंश, रोपण के ६० दिन बाद, फसल कटाई के समय के मानो की तुलना में अधिक थे ! कटाई के समय तथा ६० डी. ए. टी. दोनों अवस्थाओं में, बी.जी.ए. + पी. जी. पी. आर. + रासायनिक उर्वरक के माध्यमसे २/३ एन ने सभी सूक्ष्मजीव सम्बन्धी प्राचलों हेतु उच्चतर मान दर्शाए! कम्पोस्ट अनुप्रयोग (१/३ एन) + उर्वरक के माध्यम से २/३ एन ने सभी तीन प्रभाजों अस्थिर तथा कम अस्थिर, में उच्चतम कार्बन अंश दर्शाया | मृदा की दोनों गहराइयों पर एन. से कार्बन के सभी प्रभाजों में कम अंश रेकॉर्ड किए गए | उच्चतम माध्य मार व्यास (एम् डब्लू डी) जो बेहतर मृदा समुच्चयन को दर्शाता है, 0 - ७.५ सेमी गहराई पर, कम्पोस्ट अनुप्रयोग (१/३ एन) + रासायनिक उर्वरक के माध्यम से २/३ एन उपचार में, मुल्तानी मिटटी आधारित बी जी ए संरोप्य + उर्वरक के माध्यम से २/३ एन तथा बायोफिल्म आधारित बी जी ए जैव उर्वरक + रासायनिक उर्वरक द्वारा २/३ एन की तुलना में महत्वपूर्ण रूप से अधिक पाया गया | इस अध्ययन से यह निष्कर्ष निकलता है कि धान कि फसल में बेहतर फसल वृद्धि, उपज एवं लाभ पाने के लिए बी जी ए + पी जी पी आर एवं उर्वरक से २/३ एन का अनुप्रयोग किया जा सकता है | इस उपचार से मृदा कि भौतिक, रासायनिक एवं सूक्ष्मजीव संबंधी गुणवत्ता में तथा दाने में पोषक तत्व सान्द्रता में सुधर होता है | अध्ययन किए गए अधिकांश प्राचलों में रोपण कि विधियाँ का कोई प्रभाव नहीं था इसलिए एस आर आई में जल कि बचत होने के कारण इस विधि को वरीयता दी जानी चाहिए |

Influence of cyanobacteria based inoculants and cultivation methods on rice (*Oryza sativa* L.) yield, soil carbon content and aggregation

ABSTRACT

A field experiment was undertaken during the rainy (*Khariif*) season of 2011 to investigate the influence of methods of rice (*Pusa Basmati* 1401) establishment and cyanobacterial inoculants on plant growth, yield, nutrient uptake, soil chemical and microbial properties, soil carbon and aggregation. The experiment was laid out in split plot design with fourteen treatments combinations, comprising two methods of crop establishment viz., conventional transplanting (CT) and system of rice intensification (SRI) and seven cyanobacterial inoculants viz., N₀; Recommended dose of N (120 kg ha⁻¹); Compost application (1/3 N) + 2/3 N; Biofilm based BGA biofertilizer + 2/3 N; BGA + PGPR + 2/3 N; Compost based BGA mixture inoculant + 2/3 N and *Multani mitti* based BGA inoculant + 2/3 N. 2/3 N (80 kg ha⁻¹) and recommended dose of N (120 kg ha⁻¹) was applied through urea and treatments were replicated thrice. In CT and SRI 21 and 12 days old seedlings, respectively were transplanted. Results showed that, conventional and SRI method did not have significant effect on plant growth parameters, yield attributes and grain and straw yield of rice. Nutrient concentrations and uptake by grain and straw and soil chemical and microbial properties were not influenced by methods of planting. Soil carbon and aggregation properties also remained unaffected due to the methods of planting. BGA+PGPR and 2/3 nitrogen through fertilizer showed highest plant growth but these were statistically at par with RDN, compost application (1/3 N) + 2/3 N through fertilizer and compost based BGA mixture inoculant + 2/3 N through fertilizer. Yield attributing characters like panicle m⁻², weight and no. of grains panicle⁻¹ and grain and straw yield were found significantly higher with BGA+PGPR and 2/3 nitrogen and RDN compared to N control. However, difference between the effect of cyanobacterial inoculants along with 2/3 N and RDN was non-significant. Treatment BGA+PGPR and 2/3 nitrogen through chemical fertilizer showed highest yields but these were statistically at par with recommended dose of N, compost application (1/3 N) + 2/3 N through fertilizer and compost based BGA mixture inoculant + 2/3 N through fertilizer. Higher net return and B : C ratio was observed in SRI compared to conventional method. Highest net return was obtained by BGA + PGPR + 2/3 N (through fertilizer) under SRI method (₹ 74, 596) followed by the recommended dose of N through fertilizer (₹ 74,556) + 2/3 N through fertilizer. BGA+PGPR+2/3 N through fertilizer, RDN, compost application (1/3 N) + 2/3 N through fertilizer and compost based BGA mixture inoculant + 2/3 N through fertilizer had higher concentrations and uptake of N, Fe, Zn, Mn and Cu in grain and straw. Available N content of soil at crop harvest stage showed that compost application (1/3 N) + 2/3 N through chemical fertilizer recorded the highest available

