Journal of the Indian Society of Soil Science, Vol. 69, No. 2, pp 187-194 (2021)
DOI: 10.5958/0974-0228.2021.00029.3

Impact of Microbial Consortia on Microbial Population and Available Nutrients in Soil under Soybean Crop

Bablu Yaduwanshi, R.K. Sahu*, N.G. Mitra and B.S. Dwivedi
Department of Soil Science and Agricultural Chemistry, Jawaharlal Nehru Krishi Vishwa Vidhyalaya, Jabalpur, Madhya Pradesh, 482004, India

The present investigation was conducted to see the effect of microbial consortia on microbial population, available nutrients in soil and yield of soybean grown during 2018-19 under an ongoing All India Network Project on Soil Biodiversity and Biofertilizers at the Research Farm of Department of Soil Science and Agricultural Chemistry, Jawaharlal Nehru Krishi Vishwa Vidyalaya, Jabalpur, Madhya Pradesh, India. Coinoculation in the form of a consortium with *Pseudomonas fluorescens* (PGPR) *Actinomycetes (Actino) and Arthrobacter* (*Arthro*) was found beneficial in enhancing yield of soybean. Application of recommended dose of fertilizers (RDF) along with microbial consortium (PGPR+*Actino*+*Arthro*) recorded significantly higher amounts of available nitrogen (N), phosphorus (P) and potassium (K) which resulted in 43, 30 and 37 per cent, respectively over that of fertilized uninoculated control. Similarly, plots receiving RDF+PGPR+*Actino*+*Arthro* recorded significantly higher populations of *Actinomycetes*, *Arthrobacter* and PGPR at 25, 45 and 65 days after sowing which were 1.71, 1.42 and 1.52 log fold; 1.32, 1.28 and 1.31 log fold; and 1.56, 1.35 and 1.44 log fold, respectively over that of fertilized uninoculated control. In almost all cases, the treatment combinations of RDF+PGPR+*Arthro,* RDF+PGPR+*Actino* and RDF+*Arthro*+*Actino* exhibited similar performances. Seed and stover yields of soybean increased by 44 and 61 per cent, respectively over that of fertilized uninoculated control. It may be concluded that application of microbial consortia may be followed for enhancing soybean yield, available soil nutrients and microbial populations by direct as well as indirect beneficial effect.

Key words: Soybean, Actinomycetes, Arthrobacter and PGPR population

Soybean (*Glycine max)* is the most important oil seed crop in India. Nutritionally it contains 35-40% protein, 19% oil, 35% carbohydrate (of which 17% dietary fiber), 5% minerals and several other components including vitamins (Liu 1997). In Madhya Pradesh during *kharif* 2017, it occupies an area of 50.1 lakh ha with production of 42.0 lakh tonnes along with the productivity of 1086 kg ha^{-1} (SOPA 2019). It is evident that seed and stover yields of soybean are significantly affected by seed inoculation with biofertilizers supplemented with starter dose of inorganic fertilizers (Amule *et al.* 2018 a,b). Different beneficial microorganisms used for inoculations are of variable nature and characteristics. *Arthrobacter* is a free living diazotroph. It is a gram positive obligate aerobe exists in rod shape during exponential growth phase and cocci during stationary phase or under moisture stress conditions. While, *Pseudomonas fluorescens* is a gram negative, rod shape, aerobic, flagellated motile root colonizing bacterium and known as plant growth promoting rhizobacteria (PGPR) which influences growth, yield and nutrient uptake mediated by an array of mechanisms. Some bacterial strains directly regulate plant physiology by mimicking synthesis of plant hormones, formation of antibiotics and siderophores; whereas others increase availability of minerals and nitrogen (N) in the soils as a way to augment plant growth. These bacteria contribute to improve soil health and are metabolically active and diverse. Moreover, the isolates can exhibit more than two or three PGPR traits which may promote plant growth directly or indirectly or synergistically (Yasmin *et al*. 2007), thereby, increases plant growth and yield of the crop. The isolated strains of *Pseudomonas fluorescens* and *Pseudomonas putida* have been acknowledged significantly to increase the growth and yield of different crops including legumes. The *Actinomycetes*

*Corresponding author (Email: rakesh_sahujbp@yahoo.co.in)

are a versatile group of microorganisms widely distributed in arable dry soils that influenced by soil temperature, soil type, soil pH, organic matter content, cultivation practices, aeration and moisture content. The actinobacteria are documented for important activities in soils like production of growth promoting substances, phosphorus (P) solubilization, decomposition of organic matter, production of antibiotics and suppression of soil borne plant pathogens. In view to the beneficial aspects of the microorganisms, the present experiment was carried out to explore the effect of consortia of beneficial microorganisms on yield of soybean, microbial population and nutrients status in soil.

Materials and Methods

The experiment was conducted during 2018-19 at experimental field of the Department of Soil Science and Agricultural Chemistry, Jawaharlal Nehru Krishi Vishwa Vidyalaya, Jabalpur, Madhya Pradesh, India. The soil of the experimental site belongs to Kheri series of fine montmorillonite, Hyperthermic family of Typic Haplusterts popularly known as "black cotton soil" or Vertisols. The mean rainfall during the crop period was 1163.4 mm. The average maximum temperature during the month of June to November was between 16.8 to 41.6 °C. Occasional winter rain occurs during December and/or January.

