Response of co-inoculation of Phosphate Solubilizing Bacteria (PSB) and Arbuscular Mycorrhizal (AM) fungi to microbial population of soil and NPKuptake by wheat (*Triticum aestivum* L.) crop

ABSTRACT

Aim: To study the response of co-inoculation of Phosphate Solubilizing Bacteria (PSB) and Arbuscular Mycorrhizal (AM) fungi to microbial population of soil and nutrient uptake by wheat crop.

Study Design: Theused design was completely randomized design with three replications.

Place of study: The pot experiment was conducted during *Rabi* season of 2017 taking wheat (cv. HD 2967) as test crop in experimental unit of the department of Soil Science and Agricultural Chemistry,Bihar Agricultural College, Sabour, Bhagalpur (Bihar).

Methodology: The present study includes eight treatments with three replications. N P K uptake by wheatcrop were measured from each treatment and microbial population of soil were determined from the rhizospheric soils collected from each treatment by using standard protocol**.**

Results:Maximum microbial population *viz*., Bacteria, Actinomycetes, Fungi (39.00, 21.33, 24.66 CFU $\times 10^5$ g^{-1} oven dry soil) were recorded under treatment T_4 (T_1+PSB @ 20 g kg-^{1} seed+AM fungi @ 5.0g pot^{-1}) for bacteria and actinomycetes and T_3 (T_1+AM fungi@5.0 g pot^{-1})for fungi, at flowering stage and similar trend was followed at harvesting stage. The treatment T_4significantly increased available nitrogen, phosphorus and potassium in soil as well as also contributed to comparatively better plant growth and higher uptake of N, P and K by grain and shoot. The maximum N, P and K content of wheat was also recorded under treatment T_4 which was found to be most effective in modifying soil microbial population, microbial community structure and grain yield of wheat crop.

Conclusion: Application of co-inoculation of Phosphorus solubilizing bacteria and arbuscular mycorrhizal fungi enhance the microbial population and N P K uptake from soil by wheat crop.

Keywords: AM fungi,microbial population, phosphorus solubilizing bacteria,wheat

Introduction

Phosphorus is critical element for plant growth and their development, and is a component of the nucleic acid structure of plants and biomembrane. Consequently, it is important in cell division and tissue development. Phosphorus is also involved in the energy

metabolism of cells and is required for the biosynthesis of primary and secondary metabolites in plants. Therefore, plants have evolved a range of strategies to increase phosphorus uptake and mobility [9], the most common among which are phosphate solubilizing bacteria (PSB) and Arbuscular mycorrhiza (AM) fungi symbiosis.

Most of the applied phosphorus in the soils is not taken up by the crop, but it is retained in insoluble forms or fixed as mineral forms in the farms even as high as 90% or more. Soil P can exist in various inorganic (Pi) and organic forms (Po). Specific determination of Pi can be obtained by fractionation methods. Generally, plants take up P as the primary ($H_2PO_4^-$) and secondary orthophosphate (HPO_4^-) ions. They are easily retained in most soils when added, and in many cases this retention is so high that the element becomes largely unavailable to the plants. The P retained by the soil is generally considered as fixed P, although a part of it can be utilized. The rate of release of P from fixed form to replenish the immediately available soil P is the most important factor in determining the P supplying capacity of the soil because the quantity of P present in soil solution is not sufficient to meet the crop requirements. The release of fixed soil P depends upon the nature of its fixation and the extracting power of the crop or reagent used to determine availability of soil P. The P is generally fixed as Fe-P and AI-P in acidic soils and Ca-P in alkaline soils.

Comment [s1]: Do not use abbreviations

Comment [s2]: Do not use abbreviations

P-solubilization ability of the microorganisms is considered to be one of the most important traits associated with plant phosphorus (P) nutrition. Recently, phosphate solubilizing microorganisms have attracted the attention of agriculturists as soil inoculums to improve the plant growth and yield. Conversion of the insoluble forms of P to the form which is available to plants (ortho-phosphate) is an important characteristics of phosphate- solubilizing bacteria (PSB) and Arbuscular Mycorrhizal Fungi (AMF). Bacteria such asPSB and AM fungi are usually effective on phosphate solubility due to different mechanism such as production and secretion of organic acids and by their co-inoculation they make phosphorus available to plant for different metabolic functions [3]. Release of phosphorus by PSB from insoluble and fixed or adsorbed forms is an important aspect regarding P availability in soils. There are strong evidences that soil bacteria are capable of transforming soil P to the forms available to plant. AM fungi are species of fungi that intimately associate with plant roots forming asymbiotic relationship, with the plant

