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# Interactions of Microbial Inoculations with Fertilization Options and Crop Establishment Methods on Modulation of Soil Microbial Properties and Productivity of Rice

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## ABSTRACT

A field investigation was undertaken to assess the interactive effects of different crop establishment methods (CEMs) and microbial formulations in field conditions. The two year experiment was carried out in *kharif* season (June to October) of both 2013–14 and 2014–15 at Research Farm of ICAR-Indian Agricultural Research Institute, New Delhi India. The CEMs studied consist of puddled transplanted rice (PTR), system of rice intensification (SRI) and aerobic rice system (ARS); while microbial formulations consist of *Anabaena* sp. (CR1)+*Providencia* sp. (PR3) consortium (MC1) and *Anabaena–Pseudomonas* biofilm formulation (MC2). The microbial inoculation was applied with 75% recommended dose of nutrients (RDN) (90 kg nitrogen ha<sup>-1</sup> and 19.35 kg phosphorus ha<sup>-1</sup>) and compared with 100% RDN. Soil microbiological properties showed significantly higher values in SRI in the first year; while in the second year, SRI and PTR remained on par and superior to ARS. Among the combinations of nutrient supplements, application of *Anabaena–Pseudomonas* biofilm formulation (MC2) with 75% RDN was superior in terms of all microbiological attributes studied [acetylene reduction activity (ARA), soil chlorophyll, soil dehydrogenase activity, microbial biomass carbon and alkaline phosphatase activity]. Application of microbial inoculation led to an increase in milled rice yield from 230 to 240 kg ha<sup>-1</sup>. Among the different combinations of CEMs and inputs investigated the application of 75% RDN+MC2+zinc (Zn) (soil applied 5 kg Zn ha<sup>-1</sup> through zinc sulphate heptahydrate) in SRI can be a judicious option to maximize rice yield and soil health.

KEYWORDS: Aerobic rice, cyanobacteria, microbial biomass carbon, SRI system

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**Data Availability Statement:** Legal restrictions are imposed on the public sharing of raw data. However, authors have full right to transfer or share the data in raw form upon request subject to either meeting the conditions of the original consents and the original research study. Further, access of data needs to meet whether the user complies with the ethical and legal obligations as data controllers to allow for secondary use of the data outside of the original study.

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## 1. INTRODUCTION

 $\mathbf{R}^{\mathrm{ice}}$  is a versatile crop, which grows in diverse hydrological regimes, ranging from deep water and coastal ecosystem to upland conditions in rainfed ecosystems. Rice has a significant impact on resource consumption and output generation (Li et al., 2020; Kumar et al., 2022). Improvement in soil physical properties (water holding capacity, aggregate stability, bulk density and hydraulic conductivity) with application of different organic sources of crop nutrition and biofertilizer was reported in rice-based cropping system. Lower in yield in rice-wheat cropping area in Indo-Gangetic plains was reported due to inadequate nutrient and improper water management with raising several short and long-term problems was reported in Timsina and Connor (2001). Hence, several modifications in rice cultivation and/or establishment practices (Alam et al., 2020; Bhatt et al., 2021) and input addition (Shivay et al., 2022; Singh et al., 2019) have been explored. The main aim of such modification is to enhance the resource use efficiency with or without yield penalty. The major changes in rice cultivation on the agronomic front include-crop establishment techniques, diversification of nutrient sources with increased emphasis on consortiabased microbial inoculation (Jha et al., 2013), micronutrient fertilization (Prasad et al., 2014), crop residue management at the system level (Laharwan et al., 2023) and tillage system at the individual crop (Kumar et al., 2016) and/or at system level (Singh et al., 2020). Rice diversification in the ricewheat cropping system (Shahane and Shivay, 2019) and enhancing rice water productivity (Gopalakrishnan et al., 2014) are other interventions. These new interventions in rice cultivation across diverse ecosystems create significant changes in soil properties. The qualitative and quantitative changes in the soil microbial communities are expected with changes in cultivation practices. This will influence nutrient availability, translocation and crop productivity (Prasanna et al., 2015; Thakur et al., 2013). These all lead to increasing concerns about the sustainability of rice-based cropping systems and soil health (Bhatt et al., 2016; Samal et al., 2017).

Above discussion highlights the need to investigate all soil properties, rather than just chemical properties. Therefore the present study was planned to evaluate combinations of both crop establishment methods (CEMs) and soil amendments application for their effect on selected soil microbial properties. Among CEMs of rice, aerobic rice system (ARS) is getting attention due to its water-saving potential. The soil moisture in ARS is maintained at field capacity and rice is grown on non-puddled soil (Prasad, 2011). In the case of a system of rice intensification (SRI) (Thakur et al., 2016), soil saturation is maintained. Such kind of variation in the hydrological regime has a significant influence on the growth, functioning and diversity of microorganisms. It is well established that the soil microbiota is actively involved in nutrient cycling through their nutrient mobilization (Amadou et al., 2021; Sharma et al., 2013; Adhya et al., 2015), nitrogen fixation (Kaushik, 2014) and secretion of plant growth promoting hormones (Santoyo et al., 2021). Application of cyanobacteriabased inoculations is common in puddled and saturated conditions; while the use of consortia of microbes having cyanobacteria in aerobic rice and their potential impact on soil microbial properties is less investigated. The hypothesis underlying the present investigation was that such CEMs with different inorganic and biological inputs would bring about modifications in the soil microclimate. It leads to create substantial modulation of microbiological activities and crop yields in rice.

#### 2. MATERIAL AND METHODS

#### 2.1. Experimental site

The field experiment was conducted consecutively for two years in rainy season of 2013 and 2014 at Research Farm (14 C Block) of ICAR-Indian Agricultural Research Institute, New Delhi, India (latitude of 28°38' N, longitude of 77°10' E and altitude of 228.6 m above mean sea level). The climate of Delhi is of sub-tropical and semi-arid type with hot and dry summer and cold winter, which falls under the agro-climatic zone 'Trans-Gangetic plains'. The mean annual normal rainfall and evaporation are 650 and 850 mm, respectively. Regarding temperature, May and June are hottest month with maximum temperature ranging between 41°C to 46°C; while there is a drop in temperature from September onward. The January month is coldest month with minimum temperature of 5°C to 7°C. The rainfall received during the first and second years of the rice growing season (June to October) was 1349.8 and 451.4 mm (Supplementary Table 1). The details of physico-chemical properties of soil in the experimental field are given in Table 1.

#### 2.2. Experimental details

The field study was conducted in split plot design involving three crop establishment methods (CEMs) as main plots (76.14 m<sup>2</sup>) viz. puddled transplanted rice (PTR), a system of rice intensification (SRI) and aerobic rice system (ARS) and nine nutrient management options as subplots (8.46 m<sup>2</sup>) (Figure 1). The subplot treatments consisted of application of 100% recommended dose of nutrients (RDN) (120 kg nitrogen ha<sup>-1</sup> and 25.8 kg phosphorus ha<sup>-1</sup>), 75% RDN, 75% RDN+*Anabaena* sp. (CR1)+*Providencia* sp. (PR3) consortium (MC1) and 75% RDN+*Anabaena*–*Pseudomonas* biofilm formulation (MC2). These four treatments were

Table 1: Soil initial properties of the experimental field									
Particulars	Values								
Texture (Hydrometer method) (Bouyoucos, 1962)	Sandy clay loam								
pH (1:2.5 soil: water ratio)	7.6								
Organic carbon (Walkley and Black, 1934)	5.4 g kg <sup>-1</sup>								
Nitrogen (alkaline permanganate extractable) (Subbiah and Asija, 1956)	257 kg ha <sup>-1</sup>								
Phosphorus (NaHCO <sub>3</sub> -extractable) (Olsen et al., 1954)	17 kg ha <sup>-1</sup>								
Potassium (NH <sub>4</sub> OAC-extractable) (Hanway and Heidel, 1952)	327 kg ha <sup>-1</sup>								
Zinc (DTPA-extractable) (Lindsay and Norvell, 1978)	0.85 g kg <sup>-1</sup>								
Acetylene reduction activity	0.89 n moles ethylene g <sup>-1</sup> soil h <sup>-1</sup>								
Soil chlorophyll (Nayak et al., 2004)	$0.23 \ \mu g \ g^{-1} \ soil$								
Soil dehydrogenase activity (Casida et al., 1964)	$9.4 \ \mu g \ TPF$ (triphenyl formazon) $g^{-1}$ soil $h^{-1}$								
Microbial biomass carbon (Nunan et al., 1998)	71.5 mg kg <sup>-1</sup>								
Alkaline phosphatase activity (Tabatabai and Bremner, 1969)	$13.4\mu gPNPg^{1}soilh^{1}$								

