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Influence of Crop Establishment Practices and Microbial Inoculants on Nodulation of Summer Green Gram (*Vigna radiata*) and Soil Quality Parameters

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ABSTRACT

A field experiment was carried out during summer season in 2018 at New Delhi to study the effect of crop establishment practices and microbial inoculation on nodulation of summer green gram [*Vigna radiata* (L.) Wilczek] and soil quality parameters. The experiment was laid out in split plot design and treatments were replicated thrice. The experiment included nine treatment combinations including three crop establishment practices *viz*., conventional tillage, zero tillage and zero tillage with chick pea residue @ 2.5 t/ha in main plots and three microbial inoculation treatment *viz*., control (no inoculation), dual inoculation of *Rhizobium* + PSB and combined inoculation of *Rhizobium*+ Phosphate Solubilizing Bacteria (PSB) + Arbuscular Mycorrhizal (AM) Fungi in sub-plots. Results showed that zero tillage with residue produced significantly higher no. of root nodules/plant and root nodule weight/ plant at 25, 35 and 45 DAS. The same treatment showed significantly superior soil chemical parameters *viz*., available N, P and K and soil microbial parameters like dehydrogenase activity, alkaline phosphatase activity and microbial biomass carbon (MBC) over other two treatments. Seed inoculation with *Rhizobium*+ PSB + AM Fungi was significantly superior to other two treatments with regard to no. of root nodules/ plant and root nodule weight/ plant at 25, 35 and 45 DAS, soil chemical and microbial parameters.

Key words: Microbial inoculants, Microbial parameters, Residue and zero tillage, Root nodules, Soil quality, Summer green gram, Zero tillage.

INTRODUCTION

India stands first globally in pulse production contributing about 25% of total pulse production. Pulses are also an important component of Indian agricultural economy next to food grains and oilseeds in terms of acreage, production and economic value (Choudhary *et al*., 2009). Pulses play a significant role in providing nutritionally balanced diet. Pulses are the major source of dietary protein for vegetarian population. The 68th UN General Assembly had declared 2016 as 'International Year of Pulses' with theme 'Nutritious food for a sustainable future' (FAO, 2016) which showed global importance of pulses. Pulses also act as an effective source for reversing the soil degradation process and can contribute in achieving the twin objectives of increasing crop productivity along with improving the sustainability of the cereal based cropping systems (Narayan and Kumar, 2015).

Green gram [*Vigna radiata* (L.) Wilczek] is one of the important pulse crop suited for arid and semi-arid regions of India. Green gram stands third after chickpea and pigeon pea among the pulses. It has occupied 34.00 lakh ha area and contributes 23.70 lakh tonnes in pulse production in the country (DAC, 2018-19). Green gram, a short duration pulse crop can be grown as catch crop during *kharif* and summer seasons (Kumar *et al*., 2020). The most potential technologies in crop production include improved crop establishment methods. Use of excessive and unnecessary tillage operations is harmful to soil and adds to production cost. Intensive tillage-based agriculture practices without recycling of organic resources deteriorate the soil quality (Ram *et al*., 2016), which then reduce the overall productivity of green gram. The conservation agriculture (CA) system based on no-till and residue management are considered alternative to ensure double cropping, improved farm income and livelihood (Ghosh *et al*., 2010). It has the potential to improve resource-use efficiency, crop productivity and soil health, while maintaining the environment. Crop residues are important natural resource in the stability of agricultural ecosystems which can be used as mulch plays an important role in improving soil biological activities, soil organic matter content and in turn helps improve physical, chemical and biological soil properties (Singh *et al*., 2019).

Microbial inoculants are organic products containing a specific micro-organism which are derived from the nodules of plant or from soil of root zone (rhizosphere). They offer important technology to Indian agriculture holding a promise