nitrogen content followed by compost based BGA mixture inoculant + 2/3 N through fertilizer. However available P, K and Fe in soil at crop harvest were statistically at par in all the treatments. All the microbial parameter viz., acetylene reductase activity (ARA), soil chlorophyll, dehydrogenase activity and microbial biomass carbon (MBC) content of soil were higher at 60 days after transplanting as compared to value at crop harvest. BGA + PGPR + 2/3 N through chemical fertilizer showed higher value for all the microbial parameter at both 60 DAT and at harvest. Compost application (1/3 N) + 2/3 N through chemical fertilizer showed higher carbon content in all the three fraction viz., very labile, labile and less labile. No nitrogen (N_0) recorded lower value in all fraction of carbon at both soil depths. Highest mean weight diameter (MWD) indicating for better soil aggregation was observed in compost application (1/3 N) + 2/3 N through chemical fertilizer which was significantly higher than *Multani mitti* based BGA inoculant + 2/3 N through chemical fertilizer and biofilm based BGA bio-fertilizer 2/3 N through chemical fertilizer at 0-7.5 cm depth. It was concluded that BGA+PGPR and 2/3 N from fertilizer can be applied in rice for getting better crop growth, yield and return. With this treatment physical, chemical and microbial quality of soil and nutrient concentration in grain can be improved. Both the methods of planting were same in most of the parameters studied so; SRI should be preferred due to eminent water saving in this method.

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11. Appendix

Appendix I- Weather conditions (June-October, 2011)