	PTR			SRI			ARS	
T <sub>9</sub>	T <sub>1</sub>	T <sub>8</sub>	T <sub>4</sub>	T <sub>6</sub>	T <sub>6</sub>	T <sub>2</sub>	T <sub>7</sub>	T <sub>9</sub>
$T_4$	$T_5$	<b>T</b> <sub>9</sub>	T <sub>9</sub>	T <sub>7</sub>	$T_4$	$T_7$	$T_5$	$T_7$
T <sub>3</sub>	T <sub>3</sub>	$T_4$	T <sub>7</sub>	T <sub>1</sub>	$T_1$	$T_8$	$T_6$	T <sub>6</sub>
$T_6$	T <sub>7</sub>	$T_5$	$T_3$	$T_4$	$T_2$	T <sub>5</sub>	T <sub>3</sub>	$T_8$
$T_7$	$T_2$	$T_2$	T <sub>5</sub>	$T_2$	$T_7$	T <sub>9</sub>	$T_4$	$T_3$
Τ <sub>1</sub>	<b>T</b> <sub>9</sub>	$T_6$	$T_2$	T <sub>3</sub>	Τ,	$T_3$	T <sub>9</sub>	$T_1$
$T_8$	$T_4$	$T_3$	$T_6$	T <sub>9</sub>	$T_3$	$T_6$	T <sub>1</sub>	$T_4$
$T_2$	$T_6$	$T_1$	T <sub>1</sub>	$T_5$	$T_8$	$T_4$	$T_2$	T <sub>5</sub>
$T_5$	T <sub>8</sub>	T <sub>7</sub>	T <sub>8</sub>	T <sub>8</sub>	T <sub>5</sub>	T <sub>1</sub>	$T_8$	$T_2$

Figure 1: Layout of experimental design (Split plot design) T<sub>1</sub>: Control; T<sub>2</sub>: RDN; T<sub>3</sub>: RDN+Zn; T<sub>4</sub>: 75% RDN; T<sub>5</sub>: 75% RDN+Zn; T<sub>6</sub>: 75% RDN+MC1; T<sub>7</sub>: 75% RDN+MC1+Zn; T<sub>8</sub>: 75% RDN+MC2; T<sub>9</sub>: 75% RDN+MC2+Zn); PTR: Puddled transplanted rice; SRI: System of rice intensification; ARS: Aerobic rice system; RDN\*: Recommended dose of nutrients 120 kg N ha<sup>-1</sup> and 25.8 kg P ha<sup>-1</sup>; Zn\*: Soil applied 5 kg Zn ha<sup>-1</sup> through zinc sulphate heptahydrate; MC1: (*Anabaena* sp. (CR1)+*Providencia* sp. (PR3) consortium; MC2: *Anabaena–Pseudomonas* biofilmed formulations applied with Zn (soil applied 5 kg Zn ha<sup>-1</sup> through zinc sulphate heptahydrate) and without Zn making total eight treatments. One absolute control (without any application of fertilizers and microbial inoculation) was ninth treatment. The chemical fertilizers used for N, P and K were urea, single super phosphate and muriate of potash, respectively. All treatments were taken in triplicate plots. The rice variety 'Pusa Sugandh 5' were planted in the experiment. 'Pusa Sugandh 5' is semi-dwarf high yielding aromatic rice variety suitable for north Indian condition. The variety was released in 2005 by central variety release committee (CVRC). The variety matures in 120–125 days with yield potential of 5.5 to 6.0 tonnes ha<sup>-1</sup>. The variety found resistant to gall midge and brown spot and moderately resistant to leaf folder and blast disease.

#### 2.3. Crop management

The sowing of rice in the main field in ARS and sowing of rice in nursery (for preparing seedling for transplanting in SRI and PTR) was done on same date (16th and 17th June in first year and 19th and 20th June in second year). For field preparation, one ploughing, two harrowing and one planking was done in ARS; while in PTR and SRI, puddling (two ploughings in standing water of 5 cm) was done in addition to field preparation mentioned for ARS. For sowing in ARS, 60 kg seeds ha<sup>-1</sup> were used and line sowing was done at 20 cm row to row spacing with seed drill. In case of PTR, 20 kg seeds ha-1 was sown in nursery and seedling of 23-25 days old were transplanted at 20 cm×15 cm spacing (2–3 seedlings at each hill). In SRI, 5 kg seeds ha<sup>-1</sup> was sown in nursery and seedlings of 13–14 days old were transplanted at 20cm×20 cm spacing (1 seedling at each hill). For water management, aerobic condition was maintained throughout the crop growth in ARS and available soil moisture depletion (ASMD) approach for irrigation was used (irrigation at 50% ASMD). The depth of irrigation was 2 cm, up to flowering and 5 cm from flowering to grain filling. In PTR, 5 cm depth of water at each irrigation was applied as and when water disappeared from the surface; while in SRI, saturated field condition was maintained. The depth of water application at each irrigation was 2 cm up to flowering and 5 cm from flowering to grain filling. For weed management, two hand weedings at 20 and 40 days after transplanting (DAT) were done in both PTR and SRI; while in ARS, three hand weedings at 15, 30 and 45 days after sowing (DAS) were done. For application of nutrient management treatments, P, K and Zn was incorporated just before transplanting and broadcasting of N was done in three equal splits at 5, 25 and 45 DAT in both PTR and SRI. In ARS, whole quantity of P, K and Zn as per the treatment detail was applied at the time of sowing by drilling below the seed. For N, 1/3rd dose was applied at the time of sowing by drilling below the seed and the remaining 2/3rd

dose was applied as top dressing (broadcasting) equally at 30 and 60 DAS. Potassium (K) was applied uniformly in all treatments at the rate of 49.8 kg  $ha^{-1}$ .

#### 2.4. Application of microbial inoculants

The formulations of *Anabaena* sp. (cyanobacteria) (CR1)+Providencia sp. (bacteria) (PR3) consortium (MC1) and Anabaena (cyanobacteria)-Pseudomonas (bacteria) biofilm (MC2) were prepared by mixing with vermiculite (hydrous phyllosilicate mineral): compost (paddy straw) (1:1) as the carrier (Nain et al., 2010; Prasanna et al., 2011, 2015). The paddy straw compost has C/N ratio of 16.22:1 and humus content of 13.8% (pH 7.34). The cyanobacterial and bacterial colony forming units in the formulations was  $10^4$  and  $10^8$  g<sup>-1</sup> carrier, respectively (Prasanna et al., 2015). All these cynobacterial and bacterial strains were maintained in microbial culture of Division of Microbiology of ICAR-Indian Agricultural Research Institute, New Delhi. For preparation of microbial inoculants, compost was used as a carrier (300 g). In the compost 80 g of carboxy methyl cellulose (CMC) was added. Bacterial culture nearly 100 ml of (48 hours growth) respective strain and 500 µg of chlorophyll of respective strain of cynobacteria were added. For application of microbial inoculation in rice, slurry of the formulations was made using water along with 1% carboxy methyl cellulose (CMC) as a sticker and seedlings were dipped in this slurry for 30 minutes, just before transplanting in both PTR and SRI. In ARS, pre-soaked seeds were treated with thick slurry of microbial cultures, using 1% CMC as a sticker for 30 minutes, and the coated seeds were dried in shade for 30 minutes.

#### 2.5. Measurement of soil parameters

For measurement of acetylene reduction activity (ARA), triplicate samples of fresh soil cores (0-10 cm) were collected at 70 and 100 DAS. The soil samples were placed in vials and injected with 10% v/v of acetylene (Prasanna et al., 2003). The gas chromatographic estimation of ethylene was done by measuring ARA as an index of nitrogenase activity. Commercially available standard ethylene was utilized for quantification and vials with equivalent volume of water served as controls. The samples were injected into preconditioned Bruker 450 Gas Chromatograph, housing a two-meter long Porapak R stainless steel column and flame ionization detector. The column temperature was maintained at 100°C and the injector and detector at 110°C. A flow rate of 35 ml min<sup>-1</sup> of N<sub>2</sub> served as a carrier gas. Standard ethylene gas was used for calibration and calculations. The values were expressed as n moles of ethylene formed g<sup>-1</sup> soil h<sup>-1</sup>.

For measurement of soil chlorophyll the fresh soil cores (0-10 cm) were collected and acetone: DMSO in ratio of 1:1 was added at a rate of 4 ml g<sup>-1</sup> soil. The contents were

thoroughly shaken and incubated for 48 hours in the dark at room temperature. Intermittent shaking every 24 hours was given to extract the chlorophyll completely. Optical density values were taken at 663, 645 and 630 nm and the chlorophyll concentration determined by using formula (Nayak et al., 2004) given below:

Soil chlorophyll content (µg g<sup>-1</sup> soil)=11.64(O.D.at 663)-2.16(O.D.at 645)+10(O.D.at 630)

The dehydrogenase activity was measured using 6 gram soil incubated with Triphenyl tetrazolium chloride (3%) for 24 hours in dark. Methanol was added to terminate the enzymatic reaction and the supernatant was filtered and absorbance taken at 485 nm (Casida et al., 1964). The values were expressed as  $\mu$ g of triphenylformazon (TPF) g<sup>-1</sup> soil h<sup>-1</sup>. For determination of microbial biomass carbon (MBC), fumigation method given by Nunan et al. (1998) was followed and MBC was expressed as mg kg<sup>-1</sup> soil. The alkaline phosphatase activity was assayed in soil suspended in modified universal buffer (pH 11), along with 1 ml p-nitro phenyl phosphate (PNP) (Tabatabai and Bremner, 1969) and expressed as  $\mu$ g PNP g<sup>-1</sup> soil h<sup>-1</sup>.