providing sugars for the fungi and the fungi providing nutrients such as phosphorus, to the plants. Mycorrhizal fungi can absorb, accumulate and transport large quantities of phosphate within their hyphae and release to plant cells in root tissue. Arbuscular mycorrhizal (AM) fungus plant relationships are usually described as mutually beneficial, because fungi supply mineral nutrients, especially phosphorus (P) to their host plants in return for photosynthates. The contribution of AM fungi to P uptake by positively responsive plants can be easily identified by comparison of P uptake in AM plants and non-mycorrhizal (NM) plants. The beneficial effect of arbuscular mycorrhiza on plant growth is mainly attributed to higher P uptake by plants [12].

This is the fact that phosphorus solubilizing bacteria and mycorrhizal fungi increase the availability of phosphorus. There is a great scope to make more P availability in soil by adopting new agronomic practices like use of co-inoculation of PSB and AMF with its appropriate dose, proper combinations and inorganic fertilizer etc. Considering above mentioned facts a pot experiment on response of Co-inoculation of Phosphorus solubilizing bacteria (PSB) and Arbuscular Mycorrhizal (AM) Fungi on phosphorus availability under wheat rhizosphere has been conducted.

Methods and Material

The present study was undertaken in pot to evaluate the response of co-inoculation of Phosphate Solubilizing Bacteria (PSB) and Arbuscular Mycorrhizal (AM) fungi to soil microbial population and availability of phosphorus under wheat rhizosphere during the *Rabi* season of 2017 with a promising var. HD 2967, at the experimental site of department of Soil Science and Agricultural Chemistry, Bihar Agricultural University, Sabour, Bhagalpur, India.

The microbial inoculums viz., PSB- *Burkholderia cepecia* and AM fungi- *Glomus mosseae* were procured from biofertilizer production unit, Bihar Agricultural University, Sabour, Bhagalpur, and Bihar. The soil used for the pot experiment wasUstochrept clayey in texture, having a pH of 7.78 and EC of 0.20ds m^{-1}. The organic carbon content of the soil was 0.47%, and the available nitrogen (N), phosphorus (P) and potassium (K) content was found 150.42, 12.51 and 192.66 kg ha^{-1},respectively.

The following treatment structure was formulated for the study: T_1-RDF (120:60:40), T_2-T_1+PSB @ 20 g kg^{-1} of seed, T_3-T_1+AM fungi@5.0 g pot^{-1}, T_4-T_1+PSB@20 g kg^{-1} seed+AM

fungi @ 5.0g pot^{-1}, T_5-75% RDF of Phosphorus +PSB@ 20 g kg-^{1} seed, T_6-75% RDF of Phosphorus +AM fungi @ 5.0g pot^{-1}, T_7-75% RDF of Phosphorus +PSB @ 20g kg^{-1} of seed+AM fungi@5.0 g pot^{-1}, and T_8-50% RDF of Phosphorus +PSB @ 20g/kg seed+AM fungi @5.0 gPot-1. Earthen pots of 15 cm height and 30 cm diameter were filled with 10 kg of soil. The seed treatment with PSB@ 20 kg^{-1} seed was done and AM fungi@ 5g Inoculum pot^{-1} were applied 2cm below the seed at the time of sowing. In each pot, 10 seeds of wheat (var. HD-2967) were planted. Nitrogenous, phosphatic and potassic fertilizers were applied just before the sowing according to the treatments. Using urea, single super phosphate and murate of potash as source of nitrogen, phosphorus and potash, respectively and mixed in the soil uniformly by working with spade. The irrigation was applied as and when required the crops. The plants were thin to maintain eight plants in all pots.

Rhizosphere samples were drawn from the soil adhering to the roots. The 10 g of soil samples were placed in an Erlenmeyer flask containing 90 ml of sterilized distilled water, and shaken for 30 min. Ten-fold series dilutions were prepared, and appropriate dilutions were plated in specific media. For the isolation of bacteria, fungi and actinomycetes, the Plate Count Agar, Czapek-Dox Agar [19] and Kenknight and Munaier's Medium, respectively were used. The numbers of colony forming cells were determined in each plot by serial dilution pour plate method [18]. Phosphorus concentration in straw and grain were determined by employing the vanadomolybdate yellow color method given by [6] and the distribution of P in soil was determined by using a modified version of the [5]. Analysis of variance (ANOVA) was performed as described by [2] to determine the effects of various treatments. Critical difference (CD) at 5% level of probability and P values was used to examine differences among treatment means.