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to balance many of the shortcomings of conventional chemical based technologies. Biofertilizers (microbial inoculants) have major role in nitrogen assimilation and phosphorus solubilization/mobilization and thereby bringing sustainability in soil fertility and pulse production. Microbial inoculants like *Rhizobium* possess unique ability to fix atmospheric nitrogen by living symbiotically, phosphate solubilising bacteria (PSB) plays a vital role in solubilization of various inorganic and organic phosphates added to the soil (Bhavya *et al*., 2018). Arbuscular mycorrhiza (AM) Fungi plays a vital role in supplementing major plant nutrients like nitrogen, phosphorus and micro nutrients like Fe, Zn requirement of crops. Microbial inoculants offer a cheaper, low capital intensive, non-bulky and renewable source, low price plant nutrient improving fertilizers and ecofriendly route to boost farm productivity depending upon their activity of mobilizing nutrients. Hence biofertilizers are essential to sustain crop production, preserve soil health and biodiversity (Unnikrishnan and Vijayaraghavan, 2019). Considering importance of these low cost technologies, a field experiment was conducted to evaluate the effects of crop establishment practices and microbial inoculation on nodulation of summer green gram and soil quality parameters.

MATERIALS AND METHODS

A field experiment was conducted at the research farm of ICAR-Indian Agricultural Research Institute, New Delhi during summer 2018 season. The soil of the experimental field was sandy loam in texture with 61.23% sand, 15.59% silt and 23.25% clay. The soil was low in organic C, low in available N and medium in available P and K with pH of 7.6. The experiment was conducted in a split-plot design with nine treatment combinations, keeping three methods of crop establishment *viz*. conventional tillage (CT), zero tillage (ZT) and zero tillage with residue (ZT+R) in main plots and three microbial inoculant treatments *viz*. dual inoculation of *Rhizobium*+ Phosphate Solubilizing Bacteria (PSB), the combined inoculation of *Rhizobium*+ PSB + Arbuscular Mycorrhizal (AM) Fungi and control where no seed inoculation was done. Seeds were treated with *Rhizobium* and PSB and AM Fungi applied in soil before sowing. Nitrogen, phosphorus and potassium were uniformly applied as basal at the rate of 20, 40 and 20 kg/ha, respectively to all plots. Other agronomic practices were followed as per the standard packages of practices to raise the green gram crop. Five plants were carefully removed from each plot without damaging the root nodules at 25, 35 and 45 DAS. No. of effective root nodules were counted after thorough and gentle washing of roots, their dry weight was recorded. No. of root nodules/plant and nodule weight/plant was calculated. Before sowing of the crop and after harvest of the crop, soil samples were collected randomly from 0-15 cm soil depth of the experimental plots, the soil samples were shade dried, ground with pestle and morter and passed through 2 mm sieve. The processed samples were analysed for available N, P, K as per the standard procedures [Available nitrogen-alkaline potassium permanganate ($KMnO_4$) method (Subbiah and Asija, 1956), Available phosphorus - Olsen's reagent (Watanable and Olsen, 1965) and Available potassium-flame photometric method (Jackson, 1973]. Soil samples near the rhizosphere (0-15 cm depth) were collected before sowing and at flowering stage of crop. The processed soil samples were used for analyzing microbial parameters *viz.,* dehydrogenase activity (Casida *et al*., 1964), alkaline phosphatase activity (Tabatabai and Bremner, 1969) and microbial biomass carbon (Vance *et al.,* 1987). At harvest, grain yield was recorded separately for each plot and reported at 12% moisture. The data obtained from the experiment were statistically analyzed using the F-test as per the standard procedure to determine the significance of difference between treatment means.

RESULTS AND DISCUSSION

Root nodulation

The weight of root nodules/plant and no. of nodule/ plant of green gram were significantly influenced due to the methods of crop establishment and microbial inoculations at 25, 35 and 45 DAS. Nodule weight/plant and no. of nodule/ plant increased between 25 DAS to 45 DAS and thereafter declined. The weight of root nodules and no. of nodule/ plant at 25, 35 and at 45 DAS were highest in zero tillage with residue treatment followed by conventional tillage and lowest in zero tillage treatment (Table 1). Similar result was reported by Meena *et al*. (2015). Amongst microbial inoculation treatments, combined inoculation of *Rhizobium* + PSB +AM Fungi recorded highest nodule weight and no. of nodule/ plant at 25, 35 and at 45 DAS followed by dual inoculation of *Rhizobium* + PSB and lowest in control, however no. of nodule/ plant at 25 DAS did not vary significantly between combined inoculation of *Rhizobium* + PSB +AM Fungi with dual inoculation treatment. The interaction effect between the treatments having zero tillage with residue and inoculation of *Rhizobium* + PSB + AM Fungi was highest with respect to weight of root nodules/plant at 25, 35 and 45 DAS and no. of nodule/ plant at 35 DAS and lowest interaction value was noticed in zero tillage with no microbial inoculation. Inoculation with *Rhizobium* + PSB + AM Fungi supported with residue mulch in zero tillage might have positively impacted on root nodulation. Tagore *et al*. (2013) also reported that increasing P availability was due to the fact that PSB by virtue of their property of producing organic acids solubilize insoluble or fixed form of phosphorus in the rhizosphere and make it available to the growing plants, which promotes root development in plants. Supply of adequate amount of phosphorus is helpful in better crop growth and yield, along with enormous and better quality nodule formation in legumes by benefiting the current and succeeding crop also (Dongare *et al*., 2016).