## 2.6. Estimation of crop and water productivity

After harvesting of rice crop, the threshing and followed by cleaning of rice grains was done separately for each plot and yield was measured by adjusting the moisture content at 14%. To obtain milled rice, firstly the hulling of rough rice was done in a mini 'Satake Rice Mill' (Satake, 1990) and hulled rice then passed through 'Satake Rice Whitening and Caking Machine' (Satake, 1990) for two minutes and the weight of milled rice was expressed as Mg or t ha-1. The water productivity was measured by summation of water applied through irrigation and rainfall received. The depth of irrigation water applied at each irrigation and number of irrigations applied were used for measurement of amount of water applied through irrigation; while rainfall data from observatory was taken for crop growing duration. The number of irrigations in PTR, SRI and ARS were 11, 11 and 16, respectively in first year; while during second year, it was 18, 20 and 24, respectively. The water productivity is expressed as:

Total water productivity (kg ha-mm<sup>-1</sup>)=Rice grain yield (kg ha<sup>-1</sup>)/Total water applied (mm)

# 2.7. Data analysis

The data obtained from the experiment was statistically analyzed using the F-test as per the procedure given by Gomez and Gomez (1984). Analysis of variance and the Duncan's multiple range test was used to compare the treatment variables at 5% level of significance.

# 3. RESULTS AND DISCUSSION

## 3.1. Acetylene reduction activity (ARA)

The ARA is measured as indicator of nitrogen fixation through nitrogenase enzyme by applied inoculants. The ARA in first year at 70 and 100 DAS was higher by 1.14– 1.56 and 2.3–2.8 n moles ethylene g<sup>-1</sup> soil h<sup>-1</sup>, respectively than the second year (Table 2a and 2b). The variation in ARA across years was found to be distinctly influenced by variation in rainfall and climatic conditions. The survival and growth of inoculated microbes are affected due to soil microclimate (Khare and Arora, 2015) and plant induced modification of rhizospheric soil (Kumar et al., 2013). At the same time, ARA across CEMs at 100 DAS was 1.39–1.57 times and 1.35–1.36 times higher than that of 70 DAS in first and second years, respectively (Table 2a and 2b). The ARA in SRI was significantly higher than both

PTR and ARS both at 70 and 100 DAS in first year; while for second year SRI and PTR remained on par with each other and both found to be statistically superior over ARS. The significant difference in ARA across CEMs reflects the responses of inoculants to variation in the hydrological regimes and soil microclimate modification (Chaudhary et al., 2018). It is well established that this variation in water regime can have distinct effects on the growth, survival and nitrogen fixation activity of inoculated microorganisms (Jha et al., 2004). Among the nutrient management treatments, the highest ARA was recorded with application of 75% RDN+MC2 in PTR at 70 DAS and same treatment with Zn application in SRI at 100 DAS in first year; while during second year, 75% RDN+MC2 at 70 DAS and 75% RDN+MC2+Zn at 100 DAS (both in SRI) recorded the highest value. On comparison across CEMs, the abovementioned treatment was found to be statistically superior

Table 2a: Effect of crop establishment methods and nutrient management options on acetylene reduction activity (n moles ethylene  $g^{-1}$  soil  $h^{-1}$ ) in rice in 2013

Treat- ment	$\begin{array}{c} Control \\ (N_0P_0Zn_0) \end{array}$	RDN	RDN+Zn	75% RDN	75% RDN + Zn	75% RDN +MC1	75% RDN+ MC1+ Zn	75% RDN + MC2	75% RDN+ MC2 + Zn	Mean
70 DAS										
PTR	1.97 <sup>n</sup>	4.20 <sup>m</sup>	4.49 <sup>1</sup>	$4.74^{jk}$	4.62 <sup>k</sup>	8.26 <sup>ab</sup>	8.16 <sup>bcd</sup>	8.34ª	8.33ª	5.90 <sup>(B)</sup>
SRI	1.98 <sup>n</sup>	$5.15^{\mathrm{gh}}$	$5.09^{\mathrm{gh}}$	4.77 <sup>ij</sup>	4.61 <sup>kl</sup>	8.06 <sup>d</sup>	8.21 <sup>abc</sup>	8.12 <sup>cd</sup>	8.16 <sup>bcd</sup>	6.02 <sup>(A)</sup>
ARS	1.92 <sup>n</sup>	$5.17^{\text{g}}$	5.03 <sup>h</sup>	4.89 <sup>i</sup>	5.18 <sup>g</sup>	$7.30^{\mathrm{f}}$	$7.29^{\mathrm{f}}$	7.61 <sup>e</sup>	7.64 <sup>e</sup>	5.78 <sup>(C)</sup>
Mean	1.96 <sup>D</sup>	4.84 <sup>C</sup>	4.87 <sup>c</sup>	4.80 <sup>°</sup>	4.80 <sup>°</sup>	7.87 <sup>B</sup>	7.89 <sup>B</sup>	8.02 <sup>A</sup>	8.04 <sup>A</sup>	
			Crop establishment methods	Nutrient management options	Interaction					
SEm±			0.01	0.03	0.05					
CD (p=0	0.05)		0.04	0.08	0.13					
100 DA	S									
PTR	3.13 <sup>1</sup>	$6.32^{i}$	6.42 <sup>i</sup>	6.50 <sup>i</sup>	6.33 <sup>i</sup>	11.70 <sup>e</sup>	$11.81^{\mathrm{f}}$	13.83 <sup>b</sup>	14.00 <sup>b</sup>	$8.89^{(B)}$
SRI	3.09 <sup>1</sup>	6.84 <sup>h</sup>	7.19 <sup>g</sup>	$7.22^{\mathrm{g}}$	$7.07^{\mathrm{gh}}$	$12.42^{d}$	12.45 <sup>d</sup>	$14.27^{a}$	14.35ª	9.43 <sup>(A)</sup>
ARS	2.29 <sup>m</sup>	5.93 <sup>j</sup>	5.73 <sup>j</sup>	5.24 <sup>k</sup>	5.77 <sup>j</sup>	$10.97^{\mathrm{f}}$	$11.04^{\mathrm{f}}$	12.77°	12.74 <sup>c</sup>	8.05 <sup>(C)</sup>
Mean	2.84 <sup>D</sup>	6.36 <sup>°</sup>	6.44 <sup>C</sup>	6.32 <sup>°</sup>	6.39 <sup>°</sup>	11.70 <sup>B</sup>	$11.77^{B}$	13.62 <sup>A</sup>	13.70 <sup>A</sup>	
			Crop establishment methods	Nutrient management options	Interaction					
SEm±			0.03	0.05	0.09					
CD (p=0	0.05)		0.11	0.14	0.25					

Within a column and row, means followed by the same letter are not significantly different at the 0.05 level of probability by the Duncan's multiple range test. The capital letters denotes significance for nutrient management treatment, capital letters within parenthesis denotes significance for crop establishment methods and small letter denotes the significance for interaction between nutrient management treatment and crop establishment methods