Result and Discussion

Effect on Bacterial population

It is evident from presented data that the microbial population resulted highest in the flowering stage of wheatplant growth. This might be due to accumulation of various root exudates and which in turn, established a strong and well defined root-microbe interaction [16]. The inoculation with treatment T_4 (T_1+PSB @20 gkg^{-1} seed + AM fungi @ 5.0gpot^{-1}) having significantly more bacterial population,when compared with applied treatments. It is clearly

shown that all applied treatments have given maximum bacterial count at flowering stage when compared with harvesting stage .The application of treatment T_4(T_1+PSB@20 gkg^{-1} seed+AM fungi @ 5.0$gpot^{-1}$) also produced significantly higher bacterial count by 22.23%, 4.28%,9.41%, than application of T_1(RDF 120:60:40),T_2(T_1+PSB@20gkg^{-1}seed), T_3(T_1+AMfungi@5.0g pot^{-1}) respectively. The similar trends were observed at harvesting stage.At harvesting stage application of treatment T_6(75% RDF of P +AM fungi @ 5.0$gpot^{-1}$) and treatment T_7 (75% RDF of P + PSB @ 20g kg^{-1}seed+AMfungi@5.0g pot^{-1}) given significantly higher colonization over all the applied treatments. Similar trend followed at harvesting stage. It might be due to the AM fungi are probably the most abundant fungi in agricultural soil accounting for somewhere between 5 and 50% of biomass of soil microbes live on carbohydrates obtained from the root cells. They alter root exudation considerably [8] and are therefore expected to influence rhizosphere populations as well [4]. Numerous studies have shown conclusively that AM is having synergistic interaction with other beneficial soil microorganism such as N fixers and P solubilizers. AM fungi affect the composition of bacterial communities either directly by changing host plant physiology or indirectly by changing the pattern of root exudation. The number of both rhizospheric bacteria and actinomycetes enhanced when plant formed mycorrhizae, along with the inoculation of PSB [15]. There may be two pathways for AM fungi to change microbe community structure, the first one is that the AM fungal hyphae secretion directly impacts microbe community structures; the another one is that both AM fungi in roots and on the roots alter plant physiological and biochemical processes, then directly or indirectly change the plant root secretion,thus alter those structures [20].

Effect on actinomycetes population

Same trend of enhancement of actinomycetes count has been resulted under the experimentation as compared as obtained incase of bacteria. The growth pattern was resulted synonymous to bacterial counterpart. The highest individual treatment effect was observed under the treatment T_4(T_1+PSB@20 g kg^{-1} seed+AM fungi @ 5.0$gpot^{-1}$) in flowering stage, produced maximum population of actinomycetes (21.33 CFU$\times 10^6$ g^{-1} soil). The treatmentwas significantly affectedactinomycetes population in all the applied treatments except treatment T_2 (T_1+PSB @ 20g/kg of seed). Similar trends were followed at harvesting stage. The similar results were obtained by [11] who conducted a field experiment and found that the maximum bacterial population (71.66 CFU × 105 g^{-1} soil and 40.00 CFU × 105 g^{-1} soil), fungi population (27.33

CFU × 104 g^{-1} soil and 22.66 CFU × 104 g^{-1} soil) and actinomycetes population (57.66 CFU × 105 g^{-1} soil and 46.33 CFU × 105 g^{-1} soil) are observed in the treatment T_5 {(75 % N (FYM) basal + 25 % N (V/C) at 25 DAT + *Azospirillum*@ 5 kg ha^{-1} + PSB @ 5 kg ha^{-1} + KSB @ 5 kg ha^{-1})} at both the panicle and harvesting stage in scented rice.