Date	Max temp(°C)	Mini temp(°C)	RH1 (%)	RH2 (%)	Rainfall (mm)	Sunshine (hrs)	Evaporation (mm)	Wind velocity
01.06.11	41.3	24.6	78	40	0	5.8	8.7	12.4
02.06.11	36	21.9	64	36	0	9.1	7.6	5.7
03.06.11	36	22.3	53	33	0	5.1	7.2	5.5
04.06.11	39.2	23.2	60	26	0	9.2	7.2	4.7
05.06.11	40.7	23.4	52	29	0	9.9	7.3	2.9
06.06.11	42.1	27.5	53	30	0	8.8	6.9	1.1
07.06.11	43.2	29	55	33	0	8	9.2	3.63
08.06.11	43.6	26.9	65	45	0	7.8	9.8	9.8
09.06.11	37	27.2	61	51	0	9	9.8	11.8
10.06.11	36.2	19.6	84	61	10.4	7	5.1	13.2
11.06.11	32.5	26	64	44	0	6.4	5.4	5.8
12.06.11	40.5	28.3	72	54	0	6	9	8
13.06.11	35.7	26	75	45	0	7.9	7.2	8.6
14.06.11	39.2	27.4	65	36	0	5.4	6.2	3.2
15.06.11	42.2	28.2	68	76	36.2	7.7	3.2	8.3
16.06.11	33	23.4	81	62	0	4.7	4.8	7.8
17.06.11	35.5	26	86	65	0	2.6	5.8	7.1
18.06.11	36.5	27.3	90	61	4	3	4.6	6.4
19.06.11	36.2	24.7	95	61	7.6	6.1	5.2	5.5
20.06.11	38	23.2	77	58	0	7.5	4	4.2
21.06.11	36.2	25.8	75	48	0	6.2	5	3.7
22.06.11	38	27.1	74	53	0	3.4	5	3.4
23.06.11	37.4	28	72	56	0	3.8	5.2	4.7
24.06.11	37.8	29	76	49	0	8.2	6	7.6
25.06.11	38.2	28.7	92	62	1.2	6.2	5.2	3.5
26.06.11	35	27.7	98	89	26.8	0	4.8	6.2
27.06.11	30.5	24.5	92	79	7	0	2.5	3.5
28.06.11	32.8	25.8	94	73	9.6	1	3.1	2.3
29.06.11	32.3	26	81	90	0	3.4	1.7	4.8
30.06.11	31.1	26.2	89	66	1.4	1.4	2.6	3.1
01.07.11	34.3	25	82	61	0	3.8	3.8	6.4
02.07.11	35.6	25.7	80	75	0	1.6	4.5	8.1
03.07.11	36.1	26.4	68	46	0	3	4.3	7.1
04.07.11	38.1	26.9	68	44	0	6.7	5	9.5
05.07.11	38	28.1	69	68	0	3.8	5.1	6
06.07.11	37.2	26.7	92	83	0	1.7	3.7	4.6
07.07.11	33.5	27	85	60	0	1.1	2.5	5.8
08.07.11	35.5	26.5	81	78	0	3.4	4	7.9
09.07.11	30	25.4	95	92	0.2	0	2.6	4

10.07.11	31.5	24.6	94	83	10	1.3	2	4.1
11.07.11	32	25.3	91	61	0	2.3	2.5	3.6
12.07.11	35.8	26.4	97	67	0	9.5	3.9	3.5
13.07.11	35.6	27	88	67	0	2.7	3.5	3.6
14.07.11	35.5	26.9	85	79	0	3.9	3.4	4.2
15.07.11	34	25.6	87	85	0	1.9	3.2	5.9
16.07.11	30.2	24.4	92	69	15.4	0	2.9	6.1
17.07.11	33.2	24.8	91	67	0	2.2	3.9	2.9
18.07.11	33	25.8	90	77	2.8	5	4.8	4.7
19.07.11	31.5	25.4	94	58	0	3	3.3	4
20.07.11	35	27.2	85	50	0	4.9	4.5	2.1
21.07.11	37.5	26	87	66	0	2.4	4	6.2
22.07.11	34.2	25.7	92	58	0	0.3	3.5	3.1
23.07.11	34.5	27.2	88	63	0	0	3.7	3.3
24.07.11	33.2	25.1	90	61	4.9	0.8	5.1	8.1
25.07.11	33.6	25	83	68	0	8.2	3.5	9.4
26.07.11	33.5	27	80	89	0	2	3.9	4.5
27.07.11	34.3	26.2	88	83	3	2	3	1.8
28.07.11	33.7	25	95	75	2.5	0.5	3.3	3.6
29.07.11	33.5	25.4	94	64	5	0.1	2.6	3.1
30.07.11	34.4	26.3	92	66	0	1.9	4.5	0
31.07.11	35.4	26.4	88	89	0	0.2	5	0
01.08.11	37.4	27	80	58	0	6	6	2.3
02.08.11	36.8	27.8	81	56	0	5.8	5.6	0.2
03.08.11	38.2	27.4	66	60	0	7.7	5.6	0.1
04.08.11	34.4	27	58	80	0	3.6	4.2	0
05.08.11	33	26.3	88	62	21	0	2	0
06.08.11	34	25.8	94	80	0.5	0.9	3	1.5
07.08.11	31.6	26.3	92	65	0	0	2.2	0.2
08.08.11	34	26.9	88	65	0	4.2	3.6	0.3
09.08.11	34.4	27.2	83	65	0	6.7	3.5	3.3
10.08.11	34.2	24.2	92	79	1.8	6.2	4.2	2.7
11.08.11	31.5	25.8	91	91	4	3.2	3	0.9
12.08.11	31	24.4	98	89	46.4	2.2	2.6	0
13.08.11	30	24.6	89	73	10.2	2.7	3.9	0.5
14.08.11	33.5	24.7	85	73	0	6.6	3.7	0.8
15.08.11	31.5	24.2	98	97	31.7	3.5	3.9	2.4
16.08.11	28.5	25.2	97	87	12.6	0	3.9	0.3
17.08.11	30	24.4	95	67	0	0	1.6	0
18.08.11	32	24.2	86	59	0	7.2	3	0
19.08.11	32.6	24	78	81	0	8.4	3.7	0
20.08.11	35.4	24.2	90	65	59.1	1	2.1	0.6
21.08.11	30.5	26.2	83	59	0	7.9	3.3	0.4
22.08.11	32.5	26	84	57	0	8.9	3.5	0
23.08.11	34.5	27	81	91	0	4.1	4	0