	0 ,									
Treat- ment	$\begin{array}{c} Control \\ (N_0P_0Zn_0) \end{array}$	RDN	RDN+Zn	75% RDN	75% RDN +Zn	75% RDN +MC1	75% RDN+ MC1+ Zn	75% RDN+ MC2	75% RDN+ MC2 +Zn	Mean
70 DAS	5									
PTR		$3.25^{\text{efg}}$	3.38 <sup>ef</sup>	$3.25^{efg}$	$3.09^{\mathrm{fg}}$	$7.37^{ab}$	$7.12^{b}$	$7.07^{\mathrm{b}}$	7.11 <sup>b</sup>	4.76 <sup>(B)</sup>
SRI	$0.94^{h}$	$3.27^{\text{efg}}$	3.43 <sup>e</sup>	$3.25^{efg}$	$3.15^{efg}$	$7.27^{ab}$	7.37 <sup>ab</sup>	7.52ª	$7.14^{b}$	4.82 <sup>(A)</sup>
ARS	$0.97^{\rm h}$	$3.09^{\mathrm{fg}}$	3.04 <sup>g</sup>	$3.05^{\mathrm{fg}}$	$3.38^{\text{ef}}$	5.80 <sup>d</sup>	6.08 <sup>d</sup>	6.11 <sup>cd</sup>	6.43 <sup>c</sup>	4.22 <sup>(C)</sup>
Mean	1.05 <sup>°</sup>	3.21 <sup>B</sup>	3.28 <sup>B</sup>	3.19 <sup>B</sup>	3.21 <sup>B</sup>	6.81 <sup>C</sup>	6.86 <sup>C</sup>	6.90 <sup>°</sup>	6.89 <sup>c</sup>	
			Crop establishment methods	Nutrient management options	Interaction					
SEm±			0.02	0.07	0.12					
CD (p=	0.05)		0.09	0.19	0.33					
100 DA	S									
PTR	1.731	4.00 <sup>ij</sup>	$5.40^{efg}$	5.43 <sup>ef</sup>	$5.25^{\mathrm{efgh}}$	9.12 <sup>abc</sup>	9.15 <sup>abc</sup>	9.17 <sup>ab</sup>	9.45ª	6.52 <sup>(A)</sup>
SRI	2.15 <sup>k</sup>	5.57°	4.96 <sup>h</sup>	$5.10^{\mathrm{fgh}}$	4.90 <sup>h</sup>	8.79°	8.90 <sup>bc</sup>	9.12 <sup>abc</sup>	9.49ª	6.55 <sup>(A)</sup>
ARS	1.32 <sup>m</sup>	$5.05^{\mathrm{gh}}$	4.33 <sup>i</sup>	3.84 <sup>j</sup>	4.34 <sup>i</sup>	8.06 <sup>d</sup>	8.13 <sup>d</sup>	8.06 <sup>d</sup>	8.23 <sup>d</sup>	5.71 <sup>(B)</sup>
Mean	$1.74^{\mathrm{D}}$	4.87 <sup>C</sup>	4.89 <sup>c</sup>	4.79 <sup>c</sup>	4.83 <sup>C</sup>	8.66 <sup>B</sup>	8.73 <sup>B</sup>	$8.78^{B}$	9.06 <sup>A</sup>	
			Crop establishment methods	Nutrient management options	Interaction					
SEm±			0.04	0.08	0.13					
CD (p=	0.05)		0.15	0.22	0.37					

Table 2b: Effect of crop establishment methods and nutrient management options on acetylene reduction activity (n moles ethylene  $g^{-1}$  soil  $h^{-1}$ ) in rice in 2014

to the same treatment applied in other CEMs (except at 100 DAS during second year). Both microbial inoculants applied (MC1 and MC2) differed significantly from each other, with superior performance of MC2 for first year and both remained on par in the second year. The increase in ARA, due to application of inoculation was higher as against the control (3.1-3.7 n moles ethylene g<sup>-1</sup> soil h<sup>-1</sup> at 70 DAS and 4.0–4.4 n moles ethylene g<sup>-1</sup> soil h<sup>-1</sup> at 100 DAS). Microbial strains differ in their ARA (Prasanna et al., 2012, 2015), which accounts for the positive interactions among the CEMs and nutrient management options in rice. Significantly higher ARA with microbial inoculation illustrated that although nitrogen fixation by native microbes was very low in the study area, the inoculants were robust, and exhibited high functional ability in the diverse soil microclimates (CEMs).

#### 3.2. Soil chlorophyll

The soil chlorophyll was measured as an indicator of cyanobacterial colonisation and growth in the different CEMs-induced soil microclimates. It is well known that the addition of photosynthetic biomass influences the microbial communities and nutrient availability in soil (Nayak et al., 2004; Swarnalakshmi et al., 2013). This was illustrated by the trend in ARA and soil chlorophyll across CEMs and nutrient management treatments which remained mostly similar. This indicates that soil chlorophyll can be also act as an indicator of the proliferation of nitrogen fixing microorganisms, particularly photosynthetic diazotrophs. The SRI plots recorded significantly higher soil chlorophyll to the tune of 0.02–0.11 and 0.02–0.28  $\mu$ g g<sup>-1</sup> over PTR and ARS at 70 DAS and a similar increase of 0.30–0.42 and 0.13–0.85  $\mu$ g g<sup>-1</sup> at 100 DAS, respectively (Table 3a and 3b).

Table 3a: Effect of crop establishment methods and nutrient management options on soil chlorophyll ( $\mu g g^{-1}$ ) in rice in 2013 Treat-Control RDN RDN+Zn 75% RDN 75% RDN 75% 75% 75% 75% Mean RDN+  $(N_0P_0Zn_0)$ +Zn RDN RDN+ RDN+ ment +MC1 MC1+ MC2 MC2 Zn +Zn 70 DAS PTR  $1.46^{(B)}$ 0.31<sup>h</sup> 1.19<sup>ef</sup> 1.15<sup>ef</sup> 1.11<sup>f</sup> 1.16<sup>ef</sup> 1.86° 1.90° 2.23ª 2.22<sup>ab</sup> SRI  $1.57^{(A)}$ 0.31<sup>h</sup> 1.27<sup>ef</sup> 1.38<sup>de</sup> 1.19<sup>ef</sup> 1.24<sup>ef</sup> 1.98<sup>bc</sup> 2.08<sup>ab</sup> 2.31ª 2.39ª ARS  $0.25^{h}$ 0.84<sup>c</sup>  $0.79^{\mathrm{g}}$  $0.86^{\text{g}}$  $1.52^{d}$ 1.55<sup>d</sup> 1.85° 1.88°  $1.15^{(C)}$ 0.81<sup>g</sup> Mean 0.29<sup>D</sup> 1.10<sup>C</sup> 1.11<sup>c</sup> 1.04<sup>c</sup> 1.09<sup>c</sup> 1.79<sup>B</sup> 1.85<sup>B</sup> 2.13<sup>A</sup> 2.16<sup>A</sup> Nutrient Crop Interaction establishment management methods options SEm± 0.01 0.05 0.08 CD (*p*=0.05) 0.05 0.14 0.24 100 DAS PTR 1.56<sup>de</sup> 1.57<sup>d</sup> 3.07<sup>b</sup> 2.30<sup>(B)</sup>  $0.76^{\text{fg}}$ 1.66<sup>d</sup> 1.66<sup>d</sup> 3.14<sup>b</sup> 3.54<sup>ab</sup> 3.74<sup>a</sup> SRI 1.62<sup>d</sup> 1.92<sup>d</sup> 1.93<sup>d</sup> 3.46<sup>ab</sup> 3.49<sup>ab</sup> 3.92ª 2.58<sup>(A)</sup> 1.06<sup>f</sup> 1.88<sup>d</sup> 3.91ª 1.73<sup>(C)</sup> 3.05<sup>b</sup> ARS  $0.32^{g}$ 1.13<sup>ef</sup>  $1.04^{\text{f}}$  $0.96^{\mathrm{f}}$  $1.03^{\rm f}$ 2.45° 2.52° 3.10<sup>b</sup>  $0.71^{\text{D}}$ 1.47<sup>c</sup> 1.54<sup>c</sup> 1.47<sup>C</sup> 1.51<sup>c</sup> 2.99<sup>B</sup> 3.05<sup>B</sup> 3.50<sup>A</sup> 3.59<sup>A</sup> Mean Crop Nutrient Interaction establishment management methods options SEm± 0.05 0.10 0.18 CD (p=0.05) 0.18 0.29 0.51

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Within a column and row, means followed by the same letter are not significantly different at the 0.05 level of probability by the Duncan's multiple range test. The capital letters denotes significance for nutrient management treatment, capital letters within parenthesis denotes significance for crop establishment methods and small letter denotes the significance for interaction between nutrient management treatment and crop establishment methods

Application of MC1 leads to 0.63–0.75 and 0.39–1.52  $\mu$ g g<sup>-1</sup> higher chlorophyll than application of RDN at 70 and 100 DAS, respectively. Similar trend was recorded with MC2, with 0.68–1.09 and 0.46–2.03  $\mu$ g g<sup>-1</sup> higher soil chlorophyll. Significantly higher soil chlorophyll was observed in RDN plots than control. The treatment significance values across CEMs were more consistent during the first year, showing superior performance of all treatments in SRI and PTR over ARS. During second year, 100% RDN, 75% RDN+MC1 +Zn and 75% RDN+MC2 had significantly higher soil chlorophyll in SRI; while other treatments remained on par in SRI and ARS at 70 DAS. At 100 DAS, all nutrient management treatments performed equally among SRI and ARS. All treatments receiving microbial inoculation had significantly higher ARA and soil chlorophyll over fertilizer applied treatments which is an indication of suitability of the selected microbial inoculants. This finding is supported by previous reports on the significant role of cyanobacterial

inoculation as agents for better crop growth and system sustainability (Nain et al., 2010; Prasanna et al., 2012).