Comment [s3]: Clarify the description

Effect on fungal population

It is depicted from the data that the fungal count resulted highest in the flowering stage of growth as compared to harvesting stage. The application of treatment T_3(T_1+AMfungi @ 5.0 g pot^{-1}) given maximum population of fungi (24.66 propagules ×10^3 g^{-1} soil). This treatment also given significantly higher fungal population by 17.02%, 40.53%,16.21 %, 44.59%, 12.16%, 16.21%, over application of treatment T_1 (RDF (120:60:40), T_2 (T_1+PSB@ 20gkg^{-1} seed), T_4 (T_1+PSB @ 20 gkg^{-1} seed+AM fungi @ 5.0$gpot^{-1}$), T_5 (75% RDF of P +PSB @ 20gkg^{-1} seed), T_6 (75% RDF of P +AM fungi @ 5.0$gpot^{-1}$) and T_7(75% RDF of P +PSB @ 20gkg^{-1} seed+AM fungi@5.0 $gpot^{-1}$) respectively. Similar trend observed at harvesting stage. At harvesting stage the maximum number of fungal population (15.667 propagules×10^3 g^{-1} soil) was recorded with the application of treatment T_3(T_1+AMfungi @ 5.0 g pot^{-1}). It might be due accumulation of various root exudates and which in turn, established a strong and well defined root-microbe interaction [10]also as compared to bacteria and fungi. The similar results were found by the[16].

Response of PSB and AMF species to availability of nutrient in soil after harvestAvailable Nitrogen (kg ha-^{1}).

Comment [s4]: Not clear.. is it subheading ? or sentence

The maximum available nitrogen (210.24 kg ha^{-1}) was recorded under the application of treatmentT_4 (T_1+PSB @ 20 gkg^{-1} seed +AM fungi @ 5.0gPot^{-1}). It has been clearly observed that the available nitrogen in soil was significantly higher than all the applied treatments. It might be due to the production of more shoot biomass and root biomass by the application of given treatments and might be due to the structural changes in the microbial community. These changes in the microbial community may alter the nutrients dynamics in the rhizosphere. The similar results were observed by the [1].

Available phosphorus (kg ha^{-1})

The data regarding available phosphorus in soil revealed that the maximum phosphorus (19.25 kg ha^{-1}) was found by the application of treatment T_4 (T_1+PSB @ 20 gkg^{-1} seed+AM fungi @ 5.0gPot^{-1}). The application of treatment T_4 given significantly higher availablephosphorus by 31.68% , 15.37%, 19.16%, 10.38% and 5.66% when compared with the

treatment T_1 { (RDF (120:60:40)}, T_2 (T_1+PSB @ 20gkg^{-1} of seed), T_3(T_1+AM fungi @ 5.0 gPot^{-1}) ,T_5(75% RDF of P +PSB @ 20gkg^{-1} seed), T_6(75% RDF of P +AM fungi @ 5.0gPot^{-1}), and numerically the least value of available phosphorus was found under the treatment T_1(RDF (120:60:40)}. Recorded data shows that the inoculation with T_7 (75% RDF of P +PSB @ 20 g kg^{-1} of seed+AM fungi@ 5.0 gPot^{-1}) having also more significantly availability of phosphorus by 28.72%, 11.70%, 15.66%, 6.50% and 22.76% over the treatments T_1 , T_2 , T_3 , T_5 and T_8 respectively.It may be due to the *Glomus mosseae* had pronounced effect for phosphorus acquisition in soil inoculated with PSB have a great result. [13] who conducted an experiment on *Coriander sativum* L. to study the effect of arbuscular mycorrhizal fungus *Glomus mosseae* and phosphorus application on plant growth rate,essential oil content and composition of coriander, and found that the mycorrhizal inoculation significantly increased growth responses and P and N plant nutrients in shoot and root tissue, also after inoculation of arbuscular mycorrhizal fungi in to coriander plant is a feasible alternative to increase growth ,nutrition, essential oil production and reduce the use of P fertilizers required to obtain economic production of coriander under phosphorus deficient soil condition.

5.3.3. Available potassium (kg ha-^{1})

Comment [s5]: Follow same formatting style

The maximum available potassium (210.07 kg ha^{-1}) in soil was recorded bythe application of T_4 (T_1+PSB@20 gkg^{-1} seed+AM fungi @ 5.0gPot^{-1}). This treatment also gave significantly higher amount of available potassium in soil by 13.66%, 11.68%, 12.81%, 12.25% ,12.13%, 4.72% over the treatments T_1 {(RDF (120:60:40)},T_2(T_1+PSB@20gkg^{-1} seed),T_5(75% RDF of P +PSB @ 20gkg^{-1} seed),T_6(75% RDF of P +AM fungi @ 5.0gPot^{-1}),T_7(75% RDF of P +PSB @ 20gkg^{-1} of seed+AM fungi@5.0 gPot^{-1}) and T_8(50% RDF of P+PSB @ 20gkg^{-1} seed+AM fungi @5.0 gPot^{-1}) while the treatment T_3(T_1+AM fungi@5.0 gPot^{-1}) given significantly zero value.It has been shown that in co-inoculated treatment of PSB and AM fungi having more amount of available potassium present with respect to un-inoculated condition like T_1,T_2 ,T_3etc. Another treatment which is inoculated with T_8 given significantly higher availability of potassium except T_3 and T_4 but another by 9.37%, 7.30%, 8.49% and 7.77% than T_1, T_2 , T_5 , T_7 respectively. The obtained results are in the agreement of [1], who conducted an investigation to evaluate the response of selected species of mycorrhizae for nutrient acquisition and phosphorus uptake by maize in an alluvial soils of Bihar and found that value of available potassium has increased .