The experiment consisted of nine treatments with 3 replications were carried out in a randomized block design. The treatments were:

T1 : Recommended dose of fertilizer + *Actinomycetes* (RDF+*Actino*)
T2 : RDF + *Arthrobactor* (RDF+*Arthro*)
T3 : RDF + Plant growth promoting rhizobacteria (RDF+PGPR)
T4 : RDF + Actinomycetes + *Arthrobactor* (RDF+ *Actino*+*Arthro*)
T5 : RDF + PGPR + *Actinomycetes* (RDF+ PGPR+*Actino*)
T6 : RDF + PGPR + *Arthrobactor* (RDF+ PGPR+*Arthro*)
T7 : RDF + PGPR + *Actinomycetes* + *Arhtrobacto*r (RDF+ PGPR+*Actino*+*Arthro*)
T8 : Fertilized uninoculated (FUI)
T9 : Unfertilized uninoculated (UFUI)

The recommended dose of fertilizers (RDF) applied were 20, 80 and 20 kg ha^{-1} of N, P_2O_5 and K_2O, respectively. Consortia of liquid inoculants like *Actinomycetes*, *Arthrobactor* and *Pseudomonas fluorescens* (as PGPR) were used in different combinations for seed treatment. Soybean (cv. 9752) was sown on July 7, 2018 and harvested on 12 October, 2018.

Initial surface (0-15 cm depth) soil sample was drawn from 10 different places from the entire experimental field and mixed together to form a representative composite sample. After the harvest of crop, plot-wise soil samples (0-15 cm) were collected, air-dried and processed for further chemical analysis. The available N was determined following alkaline $KMnO_4$ method (Subbiah and Asija 1956). Available P was extracted with 0.5 *M* $NaHCO_3$ solution (8.5 pH) and the P content in the extract was estimated spectrophotometrically (Olsen *et al.* 1954). Available potassium (K) was determined by extraction of soil with neutral normal ammonium acetate (pH 7.0 in 1:5, soil: solution ratio) and estimated with the help of flame photometer (Jackson 1973). Fresh soil samples were used as far as possible without grinding, sieving or any modifications for population counts of *Actinomycetes*, *Arthrobacter* and PGPR (*Pseudomonas fluorescens*) in rhizosphere soil. The samples were stored in low density polyethylene bags. Soil samples were collected from the rhizospheric soil (0-15 cm) at 25, 45 and 65 days after sowing (DAS) of soybean crop for microbial population counts. Soil-water serial dilution was carried out by suspending 10 g of fresh soil sample in 90 mL sterilized water and shaken thoroughly which resulted in 10^{-1} dilution. Subsequent serial dilutions were made up to 10^{-9} dilution levels.

Plating (pour method)

Inoculation in sterilized petri-plates were done by taking 1 mL each of 10^{-1} to 10^{-9} dilutions as required for *Actinomycetes*, *Arthrobactor* and PGPR population counts in soil. Inoculation in petri-plates was performed in triplicate from aliquots of each dilution. The standard media compositions for *Actinomycetes*, *Arthrobactor* and PGPR population counts are given in table 1.

The serial dilutions obtained from soil samples at 25, 45 and 65 DAS of soybean were used for plating adopting pour plate method. Sterilized water (9.0 mL) was taken in test tube along with soil (1.0 g). Then 1.0 mL of first diluents was transferred and shaken uniformly by rolling the tube between palms of hand to provide horizontal shaking. The series in similar manner to get up to 10^9 dilution levels were prepared and marked properly for the dilutions on the test tubes. Then, 1 mL from each dilution was transferred onto petri-plates. After that 15 mL of melted media was poured aseptically under laminar air flow chamber. Plates were rotated gently clock and anti-clock wise

Table 1. Media composition for population counts of PGPR (*Pseudomnas fluorescens*), *Actinomycetes* and *Arthrobacter*

Chemical /reagents	Media composition L^{-1}		
	King's medium B (KMB) (for PGPR - *P. fluorescence*)	Caseinate medium (for Actinomycetes)	Luria-Bertani (for *Arthrobactor*)
KH_2PO_4	1.5 g	0.50 g	-
$MgSO_4.7H_2O$	1.5 g	0.20 g	-
$FeCl_3$	-	0.01 g	-
Sodium caseinate	-	0.20 g	-
Beef extract	-	-	1.0 g
Glycerol	10.0 mL	-	-
Peptone	10.0 g	-	5.0 g
Yeast extract	-	-	2.0 g
NaCl	-	-	5.0 g
Agar-agar	20.0 g	15.0 g	15.0 g
Distilled water	1000 mL	1000 mL	1000 mL

to mix the soil dilution within media. After solidification of media the plates were inverted and incubated at 28±2 °C for developing colonies of the respective microorganisms on media. After 24 h, every day the characteristic growth was observed and the number of colonies were counted on 3-4 days.