#### 3.3. Dehydrogenase activity

The wide variation in dehydrogenase enzyme activity among the treatments was observed in our investigation. This indicates the significant role of nutrient inputs (chemical fertilizers or microbial inoculants), as reported earlier by several researchers (Mandal et al., 2007; Prasanna et al., 2015) and crop management (CEMs), on the activities of soil microbial communities and their effective functioning. The values ranged between 5.79–40.6 and 11.6–77.3 µg triphenylformazon (TPF) g<sup>-1</sup> soil h<sup>-1</sup> at 70 and 100 DAS, respectively (Table 4a and 4b). Among the CEMs studied, SRI was statistically superior with 4.4–8.4 and 0.9–8.9 µg TPF g<sup>-1</sup> soil h<sup>-1</sup> higher values of dehydrogenase activity than ARS at 70 and 100 DAS in the first year. Application of 75% RDN+MC2 with and without Zn applied in SRI

$Table \ 3b: Effect \ of \ crop \ establishment \ methods \ and \ nutrient \ management \ options \ on \ soil \ chlorophyll \ (\mu g \ g^{-1}) \ in \ rice \ in \ 2014$										
Treat- ment	$\begin{array}{c} Control \\ (N_0P_0Zn_0) \end{array}$	RDN	RDN+Zn	75% RDN	75% RDN +Zn	75% RDN +MC1	75% RDN+ MC1+ Zn	75% RDN+ MC2	75% RDN+ MC2 +Zn	Mean
70 DAS										
PTR	0.19 <sup>n</sup>	$0.75^{\mathrm{hijkl}}$	$0.79^{\mathrm{ghijk}}$	$0.74^{ijklm}$	$0.76^{\mathrm{hijkl}}$	$1.36^{\text{zbcd}}$	1.35 <sup>abcde</sup>	1.43 <sup>ab</sup>	1.44ª	0.98 <sup>(A)</sup>
SRI	0.29 <sup>n</sup>	$0.87^{\mathrm{fghij}}$	$0.76^{\rm hijkl}$	$0.70^{jklm}$	$0.73^{ijklm}$	1.37 <sup>abc</sup>	1.42 <sup>ab</sup>	1.40 <sup>abc</sup>	1.48ª	1.00 <sup>(A)</sup>
ARS	0.08°	$0.43^{mn}$	$0.48^{klmn}$	0.46 <sup>lmn</sup>	$0.50^{klmn}$	$1.06^{\text{defgh}}$	$1.04^{\rm efghi}$	$1.09^{\text{cdefg}}$	$1.12^{\rm bcdef}$	0.70 <sup>(B)</sup>
Mean	0.19 <sup>c</sup>	0.69 <sup>B</sup>	0.68 <sup>B</sup>	0.63 <sup>B</sup>	0.66 <sup>B</sup>	1.27 <sup>A</sup>	1.27 <sup>A</sup>	1.31 <sup>A</sup>	1.35 <sup>A</sup>	
			Crop establishment methods	Nutrient management options	Interaction					
SEm±			0.03	0.06	0.11					
CD (p=0.	.05)		0.11	0.18	0.31					
100 DAS										
PTR	0.24 <sup>g</sup>	$1.25^{\text{cdef}}$	$1.21^{\text{def}}$	$1.24^{\text{cdef}}$	$1.21^{\text{def}}$	$1.61^{\text{abcd}}$	1.68 <sup>abc</sup>	$1.65^{\text{abcd}}$	$1.65^{\text{abcd}}$	1.30 <sup>(A)</sup>
SRI	0.24 <sup>g</sup>	$1.21^{\text{def}}$	$1.10^{\mathrm{f}}$	$1.26^{\mathrm{bcdef}}$	$1.27^{\text{bcdef}}$	$1.70^{ab}$	1.73ª	$1.68^{\text{abc}}$	1.72ª	1.32 <sup>(A)</sup>
ARS	0.21 <sup>g</sup>	$1.16^{\text{ef}}$	1.15 <sup>ef</sup>	$1.05^{\mathrm{f}}$	$1.08^{\mathrm{f}}$	1.40 <sup>abcdef</sup>	$1.47^{\text{abcdef}}$	$1.58^{\text{abcde}}$	$1.56^{\text{abcde}}$	1.19 <sup>(A)</sup>
Mean	0.23 <sup>C</sup>	1.21 <sup>B</sup>	1.15 <sup>B</sup>	1.18 <sup>B</sup>	1.18 <sup>B</sup>	1.57 <sup>A</sup>	1.63 <sup>A</sup>	1.64 <sup>A</sup>	1.65 <sup>A</sup>	
			Crop establishment methods	Nutrient management options	Interaction					
SEm±			0.05	0.09	0.16					
CD (p=0.	.05)		NS	0.26	0.44					

recorded the highest values, while it remained statistically on par when applied in PTR and ARS. Among the microbial inoculation treatments, MC2 found significantly superior to MC1 when applied both with and without Zn in PTR; while in SRI, significance of MC2 over MC1 was found without Zn and in ARS, values were significant with Zn application in the first year at 70 DAS. During second year, both inoculations remained on par except in PTR without Zn application at 70 DAS and with Zn at 100 DAS. The Zn fertilization brought variation in dehydrogenase activity in conjunction with microbial inoculation. Dehydrogenase enzyme activity is considered as an important indicator of the functionality of the microbial abundance (Jarvan et al., 2014). The greater sensitivity to crop management (Chaudhary et al., 2018) and input addition (Bhaduri et al., 2017) was also reported. The variation in soil moisture regime and variation in rhizospheric effect due to plant growth response to applied treatments across CEMs, affects

the soil microclimate and crop metabolism, thereby soil dehydrogenase activity.

#### 3.4. Microbial biomass carbon (MBC)

The microbial biomass carbon (MBC) is very sensitive fraction of total soil carbon. The present investigation showed distinct responses to changes in crop management practices, as reported by several researchers (Kushwaha et al., 2000; Bhaduri et al., 2017). The significant variations in MBC due to application of fertilizers (Zhao et al., 2014; Bhardwaj et al., 2019) and microbial inoculation were also observed in our experiment. It is also an important criterion for calculating the soil quality index (Liu et al., 2017). In our study, the degree of significant effects of applied treatments in increasing MBC was in the order: rate of N and P application>microbial consortia application>CEMs>Zn fertilization among all observations taken in both years [Figure 2 (a-d)]. Among these factors, the contribution of

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Treat- ment	$\begin{array}{c} Control \\ (N_0P_0Zn_0) \end{array}$	RDN	RDN+Zn	75% RDN	75% RDN +Zn	75% RDN +MC1	75% RDN+ MC1+Zn	75% RDN+ MC2	75% RDN+ MC2+Zn	Mean
70 DAS										
PTR	14.2 <sup>j</sup>	$26.5^{\text{gh}}$	$27.0^{\mathrm{fgh}}$	$26.0^{h}$	$26.3^{h}$	30.6 <sup>de</sup>	31.3 <sup>d</sup>	34.8°	34.1°	27.9 <sup>(B)</sup>
SRI	16.4 <sup>j</sup>	28.7 <sup>ef</sup>	$28.8^{\text{ef}}$	$28.6^{efg}$	$28.9^{\text{ef}}$	37.7 <sup>b</sup>	37.8 <sup>b</sup>	40.6ª	39.9 <sup>ab</sup>	31.9 <sup>(A)</sup>
ARS	10.0 <sup>k</sup>	23.9 <sup>i</sup>	23.6 <sup>i</sup>	22.9 <sup>i</sup>	22.8 <sup>i</sup>	$26.1^{h}$	$26.0^{h}$	$27.0^{\mathrm{fgh}}$	29.2 <sup>e</sup>	23.5 <sup>(C)</sup>
Mean	13.5 <sup>D</sup>	26.3 <sup>c</sup>	26.5 <sup>°</sup>	25.8 <sup>°</sup>	26.0 <sup>°</sup>	31.5 <sup>B</sup>	31.7 <sup>B</sup>	34.2 <sup>A</sup>	34.4 <sup>A</sup>	
			Crop establishment methods	Nutrient management options	Interaction					
SEm±			0.46	0.75	1.30					
CD (p=0	).05)		1.82	2.14	3.71					
100 DAS	<u>S</u>									
PTR	38.1 <sup>h</sup>	58.9°	$59.7^{\text{de}}$	57.9 <sup>ef</sup>	$58.1^{\text{ef}}$	67.0°	66.2°	72.9 <sup>b</sup>	74.2 <sup>b</sup>	61.4 <sup>(B)</sup>
SRI	$39.0^{\rm h}$	62.3 <sup>cd</sup>	$62.7^{\mathrm{cd}}$	59.4°	59.9 <sup>de</sup>	71.2 <sup>b</sup>	71.5 <sup>b</sup>	77.2ª	77.3ª	64.5 <sup>(A)</sup>
ARS	32.1 <sup>g</sup>	$55.1^{\mathrm{fg}}$	54.6 <sup>g</sup>	54.5 <sup>g</sup>	53.9 <sup>g</sup>	59.6 <sup>de</sup>	$60.4^{de}$	65.4°	65.1°	55.6 <sup>(C)</sup>
Mean	36.4 <sup>D</sup>	58.8 <sup>c</sup>	59.0 <sup>°</sup>	57.3 <sup>c</sup>	57.3 <sup>c</sup>	65.9 <sup>B</sup>	66.0 <sup>B</sup>	71.8 <sup>A</sup>	72.2 <sup>A</sup>	
			Crop establishment methods	Nutrient management options	Interaction					
SEm±			0.65	1.07	1.85					
CD (p=0	).05)		2.55	3.04	5.26					