Soil organic carbon (%)

The data revealed with percent organic carbon in soil showed that the application ofall treatments were non-significant. The data regarding available organic carbon in a soil maximum (0.54%) was recorded by the application of treatment T_2 (T_1+ PSB @ 20g/kg of seed),T_3(T_1+AM fungi @5.0 $gPot^{-1}$),T_4 (T_1+PSB @20 gkg^{-1} seed +AM fungi @ 5.0$gPot^{-1}$), T_5 (75% RDF of P +PSB @ 20 g kg^{-1} seed), T_6 (75% RDF of P +AM fungi @ 5.0$gPot^{-1}$), T_7 (75% RDF of P +PSB @ 20gkg^{-1} of seed+AM fungi@5.0 $gPot^{-1}$) and T_8(50% RDF of P +PSB @ 20gkg^{-1} seed+AM fungi @5.0 $gPot^{-1}$). It might be due to the AM colonization produced more root biomass and plant biomass [1],who conducted an experiment Evaluation of Arbuscular Mycorrhiza Fungi Species for Their Efficiency Towards Nutrient Acquisition in Rhizospheric Soil of Maize and revealed thatthe organiccarbon content exhibited significant positive correlation withcontent. This positive correlation with organic carbon indicated that cationic micronutrients formed complexes with organic matter and consequentially remained in the forms, easily available to the plants.

Effect of microbial inoculants on N P K content and uptake by wheat

The data regarding N P K content and uptake in shoot and grain of wheat clearly depicted that the application of treatment T_4(T_1+PSB@20 gkg^{-1} seed+AM fungi @ 5.0$gPot^{-1}$) given maximumN P K content and their uptake over all the applied treatments,which is found to be significantly higher over the applied treatment T_1{(RDF (120:60:40)},T_2(T_1 +PSB @ 20gkg^{-1} seed),T_5 (75% RDF of P +PSB @ 20gkg^{-1} seed),T_6 (75% RDF of P +AM fungi @ 5.0$gPot^{-1}$),T_7(75% RDF of P +PSB @ 20gkg^{-1} seed+AM fungi@5.0 $gPot^{-1}$) and T_8 (50% RDF of P+PSB @ 20gkg^{-1} seed+AM fungi @5.0 $gPot^{-1}$).

The data pertaining N content (%) has been clearly observed that the nitrogen content percentage in shoot was maximum (0.149%)with the treatment T_4(T_1+PSB@20 g kg^{-1}seed+AM fungi @ 5.0$gpot^{-1}$),which is significantly zeroover the applied treatment T_1{(RDF (120:60:40)},T_2 (T_1+PSB@20gkg^{-1} of seed),T_3(T_1+AM fungi@5.0 $gpot^{-1}$),T_5,(75% RDF of Phosphorus +PSB @ 20gkg^{-1} seed),T_6(75% RDF OF Phosphorus +AM fungi @ 5.0$gpot^{-1}$) and T_7(75% RDF of Phosphorus +PSB @ 20g kg^{-1} of seed+AM fungi@5.0 g pot^{-1})The lowest nitrogen content (0.140%) was found under the treatment T_1.While the treatment T_3 given significantly higher value by 2.75%,2.06%,2.7% over the applied treatment T_1,$T_2$and T_5respectively while T_4 and T_8 are at par values. In the same way treatment T_8(50% RDF of

Phosphorus +PSB @ 20gkg^{-1} seed+AM fungi @5.0 gpot^{-1}) have given significantly higher value by 5.40%,4.05%,2.02%,4.72%,2.70%,over the applied treatment T_1,T_2,T_3,T_5 and T_7 while the treatment T_4 have at par values.