Viable cells (cfu g^{-1} soil) =

$$\frac{\text{Number of colonies}}{\text{Oven dry weight of soil (1.0 g)}} \times \text{dilution factor}$$

Statistical analysis

Statistical analysis was performed by the Windows-based SPSS program. The SPSS procedure was used for analysis of variance to determine the statistical significance of the treatments of microorganisms on various attributes of soybean. The differences between means of the different treatments were compared by the least significant difference (LSD) at 5% level of significance.

Results and Discussion

Data on available N, P and K status in soil (0-15 cm) after the harvest of soybean crop are presented in table 2. Results showed that significantly maximum amount of available N (286.1 kg N ha^{-1}) was observed in plots receiving RDF along with microbial consortium of PGPR+*Actino*+*Arthro* (T_7) which was 43 per cent higher than that of fertilized uninoculated plot (199.0 kg N ha^{-1}). This was followed by the performance of treatments receiving RDF+PGPR+ *Arthro* (T_6), RDF+PGPR+*Actino* (T_5), RDF+*Actino* + *Arthro* (T_4) and RDF+PGPR (T_3) with available N in soil of 257.1, 251.7, 242.5 and 236.4 kg N ha^{-1} corresponding to 29.2, 26.5, 21.9 and 18.8 per cent greater responses, respectively over fertilized uninoculated plot. This increase in available N content might be attributed to the greater multiplication of soil microbes which converted organically bound N to inorganic form as reported by Katkar *et al.* (2006)

Table 2. Effect of microbial consortia on content of available nutrients of N, P and K in soil (0-15 cm depth) at harvest of soybean

Treatment	Available nutrient content (kg ha^{-1})		
	N	P	K
T_1: RDF+*Actinomycetes*	217.7	18.0	250.0
T_2: RDF+*Arthrobactor*	224.7	17.7	259.3
T_3: RDF+PGPR	236.4	18.6	266.2
T_4: RDF+Actinomycetes+*Arthrobactor*	242.5	19.4	277.0
T_5: RDF+PGPR+*Actinomycetes*	251.7	20.3	298.1
T_6: RDF+PGPR+*Arthrobactor*	257.1	21.3	314.9
T_7: RDF+PGPR+*Actinomycetes*+*Arhtrobactor*	286.1	22.6	331.3
T_8: Fertilized uninoculated (FUI)	199.0	17.4	242.4
T_9: Unfertilized uninoculated (UFUI)	186.0	16.2	231.2
Mean	233.5	19.1	274.5
SEm±	12.4	1.2	20.7
LSD (P=0.05)	37.1	3.8	62.1

and Khandagle *et al.* (2020). The production of growth promoting substances and high colonization ability of rhizobacteria such as *Pseudomonas* enhanced the N_2 fixation of soybean when co-inoculated with *Bradyrhizobium japonicum* (Chebotar *et al.* 2001). Sivasakthi *et al.* (2014) reported the ability of actinomycetes such as *Frankia* to fix atmospheric N_2 and increase the available N in soil.

The highest content of available P in soil (22.6 kg P_2O_5 ha^{-1}) was recorded with the application of RDF+PGPR+*Actino*+*Arthro* (T_7) as compared to that of fertilized uninoculated plot (17.4 kg P_2O_5 ha^{-1}) and it had a greater response by ~30 per cent over control followed by the effect of treatment combination of RDF+PGPR+*Arthro* (T_6) with available P of 21.3 kg P_2O_5 ha^{-1} with ~22.0 per cent greater response over control plot. Similar findings were reported by Sarkar *et al.* (2002) where they reported that actinomycetes along with PGPR and *Arthrobacter* were capable of solubilizing tricalcium phosphate (TCP), while rock phosphate (RP) was found less soluble than TCP. Henderson and Duff (1963) reported that certain bacteria, fungi and actinomycetes are able to solubilize P-minerals into soluble form due to several mechanisms such as enzymatic oxidation-reduction reactions, formation of chelates and complexes with proteins, amino acids, organic acids, *etc*. The *Actinobacteria*, *Arthrobacter* and PGPR help in influencing the soil fertility through the involvement of many components and serve as nutrient enhancer. Besides producing siderophores and solubilizing P, they are known to produce cocktail of enzymes which include amylase, chitinase, cellulase, invertase, lipase, keratinase, peroxidase, pectinase, protease, phytase and xylanase which make the complex nutrients into simple mineral forms. This nutrient cycling capacity makes them as an ideal microflora as natural fertilizers. Many soil bacteria such as *Pseudomonas, Azospirillum, Bacillus, Rhizobium, Burkholderia, Arthrobacter, Enterobacter, Flavobacteriu and Erwinia*; while, fungi especially *Aspergillus*, *Penicillium* and *Trichoderma* have the ability to solubilize P and make it available to plants (Rodrýìguez and Fraga 1999; Oberson *et al.* 2001). The maximum available K content of 331.3 kg K ha^{-1} in soil at surface layer was found statistically superior with application of RDF+PGPR+*Actino*+*Arthro* (T_7) by ~37.0 per cent as compared to fertilized uninoculated control (242.4 kg K ha^{-1}), followed by the effect from RDF+PGPR+*Arthro* with P content of 314.9 kg ha^{-1} along with ~30.0 per cent greater response over that of fertilized uninoculated control. Similar findings were reported by Patel *et al.* (2018).