Table 4a: Effect of crop establishment methods and nutrient management options on dehydrogenase activity [ $\mu$ g TPF (triphenyl formazon) g<sup>-1</sup> soil h<sup>-1</sup>] in rice in 2013

Table 4b: Effect of crop establishment methods and nutrient management options on dehydrogenase activity [ $\mu$ g TPF (triphenyl formazon) g<sup>-1</sup> soil h<sup>-1</sup>] in rice in 2014

Treat- ment	$\frac{\text{Control}}{(N_0P_0Zn_0)}$	RDN	RDN+Zn	75% RDN	75% RDN +Zn	75% RDN	75% RDN+	75% RDN+	75% RDN+	Mean
						+MC1	MC1+Zn	MC2	MC2+Zn	
70 DAS	5									
PTR	9.8 <sup>1</sup>	$20.6^{\text{ef}}$	21.1 <sup>e</sup>	$19.9^{\text{efgh}}$	$20.1^{\text{efg}}$	23.0 <sup>d</sup>	23.7 <sup>cd</sup>	25.1 <sup>bc</sup>	24.6 <sup>b</sup>	20.9 <sup>(A)</sup>
SRI	8.1 <sup>1</sup>	$19.0^{\rm fghi}$	$19.1^{\mathrm{fghi}}$	$18.7^{\mathrm{fghijk}}$	$18.8^{\mathrm{fghij}}$	26.3 <sup>ab</sup>	26.3ª	27.1ª	26.5ª	$21.1^{(A)}$
ARS	5.8 <sup>m</sup>	$18.2^{\mathrm{ghijk}}$	$18.0^{\mathrm{hijk}}$	$17.0^{jk}$	16.8 <sup>k</sup>	$18.7^{\mathrm{fghijk}}$	$18.6^{\mathrm{ghijk}}$	17.5 <sup>ijk</sup>	$19.9^{\text{efg}}$	$16.7^{(B)}$
Mean	7.9 <sup>c</sup>	19.3 <sup>B</sup>	19.4 <sup>B</sup>	18.5 <sup>B</sup>	18.6 <sup>B</sup>	22.7 <sup>A</sup>	22.9 <sup>A</sup>	23.2 <sup>A</sup>	23.7 <sup>A</sup>	
			Crop establishment methods	Nutrient management options	Interaction					
SEm±			0.37	0.67	1.16					
CD (p=	0.05)		1.44	1.90	3.29					

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Treat-	Control	RDN	RDN+Zn	75% RDN	75% RDN	75%	75%	75%	75%	Mean
ment	$(\mathbf{N}_0\mathbf{P}_0\mathbf{Z}\mathbf{n}_0)$				+Zn	RDN	RDN+	KDN+	RDN+	
						+MC1	MC1+Zn	MC2	MC2+Zn	
100 DA	<u>IS</u>									
PTR	13.0	38.2	39.1	40.2	38.8	42.6	41.4	43.1	44.2	37.9 <sup>(A)</sup>
SRI	11.6	39.6	39.7	39.3	38.3	44.6	44.4	45.1	45.0	38.6 <sup>(A)</sup>
ARS	12.9	40.7	39.9	40.1	40.5	41.1	41.5	41.4	41.0	37.7 <sup>(A)</sup>
Mean	12.5 <sup>c</sup>	39.5 <sup>B</sup>	39.6 <sup>B</sup>	39.9 <sup>B</sup>	39.2 <sup>B</sup>	42.8 <sup>A</sup>	42.4 <sup>A</sup>	43.2 <sup>A</sup>	43.4 <sup>A</sup>	
			Crop establishment methods	Nutrient management options	Interaction					
SEm±			0.47	0.78	1.36					
CD ( <i>p</i> =	0.05)		NS	2.23	NS					





Figure 2: Effect of crop establishment methods and nutrient management options on microbial biomass carbon (mg kg<sup>-1</sup>) at -70 DAS in 2013 (a), 100 DAS in 2013 (b), 70 DAS in 2014 (c) and 100 DAS (d). T<sub>1</sub>: Control; T<sub>2</sub>: RDN; T<sub>3</sub>: RDN+Zn; T4: 75% RDN; T<sub>5</sub>: 75% RDN+Zn; T<sub>6</sub>: 75% RDN+MC1; T<sub>7</sub>: 75% RDN+MC1+Zn; T<sub>8</sub>: 75% RDN+MC2; T<sub>9</sub>: 75% RDN+MC2+Zn; PTR: Puddled transplanted rice; SRI: System of rice intensification; ARS: Aerobic rice system; RDN: Recommended dose of nutrients 120 kg N ha<sup>-1</sup> and 25.8 kg P ha<sup>-1</sup>; Zn: Soil applied 5 kg Zn ha<sup>-1</sup> through zinc sulphate heptahydrate; MC1: (*Anabaena* sp. (CR1)+*Providencia* sp. (PR3) consortium; MC2: *Anabaena–Pseudomonas* biofilmed formulations

rate of N and P and microbial consortia in increasing MBC was higher at 100 DAS than 70 DAS; while for CEMs, it was higher at 70 DAS. The contribution of Zn fertilization was lowest and more or less remained same at both 70 and 100 DAS. The contribution of rate of N and P application, application of microbial inoculation, CEMs and Zn fertilization to MBC at 70 DAS varied between 80.1–81.3, 17.2–24.3, 13.7–21.5 and 0.2–0.6 mg kg<sup>-1</sup>, respectively; while at 100 DAS, the values were 108–109.9, 19.0–27.7, 10.9–18.7 and 0.3–2.6 mg kg<sup>-1</sup>, respectively. The better

crop and root growth and higher root secretion, generates substrates for microbial growth, reflected as higher increases in MBC with fertilization (rate of N and P application), as also recorded in our investigation. The positive effect of fertilization on MBC in long-term experiments in maize-wheat-cowpea cropping system has been reported by Kanchikerimath and Singh (2001).

The SRI had the highest MBC in first year and found significantly superior to PTR and ARS; while during second year, the differences were not significant among the CEMs.

The variation in MBC among inoculated and un-inoculated treatment was similar as that of dehydrogenase activity. All the inoculated treatments had significantly higher MBC than un-inoculated at all observations. At the same time, MBC was higher at 100 DAS in both years than 70 DAS. The application of RDN and 75% RDN did not differ significantly, and both found significantly superior over control in both years. The significantly higher MBC in inoculated treatments over uninoculated is an indication of the strong and beneficial influence of microbial inoculation on the soil microbial population and activities. This change in MBC is also governed by the rhizosphere activities (root secretion) and indirectly impacting plant growth.

# 3.5. Alkaline phosphatase activity (APA)

The APA is an indicator of P mobilizing and solubilization which can be used by soil macro- and micro-biota and

plants. The increase in APA in first year due to microbial inoculation was 5.8-8.5 and 11.4-15.6 µg PNP g<sup>-1</sup> soil h<sup>-1</sup> at 70 and 100 DAS, respectively. Similar increase was also observed during the second year of study (Table 5a and 5b). The APA values did not differ significantly due to nutrient application rate (100% and 75% RDN) in the first year and 70 DAS in the second year; while at 100 DAS in second year, it differed significantly in SRI and ARS (with Zn application). The lower value of APA in control over other treatments indicates that, application of chemical fertilizers had a positive effect on the population build-up of inherent P mobilizing microorganisms. The positive effect of fertilizer application on P solubilisation might be due to the better root growth, leading to greater root carbon secretion indirectly and directly due to provision of usable P for solubilisation by inherent microorganisms. The increase in APA due to application of microbial inoculation indicates

Table 5a Effect of crop establishment methods and nutrient management options on alkaline phosphatase activity ( $\mu$ g PNP g<sup>-1</sup> soil h<sup>-1</sup>) in rice in 2013