Available N, P and K in soil

Available N, P and K in soil at crop harvest were significantly influenced by methods of crop establishment and microbial inoculants. Amongst three method of crop establishment,

Table 1: Influence of establishment methods and microbial inoculation on nodulation at different stage of green gram.

Treatment		Root nodule weight (mg/plant)			No. of nodules/plant		
		25 DAS	35 DAS	45 DAS	25 DAS	35 DAS	45 DAS
Method of crop establishment (ME)							
Conventional tillage		8.52	13.5	18.5	4.93	13.7	21.5
Zero tillage		7.65	12.2	17.2	4.23	10.4	18.7
Zero tillage with residue		10.58	15.5	20.4	5.51	15.8	23.0
SEm±		0.12	0.15	0.15	0.13	0.49	0.22
LSD (P=0.05)		0.49	0.53	0.60	0.52	1.91	0.85
Microbial inoculants (MI)							
Control (no inoculation)		8.19	12.9	17.9	4.26	12.4	19.9
Rhizobium+ PSB		8.93	13.7	18.6	5.02	13.4	20.8
Rhizobium+ PSB + AM Fungi		9.84	14.6	19.6	5.40	14.1	22.5
SEm±		0.13	0.14	0.15	0.14	0.20	0.37
LSD (P=0.05)		0.39	0.44	0.47	0.43	0.63	1.15
Interaction (EM × MI)							
Factor (B) at same level of A	SEm±	0.22	0.10	0.18	0.23	0.84	0.37
	LSD (P=0.05)	0.74	0.56	0.76	NS	1.33	NS
Factor (A) at same level of B	SEM (m)	0.22	0.15	0.21	0.24	0.56	0.57
	LSD (P=0.05)	0.74	0.50	0.71	NS	2.14	NS

zero tillage with residue treatment recorded highest available N, P and K in soil (218.31, 14.82 and 269.89 kg/ha, respectively) and available N, P and K of this treatment varied significantly over other methods (Table 2). The increase of N in soil having residue suggests that the N-supplying power of soil can be improved by returning straw to the soil and eliminating tillage (Malhi *et al*., 2011). De-Vita *et al*. (2007) reported that it was due to lower water evaporation, radiation insulation effect of residue and shedding effect on soil surface. Crop residue influenced the nutrients by providing the better micro climate and energy sources for soil micro-organisms. These microbes ultimately improved the soil aggregation.

Among microbial inoculation treatments combined inoculation of *Rhizobium* + PSB+ AM Fungi contributed highest to soil available N, P and K (217.19, 14.03 and 260.67 kg/ha, respectively) and lowest was documented in control (Table 2). Balai (2002) reported that seed treatment with PSB+ *Rhizobium* increased the soil N, P, K and S content after harvesting of mung bean crop. Yadav *et al*. (2017) reported that phosphorus concentration in soil after crop harvest increased significantly due to inoculation with PSB + VAM. It was accounted due to build-up of soil nutrients. Solubilization of phosphorus by bio fertilizers is attributed to extraction of acids like glutamic, succinic, lactic, oxalic, glyoxalic, malic, fumaric, a-ketobutric, propionic and formic acid. Some of these acids (hydroxyl-acid) may form chelates with cations such as Ca^{++} and Fe^{++} which resulted in effective solubilization of phosphates. In addition to phosphate solubilization, these microbes can mineralize organic phosphorus and render more P into soil solution than required for their own growth and metabolism, the surplus is for plant to absorb. This might be the fact that PSB inoculation increase availability of soil nutrients. The root system of legumes has capacity to solubilize soil phosphorus through excretion of amino acids and encourage the growth and multiplication of soil microbes which finally led to mineralization of unavailable P to available P in soil (Singh *et al*., 2016). Dadhich *et al.* (2006) also observed that co-inoculation of VAM along with PSB significantly improved nodulation, seed yield, mineral uptake and available P in soils.