Population of Actinomycetes in rhizospheric soil at different growth stages of soybean

The data on population of actinomycetes in rhizospheric soil counted at 25, 45 and 65 DAS of the crop growth are given in table 3. The data on actinomycetes population in rhizospheric soil at 25 DAS revealed that the treatment combination RDF+PGPR+*Actino*+*Arthro* (T_7) responded maximum population of 5.02 log cfu (10.6×10^4 cfu g^{-1} soil) with the relative response of 1.71 log fold increase over the fertilized uninoculated plot, followed by the response of RDF+PGPR+*Actino* (T_5), RDF+Actino+*Arthro* (T_4) and RDF+PGPR+*Arthro* (T_6) by 5.01 log cfu (10.3×10^4 cfu g^{-1} soil), 5.00 log cfu (10.0×10^4 cfu

Table 3. Effect of microbial consortia on population of actinomycetes in rhizospheric soil at different growth stages of soybean

Treatments	Population of Actinomycetes		
	25 DAS	45 DAS	65 DAS
T_1: RDF+*Actinomycetes*	3.97 (9.4×10^3)	6.02 (10.6×10^5)	5.01 (10.1×10^4)
T_2: RDF+*Arthrobactor*	3.94 (8.8×10^3)	6.01 (10.2×10^5)	4.98 (9.7×10^4)
T_3: RDF+PGPR	3.97 (9.4×10^3)	6.01 (10.4×10^5)	4.99 (9.9×10^4)
T_4: RDF+*Actinomycetes*+*Arthrobactor*	5.00 (10.0×10^4)	7.05 (11.3×10^6)	6.03 (10.6×10^5)
T_5: RDF+PGPR+*Actinomycetes*	5.01 (10.3×10^4)	7.07 (11.9×10^6)	6.06 (11.6×10^5)
T_6: RDF+PGPR+*Arthrobactor*	4.99 (9.9×10^4)	7.04 (11.0×10^6)	6.01 (10.4×10^5)
T_7: RDF+PGPR+*Actinomycetes*+*Arhtrobactor*	5.02 (10.6×10^4)	7.08 (12.2×10^4)	6.06 (11.7×10^5)
T_8: Fertilized uninoculated (FUI)	2.92 (8.5×10^2)	4.98 (9.7×10^4)	3.978 (9.6×10^3)
T_9: Unfertilized uninoculated (UFUI)	2.57 (8.3×10^2)	4.97 (9.6×10^4)	3.95 (9.1×10^3)
Mean	4.19	6.25	5.23
SEm±	0.48	0.45	0.34
LSD (P=0.05)	1.45	1.36	1.02

Note: RDF = Recommended dose of fertilizers; PGPR = Plant growth promoting rhizobacteria

The values outside parentheses are in log cfu g^{-1} soil from log transformation of exponential values given in parentheses in cfu g^{-1} soil (oven dry basis) from plate counts

g^{-1} soil) and 4.99 log cfu (9.94×10^4 cfu g^{-1} soil), respectively with the responses of 1.71, 1.71 and 1.70 log folds increase over the fertilized uninoculated plot (2.92 log cfu = 8.53×10^2 cfu g^{-1} soil). At 45 DAS of crop growth, the maximum actinobacterial response of 7.08 log cfu (12.2×10^6 cfu g^{-1} soil) was found in plots receiving RDF+PGPR+*Actino*+*Arthro* (T_7) which was 1.42 log fold increase over the control (T_8). This was followed by the effect of RDF+PGPR+*Actino* (T_9), RDF+*Actino*+*Arthro* (T_4) and RDF+PGPR+*Arthro* (T_6) with the population of 7.07 log cfu (11.9×10^6 cfu g^{-1} soil), 7.05 log cfu (11.3×10^6 cfu g^{-1} soil) and 7.04 log cfu (11.0×10^6 cfu g^{-1} soil) representing 1.41 1.40 and 1.40 log folds responses, respectively as compared to the fertilized uninoculated plot (4.98 log cfu = 9.72×10^4 cfu g^{-1} soil).

At 65 DAS of crop growth, the actinomycetes population in rhizospheric soil due to application of treatment combination of RDF+PGPR+*Actino*+*Arthro* (T_7) responded maximum population of 6.06 log cfu (11.7×10^5 cfu g^{-1} soil) with the response of 1.52 log fold increase over the fertilized uninoculated plot. This was followed by the response of RDF+PGPR+*Actino* (T_5), RDF+*Actino*+*Arthro* (T_4) and RDF+PGPR+*Arthro* (T_6) for the actinomycetes population of 6.06 log cfu (11.6×10^5 cfu g^{-1} soil), 6.03 log cfu (10.6×10^5 cfu g^{-1} soil) and 6.01 log cfu (10.4×10^5 cfu g^{-1} soil), respectively corresponding to 1.52, 1.51 and 1.51 log fold increase over the fertilized uninoculated plot (3.97 log cfu = 9.64×10^3 cfu g^{-1} soil). Similar range of actinomycetes population of 5×10^5 cfu g^{-1} of dry soil sampled from most of the plots was recorded by Sahur *et al.* (2018). The seed inoculation with *Pseudomonas* at 3 g kg^{-1} showed the maximum number of actinomycetes counts of 11.4×10^5 cfu g^{-1} of soil which was 13.9 per cent greater over that of control. Mishra *et al.* (2014) reported that significant increase in actinomycetes population in comparison to control the treatments.