8 0011	)	1010								
Treat- ment	$\begin{array}{c} Control \\ (N_0P_0Zn_0) \end{array}$	RDN	RDN+Zn	75% RDN	75% RDN +Zn	75% RDN +MC1	75% RDN+ MC1+ Zn	75% RDN+ MC2	75% RDN+ MC2+ Zn	Mean
70 DAS	5									
PTR	30.8 <sup>k</sup>	$68.0^{i}$	$69.0^{\mathrm{hi}}$	$69.0^{\mathrm{hi}}$	$69.6^{\mathrm{ghi}}$	75.6 <sup>de</sup>	$75.7^{\text{cde}}$	77.6 <sup>bcd</sup>	$78.2^{bcd}$	68.1 <sup>(B)</sup>
SRI	33.0 <sup>k</sup>	$71.1^{\mathrm{fgh}}$	72.8 <sup>ef</sup>	$72.3^{\mathrm{fg}}$	$72.3^{\mathrm{fg}}$	$78.8^{\text{ab}}$	78.6 <sup>abc</sup>	81.4ª	81.3a	71.3 <sup>(A)</sup>
ARS	$27.2^{1}$	63.7 <sup>j</sup>	64.5 <sup>j</sup>	64.3 <sup>j</sup>	64.4 <sup>j</sup>	$68.6^{\mathrm{hi}}$	$69.6^{\mathrm{hi}}$	$72.0^{\mathrm{fg}}$	$72.4^{\mathrm{fg}}$	63.0 <sup>(C)</sup>
Mean	30.3 <sup>D</sup>	67.6 <sup>°</sup>	68.8 <sup>C</sup>	68.5 <sup>°</sup>	68.7 <sup>°</sup>	74.3 <sup>B</sup>	74.6 <sup>B</sup>	77.0 <sup>B</sup>	77.3 <sup>A</sup>	
			Crop establishment methods	Nutrient management options	Interaction					
SEm±			0.34	0.59	1.02					
CD (p=	0.05)		1.33	1.68	2.90					
100 DA	S									
PTR	56.1 <sup>1</sup>	$118.7^{\mathrm{ghi}}$	$119.0^{\mathrm{ghi}}$	$117.3^{hi}$	116.2 <sup>ij</sup>	128.7 <sup>cd</sup>	129.1 <sup>cd</sup>	133.4 <sup>abc</sup>	134.5 <sup>ab</sup>	117.0 <sup>(B)</sup>
SRI	$61.0^{1}$	$122.4^{\mathrm{fgh}}$	$123.3^{\mathrm{fg}}$	$122.6^{\mathrm{fg}}$	$124.5^{\text{def}}$	132.5 <sup>bc</sup>	$133.2^{\text{abc}}$	136.9 <sup>ab</sup>	137.8ª	121.6 <sup>(A)</sup>
ARS	51.2 <sup>m</sup>	112.2 <sup>j</sup>	112.6 <sup>j</sup>	110.6 <sup>k</sup>	112.0 <sup>j</sup>	$123.5^{\text{ef}}$	$124.2^{\text{def}}$	$127.0^{\text{def}}$	$128.5^{\text{cde}}$	111.3(C)
Mean	56.1 <sup>D</sup>	117.8 <sup>c</sup>	118.3 <sup>c</sup>	116.8 <sup>c</sup>	117.5 <sup>c</sup>	128.2 <sup>B</sup>	128.8 <sup>B</sup>	132.4 <sup>A</sup>	133.6 <sup>A</sup>	
			Crop establishment methods	Nutrient management options	Interaction					
SEm±			0.61	1.04	1.80					
CD (p=	0.05)		2.41	2.95	5.12					

Within a column and row, means followed by the same letter are not significantly different at the 0.05 level of probability by the Duncan's multiple range test. The capital letters denotes significance for nutrient management treatment, capital letters within parenthesis denotes significance for crop establishment methods and small letter denotes the significance for interaction between nutrient management treatment and crop establishment methods

Treat- ment	$\begin{array}{c} Control \\ (N_0P_0Zn_0) \end{array}$	RDN	RDN+Zn	75% RDN	75% RDN +Zn	75% RDN +MC1	75% RDN+ MC1+ Zn	75% RDN+ MC2	75% RDN+ MC2+ Zn	Mean
70 DAS										
PTR	- 11.8 <sup>g</sup>	49.6 <sup>cd</sup>	50.0 <sup>cd</sup>	49.2 <sup>cde</sup>	49.7 <sup>cd</sup>	53.2 <sup>ab</sup>	53.4 <sup>ab</sup>	53.6 <sup>ab</sup>	54.0ª	47.2 <sup>(A)</sup>
SRI	11.7 <sup>g</sup>	50.4°	51.4 <sup>bc</sup>	50.3°	$50.2^{cd}$	54.1ª	54.1ª	55.1ª	54.8ª	48.0 <sup>(A)</sup>
ARS	$9.7^{\mathrm{g}}$	46.8 <sup>ef</sup>	47.0 <sup>ef</sup>	$46.1^{\mathrm{f}}$	46.0 <sup>f</sup>	$47.7^{\text{def}}$	48.9 <sup>cde</sup>	49.5 <sup>cd</sup>	49.7 <sup>cd</sup>	43.5 <sup>(B)</sup>
Mean	11.1 <sup>D</sup>	48.9 <sup>c</sup>	49.4 <sup>BC</sup>	48.5 <sup>°</sup>	48.6 <sup>°</sup>	51.6 <sup>AB</sup>	52.1 <sup>B</sup>	52.7 <sup>A</sup>	52.8 <sup>A</sup>	
			Crop establishment methods	Nutrient management options	Interaction					
SEm±			0.29	0.50	0.87					
CD (p=0	).05)		1.13	1.42	2.47					
100 DA	<u>S</u>									
PTR	29.6 <sup>k</sup>	89.9 <sup>cdef</sup>	$87.8^{\mathrm{efghi}}$	$88.5^{\rm efg}$	$87.1^{\mathrm{fghi}}$	94.4 <sup>abcd</sup>	94.7 <sup>abc</sup>	95.4 <sup>abc</sup>	96.9ª	84.9 <sup>(A)</sup>
SRI	31.5 <sup>k</sup>	$90.5^{\mathrm{bcdef}}$	$93.1^{\text{abcde}}$	$90.8^{\mathrm{bcdef}}$	84.1 <sup>ghij</sup>	95.2 <sup>abc</sup>	95.9 <sup>ab</sup>	96.0 <sup>ab</sup>	97.2ª	86.0 <sup>(A)</sup>
ARS	$23.7^{1}$	82.3 <sup>ij</sup>	$82.7^{ m hij}$	80.8	$86.1^{\mathrm{fghij}}$	$88.2^{\rm efgh}$	$88.9^{\text{defg}}$	$88.1^{\rm efgh}$	89.9 <sup>cdef</sup>	79.0 <sup>(B)</sup>
Mean	28.2 <sup>°</sup>	87.6 <sup>B</sup>	87.9 <sup>B</sup>	86.7 <sup>B</sup>	85.8 <sup>B</sup>	92.6 <sup>A</sup>	93.2 <sup>A</sup>	93.2 <sup>A</sup>	94.7 <sup>A</sup>	
			Crop establishment methods	Nutrient management options	Interaction					
SEm±			0.56	1.11	1.93					
CD (p=0	).05)		2.19	3.17	5.49					

Table 5b: Effect of crop establishment methods and nutrient management options on alkaline phosphatase activity ( $\mu g PNP g^{-1} \operatorname{soil} h^{-1}$ ) in rice in 2014

the superior performance of externally inoculated microbes.

The interaction effect between CEMs and nutrient management options for APA was more consistent than ARA. This indicates less sensitivity of APA than ARA to variation in weather conditions during crop growth stages. This might be due to acid phosphatase activity of plant roots (Spohn and Kuzyakov, 2013) and adaptation of P solubilizing microorganisms to a wider range of soil microclimate and weather variations.

#### 3.6. Milled rice yield and total water productivity

The milled rice yield and total water productivity varied between 1.94 to 2.95 Mg ha<sup>-1</sup> and 1.04 to 1.99 kg hamm<sup>-1</sup> (Table 6 and 7). The CEMs differed significantly in terms of both milled rice yield and total water productivity. Application of 75% RDN+MC2+Zn in SRI had highest yield in both year and found significantly higher than same

treatment applied in PTR and ARS in second year; while in first year, SRI and PTR stand on par and found statistically superior over ARS (Table 6). The response of rice to Zn fertilization and microbial inoculation in terms of milled rice yield was ranged from 90 to 150 kg ha<sup>-1</sup> and 230 to 240 kg ha<sup>-1</sup>, respectively. This point outs their significant role in providing primary nutrients, facilitating vigorous growth and enhancing the milled rice yield. The increase in productivity due to application of microbial consortia highlights their role as cost-effective and environmentfriendly option (Shivay et al., 2022). Yield in control was found on par in all CEMs indicating the applied treatment alone created positive interactions between CEMs and nutrient management treatments. Microbial inoculated treatments differ in their response to CEMs; while application with Zn found superior over without Zn application in both year. The interaction between CEMs