24.08.11	35.2	24	93	72	23.2	0	1.9	2.7
25.08.11	33.2	24.8	94	72	14.4	1.3	2.2	0.7
26.08.11	31.5	25.2	91	81	0	4.6	3.7	0
27.08.11	34	25.8	91	75	0	0	3	2.9
28.08.11	32.2	25.7	79	60	0	0.3	3.1	3.2
29.08.11	31.6	25.8	83	67	0	8.8	4.2	2
30.08.11	34.8	26.8	84	65	0	9.9	5.2	3
31.08.11	34.5	26	88	62	2.4	7.9	4.9	2.6
01.09.11	35	27.6	91	66	0	10.1	5.4	2.5
02.09.11	35.5	27.7	83	64	0	9.3	5.4	4.3
03.09.11	35	24	90	77	7.2	7.7	4.2	3.5
04.09.11	33	24.4	98	88	29.2	2.2	3.7	3.8
05.09.11	30.6	24	98	84	14.6	0	1.6	1.7
06.09.11	31.6	23.8	97	92	18.4	0	2	2.3
07.09.11	31.2	25.8	92	97	4.8	1.1	1.8	2.1
08.09.11	32.5	24.9	97	68	16.6	3.7	1.6	1.2
09.09.11	33.4	24.4	97	94	23	6.2	1.9	1.2
10.09.11	29.2	24.6	97	74	8.2	0	1.9	0.6
11.09.11	33.4	25.8	92	66	0	2.7	2.2	1.3
12.09.11	33.3	26	95	80	0	8.1	2.5	1.5
13.09.11	33.5	26.1	88	67	0	5.6	3.4	1.9
14.09.11	32	26	86	63	0	8.8	4.9	1.1
15.09.11	32.6	25.8	95	88	5.4	7.3	3.8	2.5
16.09.11	32.5	24.2	97	81	24.6	3.8	3.6	0
17.09.11	33.5	23.4	92	69	11.6	4.4	2.2	1.8
18.09.11	33	24.3	92	73	0	8.6	3	6.9
19.09.11	33.6	24.4	89	73	0	7.2	3.4	5.5
20.09.11	33.5	24	90	68	0	8.1	3.5	9.6
21.09.11	33.2	24.3	92	62	0	9	3.7	9.2
22.09.11	34	23	90	67	0	8.4	3.2	6.4
23.09.11	34.2	24.8	90	72	0	8.8	3.8	7.9
24.09.11	35.2	22.4	82	51	0	9.6	3.4	9.2
25.09.11	34.5	21.4	84	54	0	9.4	3.9	5.4
26.09.11	34.2	23.4	80	55	0	9.4	4.1	7.1
27.09.11	35	23.4	76	58	0	8.6	5.2	10.3
28.09.11	34.6	23.8	81	57	0	8.7	5.1	9.6
29.09.11	34.8	22.6	84	47	0	9.2	6.2	10.4
30.09.11	34.7	22	84	55	0	9.5	6.3	9.1
01.10.11	34.5	22.4	80	54	0	9.6	6	9.7
02.10.11	34.2	22	71	64	0	9.2	5.6	9.9
03.10.11	34.5	22.8	84	41	0	9.2	4.5	8.1
04.10.11	34.5	21.7	73	47	0	8.7	5.5	6.9
05.10.11	34.5	21	83	44	0	8.4	4.2	4.7
06.10.11	33.6	20.4	87	48	0	9	4.5	4.2