Population of PGPR (Pseudomonas sp.) in rhizospheric soil at different growth stages

The data on PGPR (*Pseudomonas* sp.) population in rhizospheric soil at 25 DAS of soybean (Table 4) revealed that the treatment combination of RDF+PGPR+Actino+*Arthro* (T_7) responded maximum PGPR population of 8.89 log cfu (78.9×10^7 cfu g^{-1} soil) with 1.32 log fold increase, followed by the response of RDF+PGPR+*Arthro* (T_6) by 8.85 log cfu (71.5×10^7 cfu g^{-1} soil) with 1.31 log fold response over that of fertilized uninoculated plot (6.72 log cfu = 53.6×10^5 cfu g^{-1} soil). At 45 DAS, the PGPR population in rhizospheric soil exhibited maximum due to RDF+PGPR+*Actino*+*Arthro* (T_7) treated soil with 10.13 log cfu (134.6×10^8 cfu g^{-1} soil) and 1.28 log fold response, followed by the influence of RDF+PGPR+*Arthro* (T_6) and RDF+PGPR+*Actino* (T_5) for the population of 10.09 log cfu (123.6×10^8 cfu g^{-1} soil) and 10.06 log cfu (114.2×10^8 cfu g^{-1} soil) along with the responses of 1.28 and 1.27 log folds, respectively over that of fertilized uninoculated plot (7.86 log cfu = 72.3×10^6 cfu g^{-1} soil).

At 65 DAS, the population of PGPR in rhizospheric soil was increased to 8.93 log cfu (85.6×10^7 cfu g^{-1} soil) which was 1.31 log fold higher relative to that of fertilized uninoculated plot due to application of RDF+PGPR+*Actino*+*Arthro* (T_7),

Table 4. Effect of microbial consortia on population of PGPR (*Pseudomonas* sp.) in rhizospheric soil at different growth stages of soybean

Treatment	Population of *Pseudomonas* sp.		
	25 DAS	45 DAS	65 DAS
T_1: RDF+*Actinomycetes*	6.78 (61.4×10^5)	7.91 (81.5×10^6)	7.86 (73.8×10^6)
T_2: RDF+*Arthrobactor*	6.79 (64.5×10^5)	8.97 (92.3×10^7)	7.85 (71.4×10^6)
T_3: RDF+PGPR	6.80 (67.2×10^5)	8.99 (97.1×10^7)	7.87 (74.5×10^6)
T_4: RDF+*Actinomycetes*+*Arthrobactor*	7.81 (66.0×10^6)	9.02 (103.8×10^7)	8.88 (76.0×10^7)
T_5: RDF+PGPR+*Actinomycetes*	7.83 (67.6×10^6)	10.06 (114.2×10^8)	8.89 (77.7×10^7)
T_6: RDF+PGPR+*Arthrobactor*	8.85 (71.5×10^7)	10.09 (123.6×10^8)	8.91 (81.5×10^7)
T_7: RDF+PGPR+*Actinomycetes*+*Arhtrobactor*	8.89 (78.9×10^7)	10.13 (134.6×10^8)	8.93 (85.6×10^7)
T_8: Fertilized uninoculated (FUI)	6.72 (53.6×10^5)	7.86 (72.3×10^6)	6.80 (63.5×10^5)
T_9: Unfertilized uninoculated (UFUI)	6.61 (44.9×10^5)	7.82 (66.4×10^6)	6.79 (62.2×10^5)
Mean	7.47	8.98	8.08
SEm±	0.45	0.50	0.50
LSD (P=0.05)	1.37	1.49	1.50

Note: The values outside parentheses are in log cfu g^{-1} soil from log transformation of exponential values given in parentheses in cfu g^{-1} soil (oven dry basis) from plate counts

followed by the effect of RDF+PGPR+*Arthro* (T_6) and RDF+PGPR+*Actino* (T_5) for the population of 8.93 log cfu (85.6×10^7 cfu g^{-1} soil) and 8.89 log cfu (77.7×10^7cfu g^{-1} soil) with the respective responses of 1.31 and 1.30 log fold over that of fertilized uninoculated plot (6.80 log cfu = 63.5×10^5 cfu g^{-1} soil). Similar pattern of variation in *Pseudomonas* population was also published by Kachhap *et al.* (2015) where they reported that up to 75 DAS (complete flowering stage) the population of *Pseudomonas* sp. was gradually increased from log 5.6 cfu g^{-1} dry soil to log 6.3 cfu g^{-1} dry soil and then it started declining. *Pseudomonas* population in soil ranged from 6.5 to 8.02×10^4 cfu g^{-1} of soil. Seed inoculation with *Pseudomonas* gave the maximum number of *Pseudomonas* population and showed significant increase in comparison to control treatment (Mishra *et al*. 2014; Dwivedi *et al.* 2016).