Soil microbial parameters

The significant influence of methods of crop establishment and microbial inoculants on soil microbiological parameters *viz.,* dehydrogenase activity, alkaline phosphatase activity and microbial biomass carbon (MBC) at flowering stage of summer green gram was recorded. Among methods of crop establishment, zero tillage with residue produced maximum dehydrogenase activity, alkaline phosphatase activity and MBC in soil and significantly higher than other two treatments (Table 3). Choudhary and Behera (2014) reported that the microbial activity was significantly higher in zero tillage over convention tillage. Increased soil microbial and enzymatic activities in zero tillage systems have showed the consistency with the results of others researchers (Dong *et al*., 2009). An improvement in soil microbial and enzymatic activities under zero tillage practices might be due to better physico-chemical properties of soil. Furthermore, better soil aggregation helps in maintaining optimal moisture content as well as aeration in the soil. Dong *et al.* (2009) showed that after 5 years of zero till maize in Mexico, soil wet aggregate stability had increased over conventional tillage and it had higher soil enzymes, SOC and MBC. They concluded that no-till is a sustainable technology for crop production. Soil FDA hydrolysis is a measurement of the contribution of several enzymes, mainly involved in the decomposition of organic matter in soil. Hence, the higher the values of FDA hydrolysis are a sign of positive soil health and microbial activity. Regular and appropriate addition of crop residue have essential roles in improving the enzymatic activity of soil that are important for nutrient cycling as well as increasing crop productivity (Rajkumara *et al.*, 2014). Increase in enzymatic activity may be due to protection to the enzymes fraction upon increase in the soil humus content (Nath *et al.*, 2012).

Among microbial inoculation treatments, highest dehydrogenase activity, alkaline phosphatase activity and MBC in soil were found from treatment which received *Rhizobium* + PSB + AM Fungi. This treatment showed significantly higher alkaline phosphatase activity and microbial biomass carbon in soil than dual inoculation of *Rhizobium* + PSB (Table 3). There was significant influence of interaction between method of crop establishment and

Table 2: Influence of establishment methods and microbial inoculation on available N, P and K of soil at harvest of green gram.

Treatment		Grain yield (kg/ha)	Available N (kg/ha)	Available P (kg/ha)	Available K (kg/ha)
Method of crop establishment (ME)					
Conventional tillage		920.6	206.52	11.12	238.56
Zero tillage		674.7	212.20	12.56	255.89
Zero tillage with residue		968.6	218.31	14.82	269.89
SEm±		11.70	1.16	0.30	1.15
LSD (0.05)		40.20	4.56	1.21	4.56
Microbial inoculants (MI)					
Control (no inoculation)		762.9	200.52	11.01	249.89
Rhizobium + PSB		851.7	215.32	13.46	253.78
Rhizobium + PSB + AM Fungi		940.7	219.19	14.43	260.67
SEm ±		10.50	1.16	0.28	1.12
LSD (P=0.05)		33.70	3.58	0.89	3.42
Interaction (ME × MI)					
Factor (B) at same level of A	SEm±	14.04	1.43	0.34	1.17
	LSD (P=0.05)	NS	NS	NS	NS
Factor (A) at same level of B	SEm±	16.7	1.25	0.29	1.18
	LSD (P=0.05)	NS	NS	NS	NS

Initial N, P and K of soil 195.80, 10.50 and 230.00 kg/ha in conventional tillage, 198.00, 11.23 and 235.00 kg/ha in zero tillage and 200.00, 11.80 and 238.00 kg/ha in zero tillage with residue, respectively.

Table 3: Influence of establishment methods and microbial inoculation on soil microbiological parameters at flowering stage of green gram.