07.10.11	34.2	20.8	85	45	0	8.6	5	4.9
08.10.11	34.3	21.4	86	50	0	8.4	6.1	5.1
09.10.11	34.4	20.4	95	53	0	7.1	5.2	0.2
10.10.11	33.6	18.4	96	55	0	7.3	4.9	8.7
11.10.11	33.6	19.6	88	32	0	7.8	5	4.8
12.10.11	34	20.8	80	33	0	8.2	3.8	4.4
13.10.11	34	18.2	74	28	0	8.4	4.8	5.4
14.10.11	34.9	19.6	69	28	0	6.8	5	4.5
15.10.11	34	18.2	74	26	0	8	5	6.1
16.10.11	34	15.7	80	91	0	9.5	5.5	4.4
17.10.11	34	15.5	77	27	0	9.5	4	2.7
18.10.11	32.5	15.2	82	27	0	8.3	3.5	2.3
19.10.11	31.9	15	81	24	0	7	4	4
20.10.11	32.8	15	90	35	0	8.7	4.1	3
21.10.11	32.2	16.2	81	38	0	3.3	3.6	0.6
22.10.11	31	15.9	86	29	0	0.5	2.6	0.5
23.10.11	32.5	15.4	84	33	0	4	2.9	1.9
24.10.11	33	16.4	88	34	0	6.9	3.7	2.5
25.10.11	31.8	14.6	82	34	0	4.4	4	1.7
26.10.11	30.8	14.4	89	34	0	7.8	3.6	1.5
27.10.11	30.4	14.3	72	25	0	3.5	4	1.9
28.10.11	29.8	11.9	84	29	0	7.5	3.8	4
29.10.11	29.6	12.6	85	35	0	7	3.8	2.4
30.10.11	29.4	14.8	85	31	0	5.8	3.6	1.5
31.10.11	30.4	13	89	29	0	3.5	3.5	2.1

Appendix II- Cost of cultivation in Conventional and SRI method of rice cultivation

Particular	Unit	Rate (₹)	Conventional method	SRI method
			Cost (₹)	Cost (₹)
Nursery raising				
ploughing of field	2 tractor hr	175	350	328.125
puddling	3 tractor hr	175	350	913.5
making beds and sowing	10 man-day	203	2030	1091.125
nitrogen dressing	40kg N	11.5	460	1237.5
weeding	6 Man -day	203	1218	735.875
Irrigation	3	450	1350	281.25
cost of irrigation	4 Man -day	203	812	328.125
rental value of land	1 month	500	500	913.5
Main field				
Discing	4 tractor hr	175	700	634.375
ploughing by cultivar	4 tractor hr	175	700	634.375
puddling	6 tractor hr	175	1050	940.625
bundling	8 Man -day	203	1624	1446.375
seedling uprooting and transplanting	30 Man - day	203	6090	5354.125
weeding	12 Man - day	203	2436	5354.125
gap filling	5 Man -day	203	1015	1801.625
fertilizer				
P ₂ O ₅	60 kg	21.25	1275	1118.281
K ₂ O	60kg	8.5	510	447.3125
N	80/ 120 kg	11.5	854.2857	748.9375
special cost of cultivation				
seed	35kg	55	1925	295.625
irrigation	20	400	8000	3900
biofertilizer	2 kg	80	80	80
compost	8000 kg	1	8000	4000.25
seed treatment(Bavistin)	90g		200	200
Harvesting	20 Man - day	203	4060	3577.875
Processing	10 Man - day	203	2030	1801.625
Rental value on land	4 months		2000	2000
Total cost			42727.86	40126.86

Appendix III
Summarized analysis of variance (ANOVA) of the data presented in the thesis manuscript

ANOVA TABLES

Table 1.1 Mean sum of square (MSS) for plant height

Source	d.f.	30 DAT	60 DAT	90 DAT	At Harvest
Replication	2	2.391	12.866	13.380	0.080
Main plot	1	88.015	18.667	55.315	23.475
Error (a)	2	1.675	3.167	11.007	11.341
Sub-plot treatment	6	34.806	224.937	432.095	470.653
Main plot x Sub-plot	6	41.784	74.798	170.402	61.020
Error (b)	24	2.407	5.456	11.003	13.947
	41.00				
CV % Main plot		5.55	4.42	6.52	6.46
Sub-plot treatment		6.65	5.80	6.52	7.16