Population of Arthrobacter sp. in rhizospheric soil at different growth stages

The data on *Arthrobacter* population in rhizospheric soil of soybean (Table 5) showed that at 25 DAS, the consortium of RDF+PGPR+*Actino*+*Arthro* (T_7) responded maximum bacterial population of 6.22 log cfu (17.0×10^5 cfu g^{-1}soil) with 1.56 log fold response over fertilized uninoculated plot (3.64 log cfu = 9.56×10^3 cfu g^{-1} soil). At 45 DAS, the *Arthrobacter* population under RDF+PGPR+Actino+*Arthro* plot (T_7) was maximum with 8.30 log cfu (20.3×10^7 cfu g^{1} soil) which exhibited 1.62 log fold response over fertilized uninoculated plot. It was followed by plots receiving RDF+PGPR+*Arthro* (T_6), RDF+*Arthro*+*Actino* (T_4) and RDF+PGPR+*Actino* by 7.26 log cfu (16.0×10^6 cfu g^{-1} soil), 7.23 (18.4×10^6 cfu g^{-1} soil) and 7.20 (17.0×10^6 cfu g^{-1} soil) corresponding to 1.42, 1.41 and 1.40 log folds, respectively as compared to that of fertilized uninoculated plot (5.11 log cfu = 13.0×10^4 cfu g^{-1} soil).

Similarly, the same treatment of RDF+PGPR+*Actino*+*Arthro* (T_7) at 65 DAS recorded the best for the population of 7.25 log cfu (18.0×10^6 cfu g^{-1} soil) exhibiting 1.43 log fold, followed by the effects from RDF+PGPR+*Arthro* (T_6), RDF+*Actino*+*Arthro* (T_4) and RDF+PGPR+*Actino* (T_5) having population of 6.21 log cfu (16.4×10^5 g^{-1} soil), 6.18 log cfu (15.3×10^5 g^{-1} soil) and 6.15 log cfu (14.3×10^5 g^{-1} soil) with the responses of 1.23, 1.22 and 1.22 log fold, respectively over fertilized uninoculated plot (5.04 log cfu = 10.8×10^3 g^{-1} soil). These rhizosphere bacteria enhance growth of plant and yield by enhancing N_2-fixation, solubilization of P, production of phytohormones such as auxins (IAA), cytokinins and gibberellins, sequestering of iron by production of siderophores and lowering of ethylene concentration. Nitrogen fixing and P-solubilizing bacteria are of great importance for plant nutrition by increasing N and P uptake of the plants. Combined inoculation consistently enhanced growth and yield of crop to a level equal to or greater than that achieved by single inoculation and far greater than that of the uninoculated control plots. This might be due to enhanced colonization of root hairs and cortical cells that improved root surface area and architecture for better acquisition of nutrients as well as plant hormones.

Table 5. Effect of microbial consortia on population of *Arthrobacter* sp. in rhizospheric soil at different growth stages of soybean

Treatment	Population of *Arthrobacter* sp.		
	25 DAS	45 DAS	65 DAS
T_1: RDF+*Actinomycetes*	4.03 (11.0×10^3)	6.16 (14.7×10^5)	5.09 (12.3×10^4)
T_2: RDF+*Arthrobactor*	4.93 (12.4×10^3)	6.21 (16.3×10^5)	5.12 (13.3×10^4)
T_3: RDF+PGPR	4.05 (11.3×10^3)	6.17 (15.0×10^5)	5.09 (12.3×10^4)
T_4: RDF+*Actinomycetes*+*Arthrobactor*	5.15 (14.3×10^4)	7.20 (17.0×10^6)	6.18 (15.3×10^5)
T_5: RDF+PGPR+*Actinomycetes*	5.12 (13.3×10^4)	7.26 (16.0×10^6)	6.15 (14.3×10^5)
T_6: RDF+PGPR+*Arthrobactor*	5.18 (15.4×10^4)	7.23 (18.4×10^6)	6.21 (16.4×10^5)
T_7: RDF+PGPR+*Actinomycetes*+*Arhtrobactor*	6.22 (17.0×10^5)	8.30 (20.3×10^7)	7.25 (18.0×10^6)
T_8: Fertilized uninoculated (FUI)	3.98 (9.6×10^3)	6.11 (13.0×10^5)	5.04 (10.8×10^3)
T_9: Unfertilized uninoculated (UFUI)	3.93 (8.4×10^3)	6.07 (12.0×10^5)	4.02 (9.3×10^3)
Mean	4.64	6.53	5.58
SEm±	0.44	0.36	0.40
LSD (P=0.05)	1.44	1.09	1.19

Note: The values outside parentheses are in log cfu g^{-1} soil from log transformation of exponential values given in parentheses in cfu g^{-1} soil (oven dry basis) from plate counts