Table 6	Table 6: Effect of crop establishment methods and nutrient management options on milled rice yield (t ha <sup>-1</sup> ) in 2013 and 2014											
Treat- ment	$\begin{array}{c} Control \\ (N_0P_0Zn_0) \end{array}$	RDN	RDN+Zn	75% RDN	75% RDN +Zn	75% RDN +MC1	75% RDN+ MC1+ Zn	75% RDN+ MC2	75% RDN+ MC2+ Zn	Mean		
2013												
PTR	2.06 <sup>no</sup>	$2.83^{\text{defg}}$	2.99 <sup>ab</sup>	$2.58^{\rm klm}$	2.64 <sup>ijkl</sup>	$2.87^{\text{bcdef}}$	$2.98^{\text{abc}}$	$2.85^{\text{bcdef}}$	$2.88^{\text{abcdef}}$	$2.74^{(A)}$		
SRI	2.20 <sup>n</sup>	$2.87^{\text{bcdef}}$	$2.97^{\text{abcd}}$	$2.50^{lm}$	$2.66^{\text{hijk}}$	$2.86^{\text{bcdef}}$	$2.92^{\text{abcde}}$	$2.86^{\text{cdef}}$	3.02ª	$2.76^{(A)}$		
ARS	2.02°	$2.69^{\mathrm{ghij}}$	$2.80^{efgh}$	2.40 <sup>m</sup>	$2.54^{\rm klm}$	$2.56^{jkl}$	$2.77^{\mathrm{fghi}}$	$2.60^{\text{jkl}}$	$2.79^{\text{efgh}}$	$2.57^{(B)}$		
Mean	2.09 <sup>E</sup>	2.80 <sup>B</sup>	2.92 <sup>A</sup>	2.49 <sup>D</sup>	2.61 <sup>C</sup>	2.76 <sup>B</sup>	2.89 <sup>A</sup>	2.77 <sup>B</sup>	2.90 <sup>A</sup>			
			Crop establishment methods	Nutrient management options	Interaction							
SEm±			0.01	0.03	0.05							
CD (p=	0.05)		0.06	0.08	0.14							
2014												
PTR	$1.90^{i}$	2.67 <sup>c</sup>	2.86 <sup>a</sup>	$2.48^{def}$	$2.48^{def}$	2.69°	2.83 <sup>ab</sup>	2.68 <sup>c</sup>	2.74 <sup>b</sup>	2.59 <sup>(A)</sup>		
SRI	$2.04^{h}$	2.70°	2.85ª	$2.40^{efg}$	$2.50^{de}$	2.68 <sup>c</sup>	2.73 <sup>b</sup>	2.68 <sup>c</sup>	2.87ª	2.61 <sup>(A)</sup>		
ARS	$1.87^{i}$	2.53 <sup>d</sup>	2.67°	2.30 <sup>g</sup>	$2.38^{\mathrm{fg}}$	$2.39^{\mathrm{fg}}$	2.70°	$2.45^{de}$	2.65°	2.44 <sup>(B)</sup>		
Mean	1.94 <sup>D</sup>	2.63 <sup>B</sup>	2.79 <sup>A</sup>	2.39 <sup>c</sup>	2.45 <sup>°</sup>	2.59 <sup>B</sup>	2.75 <sup>A</sup>	2.60 <sup>B</sup>	2.75 <sup>A</sup>			
			Crop establishment methods	Nutrient management options	Interaction							
SEm±			0.01	0.02	0.03							
CD (p=	0.05)		0.04	0.06	0.10							

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Within a column and row, means followed by the same letter are not significantly different at the 0.05 level of probability by the Duncan's multiple range test. The capital letters denotes significance for nutrient management treatment, capital letters within parenthesis denotes significance for crop establishment methods and small letter denotes the significance for interaction between nutrient management treatment and crop establishment methods

Table 7: Effect of crop establishment methods and nutrient management options on water productivity (kg ha-mm<sup>-1</sup>) in 2013 and 2014

Treat- ment	$\begin{array}{c} Control \\ (N_0P_0Zn_0) \end{array}$	RDN	RDN+Zn	75% RDN	75% RDN +Zn	75% RDN +MC1	75% RDN+ MC1+ Zn	75% RDN+ MC2	75% RDN+ MC2+ Zn	Mean
2013										
PTR	0.991	$1.36^{\text{efg}}$	1.44 <sup>cd</sup>	1.24 <sup>i</sup>	$1.27^{i}$	$1.38^{\text{def}}$	1.43 <sup>c</sup>	$1.37^{\text{defg}}$	1.38def	1.32 <sup>(C)</sup>
SRI	1.15 <sup>j</sup>	1.49 <sup>bc</sup>	$1.55^{ab}$	$1.30^{\mathrm{gh}}$	$1.39^{\text{def}}$	1.49 <sup>bc</sup>	1.52ª	1.48 <sup>bc</sup>	1.57ª	1.44 <sup>(A)</sup>
ARS	$1.07^{k}$	1.43 <sup>cde</sup>	1.49 <sup>bc</sup>	$1.28^{h}$	$1.35^{\mathrm{fgh}}$	$1.36^{\text{efg}}$	1.47°	$1.38^{\text{def}}$	$1.48^{bc}$	1.37 <sup>(B)</sup>
Mean	$1.07^{\text{E}}$	1.43 <sup>B</sup>	1.49 <sup>A</sup>	$1.27^{\mathrm{D}}$	1.34 <sup>c</sup>	1.41 <sup>B</sup>	1.47 <sup>A</sup>	1.41 <sup>B</sup>	1.48 <sup>A</sup>	
			Crop establishment methods	Nutrient management options	Interaction					
SEm±			0.01	0.01	0.02					
CD (p=0.0	05)		0.03	0.04	0.07					

Treat- ment	$\begin{array}{c} Control \\ (N_0P_0Zn_0) \end{array}$	RDN	RDN+Zn	75% RDN	75% RDN +Zn	75% RDN +MC1	75% RDN+ MC1+ Zn	75% RDN+ MC2	75% RDN+ MC2+ Zn	Mean
2014										
PTR	1.09 <sup>1</sup>	1.54 <sup>j</sup>	1.64 <sup>h</sup>	1.43 <sup>k</sup>	1.43 <sup>k</sup>	1.55 <sup>j</sup>	$1.63^{\mathrm{hi}}$	1.54 <sup>j</sup>	$1.57^{ij}$	1.49 <sup>(C)</sup>
SRI	$1.58^{\mathrm{hi}}$	$2.09^{d}$	2.20 <sup>c</sup>	1.86 <sup>f</sup>	1.94°	$2.07^{d}$	$2.11^{d}$	$2.07^{d}$	2.22 <sup>c</sup>	2.02 <sup>(B)</sup>
ARS	1.73 <sup>g</sup>	2.34 <sup>b</sup>	2.47ª	2.13 <sup>d</sup>	2.20 <sup>c</sup>	2.21°	2.50ª	2.26 <sup>c</sup>	2.45ª	$2.25^{(A)}$
Mean	$1.47^{\mathrm{F}}$	1.99 <sup>B</sup>	2.10 <sup>A</sup>	$1.81^{E}$	1.86 <sup>D</sup>	1.94 <sup>c</sup>	2.08 <sup>A</sup>	1.96 <sup>BC</sup>	2.08 <sup>A</sup>	
			Crop establishment methods	Nutrient management options	Interaction					
SEm±			0.01	0.01	0.02					
CD ( <i>p</i> =0.05)			0.03	0.04	0.06					

and nutrient application treatment was found significant indicating the impact of soil preparation and plant on microbial survival besides impact of amendment application and microbial consortia application. Therefore, microbial consortia can be used to reduce doses of chemical fertilizers (both N and P). The calculation of water productivity for milled rice is more appropriate as it represents the final produce which is consumed. Water productivity was significantly different in both CEMs and nutrient management treatments. Among CEMs, SRI in first year and ARS in second year found significantly superior over other methods (Table 7). Application of microbial consortia had significantly higher water productivity over application of 75% RDN alone in PTR and SRI; while significance is not consistent in ARS. Application of RDN found superior over 75% RDN in all CEMs. The increase in total water productivity due to application of RDN was 0.11 to 0.21 kg ha<sup>-1</sup>-mm over sub-optimal fertilization, while application of MC increases it by 0.08 to 0.19 kg ha-mm<sup>-1</sup>. The variation in water productivity observed across CEMs is directly due to differences in the quantity of water applied; while changes in grain yield due to applied treatments indirectly helps in increasing water productivity (Singh, 2013). Hence, enhancing resource use efficiency, which is crucial in the climate change scenario and dwindling resources also leads to higher crop productivity and can be recommended to meet the demands of the burgeoning population.

# 4. CONCLUSION

Application of 75% RDN+MC2 (Anabaena-Pseudomonas biofilm)+Zn in SRI was found to be the most

promising option for high yields and enhancing all soil microbial properties (Acetylene reductase activity, soil chlorophyll, dehydrogenase activity, microbial biomass carbon and alkaline phosphatase activity). The significantly higher values of all studied microbial properties in SRI and PTR over ARS illustrate the positive effect of maintaining soil under saturated condition in rice.

#### 5. SUPPLEMENTARY MATERIALS

The values of weather parameters during rice growing season recorded at Class B non-crop weather observatory of ICAR-Indian Agricultural Research Institute, New Delhi, India is given in supplementary Table 1.

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