Table 1.2 Mean sum of square (MSS) for number of tillers

Source	d.f.	30 DAT	60 DAT	90 DAT	At Harvest
Replication	2	150.756	292.667	55.024	73.654
Main plot	1	1862.669	242.881	160.095	120.701
Error (a)	2	55.322	94.952	238.024	64.178
Sub-plot treatment	6	4592.761	7716.992	11129.159	13790.212
Main plot x Sub-plot	6	57.374	441.770	239.317	253.701
Error (b)	24	70.406	198.976	159.440	96.505
	41.00				
CV % Main plot		7.48	5.58	8.20	4.53
Sub-plot treatment		8.44	8.08	6.71	5.56

Table 1.3 Mean sum of square (MSS) for shoot dry matter accumulation m⁻².

Source	d.f.	30 DAT	60 DAT	90 DAT
Replication	2	23.496	197.859	91.731
Main plot	1	1105.238	262.550	38.864
Error (a)	2	2.163	70.249	89.509
Sub-plot treatment	6	229.071	4956.380	65922.408
Main plot x Sub-plot	6	75.073	275.274	2022.532
Error (b)	24	5.915	152.187	153.765
	41.00			
CV % Main plot		1.96	5.93	2.66
Sub-plot treatment		3.24	8.73	3.49

Table 1.4 Mean sum of square (MSS) for leaf area index (LAI)

Source	d.f.	30 DAT	60 DAT	90 DAT
Replication	2	0.093	0.064	0.029
Main plot	1	0.860	0.006	0.012
Error (a)	2	0.011	0.022	0.013
Sub-plot treatment	6	1.291	3.189	3.612
Main plot x Sub-plot	6	0.182	0.864	1.776
Error (b)	24	0.027	0.045	0.027
	41.00			
CV % Main plot		9.7	6.16	6.96
Sub-plot treatment		15.51	8.86	9.82

Table 1.5 Mean sum of square (MSS) for yield attributing characters

Source	d.f.	Panicle m ⁻²	Weight of panicle	Length of panicle	Grains panicle ⁻¹	Test weight
Replication	2	73.654	0.019	1.867	33.225	0.123
Main plot	1	120.701	0.002	0.013	10.575	0.012
Error (a)	2	64.178	0.011	0.128	10.425	0.241
Sub-plot treatment	6	13790.212	1.253	10.401	1463.551	0.143
Main plot x Sub-plot	6	253.701	0.113	6.363	655.157	0.170
Error (b)	24	96.505	0.028	0.382	61.571	0.641
	41.00					
CV % Main plot		4.53	7.14	2.49	4.08	4.58
Sub-plot treatment		5.56	12.17	4.30	9.91	7.47

Table 1.6 Mean sum of square (MSS) for grain and straw yield and harvest index

Source	d.f.	Grain yield	Straw yield	Harvest index
Replication	2	0.023	0.590	0.00074
Main plot	1	0.011	0.101	0.00001
Error (a)	2	0.005	0.069	0.00012
Sub-plot treatment	6	1.759	2.019	0.00141
Main plot x Sub-plot	6	0.325	2.457	0.00182
Error (b)	24	0.029	0.088	0.00019
	41.00			
CV % Main plot		3.19	6.46	6.37
Sub-plot treatment		7.91	7.29	8.04

Table 1.7 Mean sum of square (MSS) for nitrogen content, protein content and uptake of nitrogen in grain and straw of rice