The data pertaining P content (%) has been clearly observed that the phosphorus content percentage was maximum (0.245%) with the treatment T_4(T_1+PSB@20 g kg^{-1} seed+AM fungi @ 5.0gpot^{-1}),which is found to be significantly higher over the applied treatment T_1{(RDF (120:60:40)},T_2(T_1+PSB@20gkg^{-1} of seed) T_3(T_1+AM fungi@5.0 gpot^{-1}),T_5(75% RDF of Phosphorus +PSB @ 20gkg^{-1} seed),T_6(75% RDF OF Phosphorus +AM fungi @ 5.0gpot^{-1}),T_7(75% RDF of Phosphorus +PSB @ 20g kg^{-1} of seed+AM fungi@5.0 g pot^{-1}) and T_8(50% RDF of Phosphorus +PSB @ 20gkg^{-1} seed+AM fungi @5.0 gpot^{-1}) by22.85%,1.22%,2.04%,6.93%,4.48%,19.18% respectively.While the treatment T_2 have given significantly higher value by 21.90%,5.71%, 17.95% over the applied treatment T_1,T_5 and T_8 respectively,while the values of treatment T_2,T_3,T_7 are at par.

The data pertaining k content (%) has been clearly observed that the potassium content percentage was maximum (1.16%) with the treatment T_4(T_1+PSB@20 g kg^{-1} seed+AM fungi @ 5.0gpot^{-1}),which is found to be significantly higher over the applied treatment T_1{(RDF (120:60:40)}, ,T_2(T_1+PSB@20gkg^{-1} of seed),T_5(75% RDF of Phosphorus +PSB @ 20gkg^{-1} seed),T_6(75% RDF OF Phosphorus +AM fungi @ 5.0gpot^{-1}),T_7(75% RDF of Phosphorus +PSB @ 20g kg^{-1} of seed+AM fungi@5.0 g pot^{-1}) and T_8(50% RDF of Phosphorus +PSB @ 20gkg^{-1}seed+AMfungi@5.0 g pot^{-1}) by 13.79%, 9.48%, 11.20%,10.34%, 9.48%, 7.75% respectively while the value of T_4 and T_7 are at par .The treatment T_3 have given significantly higher value than 13.04%,1.904% over the applied treatments T_1 and T_2,while the values of T_3 and T_4 are at par.The data pertaining nitrogen uptake (g pot^{-1}) , has been clearly observed that the nitrogen uptake in shoot was maximum (0.0460%) with the treatment T_4(T_1+PSB@20 g kg^{-1} seed+AM fungi @ 5.0gpot^{-1}) of table in which the nitrogen uptake was maximum which is significantly higher than applied treatment T1{(RDF (120:60:40)},T_2(T_1+PSB@20gkg^{-1} of seed),T_3(T_1+AM fungi@5.0 gpot^{-1}) ,T_5(75% RDF of Phosphorus +PSB @ 20gkg^{-1} seed),T_6(75% RDF Of Phosphorus +AM fungi @ 5.0gpot^{-1}) T_7(75% RDF of Phosphorus +PSB @ 20g kg^{-1} of seed+AM fungi@5.0 gpot^{-1}) and T_8(50% RDF of Phosphorus +PSB @ 20gkg^{-1} seed+AM fungi @5.0 gpot^{-1}) by21.73% ,10.86%, 17.39%, 13.47%, 13.26%, 17.39% and 14.78% respectively.