Treatment		Dehydrogenase activity (µg TPF g^{-1} soil 24 h^{-1})	Alkaline phosphatase activity (µg PNP produced g^{-1} soil h^{-1})	Microbial biomass carbon (µg g^{-1} of soil day^{-1})
Method of crop establishment (ME)				
Conventional tillage		8.98	9.58	140.88
Zero tillage		9.30	10.81	155.53
Zero tillage with residue		10.92	11.98	168.44
SEm±		0.27	0.09	0.76
LSD (0.05)		1.06	0.34	2.99
Microbial inoculants (MI)				
Control (no inoculation)		8.84	9.95	143.55
Rhizobium + PSB		10.12	10.88	154.41
Rhizobium + PSB + AM Fungi		10.24	11.55	166.88
SEm ±		0.23	0.09	0.68
LSD (P=0.05)		0.71	0.27	2.08
Interaction (ME × MI)				
Factor (B) at same level of A	SEm±	0.44	0.15	1.32
	LSD (P=0.05)	NS	0.52	4.07
Factor (A) at same level of B	SEm±	0.42	0.15	1.22
	LSD (P=0.05)	NS	0.52	4.25

Initial dehydrogenase activity- 7.50 µg TPF g^{-1} soil 24 h^{-1}, Alkaline phosphatase activity- 6.80 µg PNP produced g^{-1} soil h^{-1} and Microbial biomass carbon -110 µg g^{-1} of soil day^{-1}.

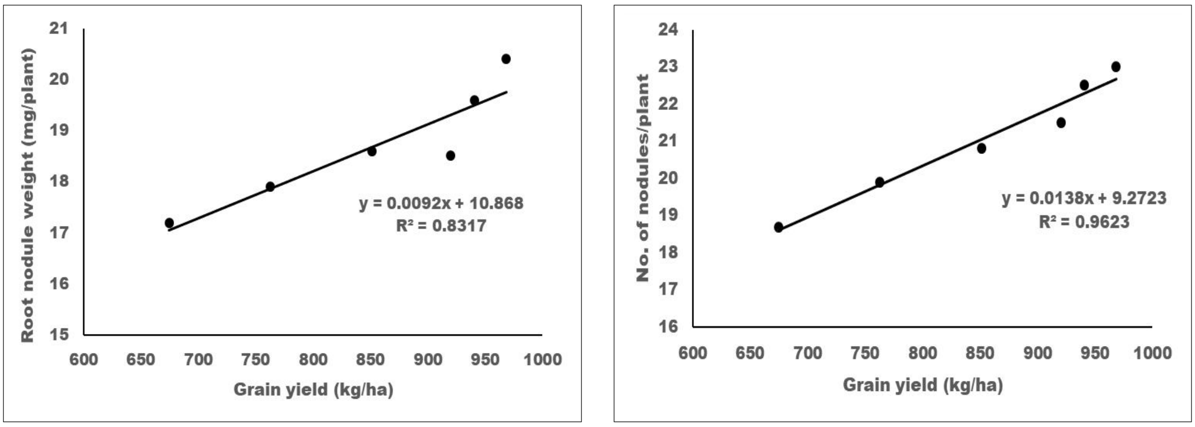

Fig 1: Relationship between grain yield and nodulation of green gram under the influence of crop establishment methods and microbial inoculation.

microbial inoculants on alkaline phosphatase activity and microbial biomass carbon in soil. The plot receiving zero tillage and residue with *Rhizobium* + PSB + AM Fungi showed highest alkaline phosphatase activity and microbial biomass carbon in soil and lowest were observed in conventional tillage along with control. Similar results were recorded by Rao *et al.* (2017).

Correlation between nodulation and grain yield

There was a highly positive correlation between grain yield and the weight of root nodules/plant and no. of nodule/ plant of green gram at 45 DAS (Fig 1). The correlation between grain yield and the weight of root nodules/plant and no. of nodule/ plant of green gram were 83.2% and 96.2%, respectively. Crop establishment methods along with microbial inoculation led to better root nodulation of summer green gram which helped in gaining higher yields.

CONCLUSION

Appropriate crop establishment practices and nutrient management practices are prerequisite for justifiable crop production without jeopardizing soil fertility and environmental health. The experiment showed that root growth parameters like nodule weight and no. of root nodules/ plant, available N, P and K in soil and soil microbial parameters like dehydrogenase activity, alkaline phosphatase activity and Microbial biomass carbon were significantly affected by method of crop establishment and microbial inoculants. These parameters were highest in zero tillage with residue treatment among method of crop establishment and in combined application of *Rhizobium* + PSB + AM among microbial inoculation treatments.

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