Source	d.f.	N in grain (%)	Protein content (%)	N in straw (%)	N uptake in grains (kg ha ⁻¹)	N uptake in straw (kg ha ⁻¹)	Total N uptake (kg ha ⁻¹)
Replication	2	0.00087	0.031	0.00015	4.730	15.263	27.952
Main plot	1	0.00024	0.008	0.00001	1.324	2.696	7.800
Error (a)	2	0.00032	0.011	0.00027	0.214	4.567	3.833
Sub-plot treatment	6	0.03025	1.071	0.02434	528.049	332.822	1697.004
Main plot x Sub-plot	6	0.00619	0.219	0.00602	89.348	103.577	335.672
Error (b)	24	0.00025	0.009	0.00054	5.522	6.237	12.892
	41.00						
CV % Main plot		2.74	2.74	6.23	1.63	9.98	3.93
Sub-plot treatment		2.45	2.45	8.87	8.28	11.67	7.21

Table 1.8 Mean sum of square (MSS) for Iron (Fe) content and uptake in grain and straw of rice

Source	d.f.	Fe conc. in grain (mg kg ⁻¹)	Fe conc. in straw (mg kg ⁻¹)	Fe uptake in grain (g ha ⁻¹)	Fe uptake in straw (g ha ⁻¹)
Replication	2	0.779	44.481	200.650	100517.0
Main plot	1	0.517	39.576	75.384	28453.9
Error (a)	2	0.966	15.090	66.824	15566.1
Sub-plot treatment	6	27.389	270.663	14633.188	424317.0
Main plot x Sub-plot	6	3.565	3392.827	1981.498	1015732.0
Error (b)	24	1.419	26.113	244.910	13656.3
	41.00				
CV % Main plot		2.52	2.09	4.81	8.26
Sub-plot treatment		3.04	2.75	9.21	7.74

Table 1.9 Mean sum of square (MSS) for Manganese (Mn) content and uptake in grain and straw of rice

Source	d.f.	Mn conc. in grain (mg kg ⁻¹)	Mn conc. in straw (mg kg ⁻¹)	Mn uptake in grain (g ha ⁻¹)	Mn uptake in straw (g ha ⁻¹)
Replication	2	0.286	0.812	20.078	1616.297
Main plot	1	3.544	1.006	87.205	686.113
Error (a)	2	0.667	1.060	23.421	385.219
Sub-plot treatment	6	7.711	4.362	1148.278	9196.832
Main plot x Sub-plot	6	2.499	3.610	217.652	6194.902
Error (b)	24	1.254	0.907	34.799	313.241
	41.00				
CV % Main plot		8.93	3.07	12.09	8.47
Sub-plot treatment		12.25	3.33	14.74	7.63

Table 1.10 Mean sum of square (MSS) for Zinc (Zn) content and uptake in grain and straw of rice

Source	d.f.	Zn conc. in grain (mg kg ⁻¹)	Zn conc. in straw (mg kg ⁻¹)	Zn uptake in grain (g ha ⁻¹)	Zn uptake in straw (g ha ⁻¹)
Replication	2	0.023	0.222	8.034	13087.42
Main plot	1	0.892	0.275	40.361	2779.46
Error (a)	2	0.307	1.739	7.154	2440.37
Sub-plot treatment	6	2.980	399.623	1123.955	122786.35
Main plot x Sub-plot	6	4.272	58.435	227.634	81027.56
Error (b)	24	0.470	2.148	17.610	2397.87
	41.00				
CV % Main plot		5.18	1.78	5.75	8.21
Sub-plot treatment		6.41	1.98	9.02	8.14

Table 1.11 Mean sum of square (MSS) for Copper (cu) content and uptake in grain and straw of rice

Source	d.f.	Cu conc. in grain (mg kg ⁻¹)	Cu conc. in straw (mg kg ⁻¹)	Cu uptake in grain (g ha ⁻¹)	Cu uptake in straw (g ha ⁻¹)
Replication	2	0.145	0.089	2.412	853.433
Main plot	1	0.002	0.089	1.062	192.737
Error (a)	2	0.002	0.012	0.224	110.255
Sub-plot treatment	6	1.069	2.003	280.137	3588.438
Main plot x Sub-plot	6	0.875	1.101	26.556	2394.065
Error (b)	24	0.131	0.511	4.836	176.028
	41.00				
CV % Main plot		0.82	0.62	2.18	7.25
Sub-plot treatment		7.28	4.00	10.14	9.16

Table 1.12 Mean sum of square (MSS) for available N, P, K and Fe content of soil at crop harvest