277 6.57%, 6.44%,17.10%,13.55% respectively. While the treatment T_3 have given significantly
278 higher value 5.26% over the applied treatment T_1 while the value T_2,T_5 and T_8 are at par.
279 Also from the data pertaining P uptake (g pot^{-1}) clearly observed that the phosphorus uptake
280 in shoot was maximum (0.0760%) with the treatment T_4(T_1+PSB@20 g kg^{-1} seed+AM fungi @
281 5.0gpot^{-1}) of table in which the phosphorus uptake was maximum which is significantly higher
282 than applied treatment T_1{(RDF (120:60:40)},T_2(T_1+PSB@20gkg^{-1} of seed),T_3(T_1+AM
283 fungi@5.0 gpot^{-1}) ,T_5(75% RDF of Phosphorus +PSB @ 20gkg^{-1} seed), T_6(75% RDF of
284 Phosphorus +AM fungi @ 5.0gpot^{-1})by35.52% ,6.57% ,6.44%, 17.10%, 13.55%,
285 17.63%,29.73% respectively. The similar results reported by [15].
286 The data pertaining K uptake (g pot^{-1}) has been clearly observed that the potassium
287 uptake in shoot was maximum (0.362%) with the application of treatment T_4(T_1+PSB@20 g kg^{-1}
288 seed+AM fungi @ 5.0gpot^{-1}) of table in which the potassium uptake was maximum which is
289 significantly higher than applied treatment T_1{(RDF (120:60:40)},T_2(T1+PSB@20gkg^{-1} of seed
290),T_3(T_1+AM fungi@5.0 gpot^{-1}) ,T_5(75% RDF of Phosphorus +PSB @ 20gkg^{-1} seed), T_6(75%
291 RDF OF Phosphorous +AM fungi @ 5.0g/pot)by27.07% and 14.91% ,6.077%, 21.27%, 19.33%,
292 respectively. While the treatment T3 have given significantly higher value by
293 22.35%,9.41%,16.17%, 14.11%, 17.94%, 15% over the applied treatment T_1, T_2, T_5, T_6, T_7 and
294 T_8 respectively with the value T_4 is at par. It clearly indicate that the potassium uptake was more
295 and more occur in inoculated species with respect to uninoculated treatment .It might be due to
296 the co-inoculation of PSB and AMF along with full dose of fertilizers,PSB secrete organic
297 acids of lower molecular weight and AM fungi make it more available to plants through its
298 hyphae and thus nutrient content and uptake concentration increases. Similar results were
299 obtained by [14], who conducted an experiment on Co-inoculation studies of vesicular
300 Arbuscular Mycorrhizal fungi (VAM) and Phosphate solubilizing bacteria (PSB) on nutrient
301 uptake of *Marsdenia volubilis* (T. Cooke) and found an excellent improvements in uptake of
302 nutrients like N, P, K, Ca, Mg, Fe, Mn and Zn concentrations in *Marsdenia volubilis* than single
303 application.

304 **Conclusion:**

305 The application of microbial inoculants in combination yield was better than separate application
306 of inoculation. The results of the study revealed that the maximum microbial population *viz*.,
307 Bacteria, Actinomycetes, Fungi were found co-inoculation of PSB and AM fungi along with

recommended dose of phosphorus. The application of PSB@20 g kg^{-1} seed+AM fungi @ 5.0g pot^{-1} along with 100% RDF significantly increased available nitrogen, phosphorus and potassium in soil as well as also contributed to comparatively better plant growth and higher uptake of N, P and K by grain and shoot. This treatment was found to be most effective in modifying soil microbial population, microbial community structure and grain yield of wheat crop.

References:

1.Beura K, Singh M, Kumar A, Rakshit R, Lal M.Evaluation of Arbuscular Mycorrhiza Fungi Species for Their Efficiency Towards Nutrient Acquisition in Rhizospheric Soil of Maize. International Journal of Bio-resourceand Stress Management.2016 ;7 (1):130-135.

2.Gome KA, Gomez AA.Statistical Procedures in Agricultural Research, By New York, Chichester, etc.Wiley 2nd edition, paperback.1984 :680.

3.Gyaneshwar P, Kumar N, Parekh LJ, Poole PS. Role of soil microorganisms in improving P nutrition of plants.Plant and Soil.2002; 245: 83-93.

4.Hayman DS.The physiology of vesicular-arbuscular mycorrhizal symbiosis.Canadian Journal of Botany.1983; 61:944-963.

5.HedleyM,Stewart J.Method to measure microbial phosphate in soils, Soil Biol. Biochem. 1982; 14, 377-385.

6.Jackson M L.Soil Chemical Analysis Prentice-Hall Inc. Englewood Cliffs, New Jersey; 1973.

7.Marschner H.Mineral nutrition of higher plants. Academic press, San Diego,CA USA. 1996;889.

8.Marschner P, Crowley DE, Higashi M.Root exudation and physiological status of a root colonizing fluorescent pseudomonas in mycorrhizal and non-mycorrhizal pepper (*Capsicum annuum*). Plant Soil.1997;189:11-20.

9.MarschnerM. Mineral nutrition of higher plants. Plant and Soil.1996; 159: 89-102.

10.Rovira AD.Interaction between plant roots and soil microorganisms. Ann. Rev. Microbiol.1965;19: 241-246.