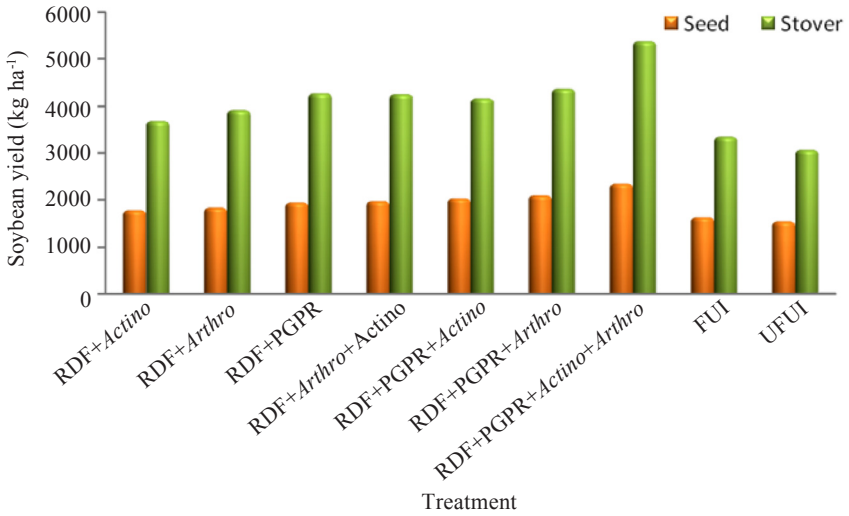

Fig.1. Seed and stover yield (kg ha^{-1}) of soybean

Seed and stover yields of soybean

The data on grain and stover yield of soybean (Fig.1) revealed that the grain yield of soybean differed significantly among all the treatments. The consortium of RDF+PGPR+*Actino*+*Arthro* (T_7) boosted significantly the grain yield to a maximum of 2350 kg ha^{-1} with 44 per cent more response over that from fertilized uninoculated plot (1636 kg ha^{-1}), followed by the performance from RDF+PGPR+*Arthro* (T_6), RDF+PGPR+*Actino* (T_5), RDF+*Actino*+*Arthro* (T_4) and RDF+PGPR (T_3) for the grain yield of 2102, 2037, 1978 and 1947 kg ha^{-1} along with the increments of 28, 25, 21 and 19 per cent, respectively over control. This increment in yields of soybean with the treatments of inoculation might be attributed to better nodulation, N_2 fixation and crop growth as against uninoculated control (Brahmaprakash *et al.* 2004; Gupta 2005; Dwivedi and Dwivedi 2016). Afzal *et al.* (2010) reported that coinoculation of *Bradyrhizobium* and *Pseudomonas* strains along with supplementation of fertilizer P enhanced the grain yield of soybean by 38 per cent in pot experiments and 12 per cent in the field experiments relative to that from P alone.

Similarly, the highest stover yield of soybean (5381 kg ha^{-1}) was recorded with the treatment combination of RDF+PGPR+*Actino*+*Arthro* (T_7) which was 61 per cent more over fertilized uninoculated plot (3345 kg ha^{-1}), followed by the effect of RDF+PGPR+*Arthro* (T_6) for the stover yield of 4369 kg ha^{-1} and 31 per cent more over control. The finding were in support to that published by Amule *et al.* (2018 a,b) and Meshram *et al.* (2018) where they found that inoculation of microbial consortium (actinomycetes, *Rhizobium* and PGPR) supplemented with recommended dose of fertilizers resulted in significant improvement in seed and stover yields of soybean over fertilized uninoculated.

Conclusions

From the above results, it may be concluded that application of consortium RDF+PGPR+*Actinomycetes*+*Arthrobacter* as seed inoculation performed the best consortium for enhancing yields of seed and stover of soybean. Similarly, for soil parameters, the same treatment combination of RDF+PGPR+*Actinomycetes*+*Arthrobacter* performed significantly better towards available nutrient contents of N, P and K and rhizospheric microbial populations at 25, 45 and 65 days after sowing with respect to *Actinomycetes*, *Pseudomonas* and *Arthrobacter*. Thus, it may be concluded that inoculation of the soybean seeds with microbial consortia may be followed for enhancing yield of soybean, nutrient availability and microbial population.

References

Afzal, A., Bano, A. and Fatima M. (2010) Higher soybean yield by inoculation with N-fixing and P-solubilizing bacteria. *Agronomy for Sustainable Development* **30**, 487-495.

Amule, F.C., Sirothiya, P., Rawat, A.K. and Mishra, U.S. (2018b) Efficacy of actinomycetes *Rhizobium* and plant growth promoting rhizobacteria consortium inoculants on symbiotic traits nodule leghemoglobin and yield of soybean. *International Journal of Chemical Studies* **6**, 593-596.

Amule, F.C., Sirothiya, P., Rawat, A.K. and Mishra, U.S. (2018a) Effect of actinobacterial *Rhizobium* and plant growth promoting rhizobacteria consortium inoculation on rhizosphere soil properties in soybean

in Jabalpur district of Madhya Pradesh. *International Journal of Chemical Studies* **6**, 583-586.

Brahmaprakash, G.P., Girisha, H.C., Navi Vithal and Hedge, S.V. (2004) Biological nitrogen fixation in pulse crops. In *Pulses in New Perspective* (Masood Ali, B.B. Singh, Shiv Kumar and Vishwa Dhar, Eds.) *Indian Society of Pulses Research and Development, Indian Institute of Pulse Research, Kanpur, India,* pp. 271-286.

Chebotar, V.K., Asis, C.A. and Akao, S. (2001) Production of growth-promoting substances and high colonization ability of rhizobacteria enhance the nitrogen fixation of soybean when coinoculated with *Bradyrhizobium japonicum*. *Biology and Fertility of Soils* **34**, 427-432.