Source	d.f.	Available N (kg ha ⁻¹)	Available P (kg ha ⁻¹)	Available K (kg ha ⁻¹)	Available Fe (kg ha ⁻¹)
Replication	2	179.831	0.656	3.809	0.039
Main plot	1	296.027	0.400	24.534	0.549
Error (a)	2	137.683	0.542	2.960	0.099
Sub-plot treatment	6	3323.762	0.874	4.652	0.503
Main plot x Sub-plot	6	526.286	2.556	133.810	0.421
Error (b)	24	189.899	0.887	6.835	0.251
	41.00				
CV % Main plot		16.83	9.10	1.31	4.35
Sub-plot treatment		19.76	11.65	2.00	6.92

Table 1.13 Mean sum of square (MSS) for soil microbial parameters at 60 DAT

Source	d.f.	ARA (N mole ethylene g ⁻¹ hr ⁻¹)	Soil Chlorophyll (µg g ⁻¹ soil)	Dehydrogen ase (µg TPF g ⁻¹ soil day ⁻¹)	Microbial biomass carbon (µg MBC g ⁻¹ soil)
Replication	2	0.064	0.012	0.325	80.310
Main plot	1	0.003	0.004	0.087	106.881
Error (a)	2	0.001	0.001	0.079	14.738
Sub-plot treatment	6	1.338	0.969	77.577	29638.413
Main plot x Sub-plot	6	0.417	0.278	12.028	32139.937
Error (b)	24	0.037	0.025	1.220	652.163
	41.00				
CV % Main plot		3.49	3.55	4.37	3.26
Sub-plot treatment		22.20	17.85	17.16	21.70

Table 1.14 Mean sum of square (MSS) for soil microbial parameters at harvest

Source	d.f.	ARA (N mole ethylene g ⁻¹ hr ⁻¹)	Soil Chlorophyll (µg g ⁻¹ soil)	Dehydrogena se (µg TPF g ⁻¹ soil day ⁻¹)	Microbial biomass carbon (µg MBC g ⁻¹ soil)
Replication	2	0.016	0.011	0.420	4.310
Main plot	1	0.023	0.000	0.107	1488.095
Error (a)	2	0.002	0.000	0.041	556.881
Sub-plot treatment	6	0.535	1.086	34.589	12975.214
Main plot x Sub-plot	6	0.456	0.304	0.816	13290.484
Error (b)	24	0.013	0.031	0.378	571.623
	41.00				
CV % Main plot		7.12	2.49	5.01	30.50
Sub-plot treatment		17.11	25.18	15.31	30.90

Table 1.15 Mean sum of square (MSS) for soil carbon content (%) at 60 DAT

Source	d.f.	Very labile		labile		Less labile	
		0-7.5 cm	7.5-15 cm	0-7.5 cm	7.5-15 cm	0-7.5 cm	7.5-15 cm
Replication	2	0.0001	0.0002	0.0005	0.0011	0.00020	0.0003
Main plot	1	0.0043	0.0002	0.0001	0.0006	0.00001	0.0001
Error (a)	2	0.0005	0.0002	0.0001	0.0003	0.00006	0.0000
Sub-plot treatment	6	0.0270	0.0055	0.0092	0.0088	0.01977	0.0052
Main plot x Sub-plot	6	0.7927	0.0040	0.0369	0.0141	0.00406	0.0013
Error (b)	24	0.0010	0.0008	0.0005	0.0028	0.00020	0.0001
	41.00						
CV % Main plot		5.92	4.22	3.47	6.60	3.39	3.01
Sub-plot treatment		8.18	9.27	6.45	19.09	6.33	7.41

Table 1.16 Mean sum of square (MSS) for soil aggregation (MWD) at 60 DAT

Source	d.f.	Mean Weight Diameter (mm)	
		0-7.5 cm	7.5-15 cm
Replication	2	0.0001	0.003
Main plot	1	0.0007	0.002
Error (a)	2	0.0003	0.001
Sub-plot treatment	6	0.2180	0.133
Main plot x Sub-plot	6	0.3085	0.042
Error (b)	24	0.0019	0.002
	41.00		
CV % Main plot		3.54	7.78
Sub-plot treatment		8.77	13.20