Dwivedi, A.K. and Dwivedi, B.S. (2016) Impact of long term fertilizer management for sustainable soil health and crop productivity: Issues and challenges. *JNKVV Research Journal* **50**, 125-139.

Dwivedi, B.S., Rawat, A.K., Dixit, B.K. and Thakur, R.K. (2016) Effect of inputs integration on yield, uptake and economics of kodo millet (*Paspalum scrobiculatum* L). *Economic Affairs* **61**, 519-526.

Gupta, S.C. (2005) Evaluation of liquid and carrier based *Rhizobium* inoculants in chickpea. *Indian Journal of Pulses Research* **18**, 40-42.

Henderson, M.E. and Duff, R.B. (1963) The release of metallic and silicate ions from minerals rocks and soils by fungal activity. *Journal of Soil Science* **14**, 236-246.

Jackson, M.L. (1973) *Soil Chemical Analysis*. Prentice Hall of India (Pvt.) Ltd., New Delhi.

Kachhap, S., Chaudhary, A. and Singh, S.D. (2015) Response of plant growth promoting rhizobacteria (PGPR) in relation to elevated temperature conditions in groundnut (*Arachis hypogaea* L.) *The Ecoscan* **9**, 771-778.

Katkar, R.N., Turkhede, A.B. and Wankhade, S.T. (2006) Residual effect of organic manures and inorganic fertilizers on soil properties and seed cotton yield. *PKV Research Journal* **30**, 36-41.

Khandagle, A., Dwivedi, B.S., Dwivedi, A.K., Panwar, S. and Thakur, R.K. (2020) Nitrogen fractions under long-term fertilizer and manure applications in soybean–wheat rotation in a Vertisol. *Journal of the Indian Society of Soil Science* **68**, 186-193.

Liu, Z.B., Hagen, C. and Cuilfoyle, T.J. (1997) A G-box-binding protein from soybean binds to the E1 auxin-response element in the soybean CH_3 promoter and contains a proline-rich repression domain. *Plant Physiology* **11**, 397-407.

Meshram, M.K., Dwivedi, B.S., Naik, K.R., Thakur, R.K. and Keram, K.S. (2018) Impact of organic and inorganic sources of nutrients on yield, nutrient uptake, soil fertility and economic performance of rice in a Typic Haplustert. *Journal of Soils and Crops* **28**, 31-36.

Mishra, G., Kumar, N., Giri, K., Pandey, S. and Kumar, R. (2014) Effect of fungicides and bioagents on number of microorganisms in soil and yield of soybean *(Glycine max)* Nusantara. *Bioscience* **6**, 45-48.

Oberson, A., Friesen, D.K., Rao, I.M., Bühler, S. and Frossard, E. (2001) Phosphorus transformations in an Oxisol under contrasting land-use systems: the role of the soil microbial biomass. *Plant and Soil* **237**, 197-210.

Olsen, S.R., Cole, C.V., Watanabe, F.S. and Dean, L.A. (1954) Estimation of available phosphorus in soils by extraction with sodium bicarbonate. *United States Department of Agriculture* Circular No. 939.

Patel, Gajendra, Dwivedi, B.S., Dwivedi, A.K., Thakur, Risikesh and Singh, Muneshwar (2018) Long-term effect of nutrient management on soil biochemical properties in a Vertisol under soybean–wheat cropping sequence. *Journal of the Indian Society of Soil Science* **66**, 215-221.

Rodrýìguez, H. and Fraga, R. (1999) Phosphate solubilizing bacteria and their role in plant growth promotion. *Biotechnology Advances* **17**, 319-339.

Sahur, A., Ala, A., Patandjengi, B. and Syamun, E. (2018) Effect of seed inoculation with Actinomycetes and *Rhizobium* isolated from indigenous Soybean and rhizosphere on nitrogen fixation growth and yield of Soybean. *International Journal of Agronomy* vol. 2018: 1-7, Article ID 4371623.

Sarkar, P.K., Florczyk, M.A., McDonough, K.A. and Nag, D.K. (2002) SSP2, a sporulation-specific gene necessary for outer spore wall assembly in the yeast *Saccharomyces cerevisiae*. *Molecular Genetics and Genomics* **267**, 348-358.

Sivasakthi, S., Usharani, G. and Saranraj, P. (2014) Biocontrol potentiality of plant growth promoting bacteria (PGPR) - *Pseudomonas fluorescens* and *Bacillus subtilis*: A review. *African Journal of Agricultural Research* **9**, 1265-1277.

SOPA (2019) The Soybean Processors Association of India. Scheme No. 53, Near Malviya Nagar, A.B. Road, Indore, https://www.sopa.org/

Subbiah, B.V. and Asija, G.L. (1956) A rapid procedure for the estimation of available nitrogen in soils. *Current Science* **25**, 259-260.

Yasmin, F., Othman, R., Saad, M.S. and Sijam, K. (2007) Screening for beneficial properties of Rhizobacteria isolated from sweet potato rhizosphere. *Journal of Biotechnology* **6**, 49-52.

Received 23 December 2020; Accepted 18 June 2021