11.Sah AK, Singh M, Prasad S, Bahera KS, ChattopadhayaN, Kiran K. Effect of organic fertilizer and microbial inoculants on soil biological properties and yield of scented rice (*Oryza sativa* L.). Journal of Applied and Natural Science.2018; 10(3): 995 – 1002.

12.Sakurai K, Tanaka N, Iwasaki K, Tanak S . Effect of arbuscular mycorrhizal application on the distributionAzcon-Aguilar C, Barea JM Applying mycorrhizal biotechnology to horticulture; 2001.

13.Salem M, Al-Amri, Khalid M, Elhindi Ahmed F,Sharaf El-Din.Effect of arbuscular mycorrhizal fungus *Glomus mosseae* and phosphorus application on plant growth rate,essential oil content and composition of coriander (*Coriander sativum* L.)Progress in Nutrition.2016;10(4):443-454.

14.Sandhya A1, Vijaya T2, Sridevi A1, Narasimha G.Annals of Plant SciencesCo-Inoculation Studies of Vesicular Arbuscular Mycorrhizal Fungi (VAM) and Phosphate Solubilizing Bacteria (PSB) On Nutrient Uptake of Marsdenia volubilis(T. Cooke);2014.

15.Singh M, Kumar N, Kumar S, Mehilal M.Effect of co-inoculation of *b. japonicum*, psb and am fungi on microbial biomass carbon, nutrient uptake and yield of soybean (*Glycine max* L. merril)Agriways.2015;3(1): 14-18. ISSN: 2321-8614 .

16.Singh M, Kumar N. Effect of FYM, Vermicompost, Vermiwash and NPK on Growth, Microbial Biomass C and Yield of Soybean. *Soybean Res*. 2012; 10: 60-66.

17.Singh M, Kumar N, Kumar S, Lal M. Effect of Co-Inoculation of *B. Japonicum*, PSB and AM fungi on microbial biomass carbon, nutrient uptake and yield of soybean (*Glycine Max* L. Merril)Agriways. 2015*;*3(1): 14-18.

18.Subba RNS.*Rhizobium* and root nodulation. In: soil microorganisms and plant growth. Oxford IBH New Delhi; 1986.

19.Thom C, Raper K.B. A manual of the *Aspergilli*.Williams and Wilkins Co.,Baltimore, U. S. A;1945.

20.Zhu HH, Long LK, Yang SZ.Influence of AM fungus on *RalstoniaSolanacearum*Population and bacterial community structure in rhizospherenMycosystema. 2005;24(11):137-142.

Table 1: Effect of Co-inoculation of PSB and AM fungi on microbial population under wheat rhizoshpere

Treatment	Bacterial count (CFU 10^6 g^{-1} soil)		Actinomycetes count (CFU 10^6 g^{-1} soil)		Fungi count (CFU 10^4 g^{-1} soil)	
	Flowering stage	Harvesting Stage	Flowering stage	Harvesting Stage	Flowering stage	Harvesting Stage
T_1	30.33	21.00	12.66	5.66	20.46	10.66
T_2	37.33	28.66	20.67	8.67	14.66	8.67
T_3	35.33	27.66	18.66	17.67	24.66	15.66
T_4	39.00	29.33	21.33	11.67	20.67	9.67
T_5	30.66	29.00	10.67	7.66	13.67	7.66
T_6	31.66	31.00	9.67	8.66	21.66	13.67
T_7	32.66	31.00	13.67	11.66	20.66	11.66
T_8	29.33	20.00	8.66	6.67	22.66	11.67
C.D.(P=0.05)	1.671	1.51	1.671	1.008	1.008	1.008
C.V.	2.876	3.18	2.876	5.896	2.899	5.17

Note: T_1-RDF (120:60:40), T_2- T_1+PSB @ 20 g kg^{-1} of seed, T_3-T_1+AM fungi@5.0 g pot^{-1}, T_4-T_1+PSB@20 g kg^{-1} seed+AM fungi @ 5.0g pot^{-1}, T_5-75% RDF of P +PSB@ 20 g kg-^{1} seed, T_6-75% RDF of P +AM fungi @ 5.0g pot^{-1}, T_7-75% RDF of P +PSB @ 20g kg^{-1} of seed+AM fungi@5.0 g pot^{-1}, and T_8-50% RDF of P +PSB @ 20gkg^{-1} seed+AM fungi @5.0 gpot^{-1}.

Table 2: Effect of co-inoculation of PSB and AM fungi on availability of N P K(kg ha-^{1}) and organic carbon in soil after wheat harvest.

Treatment	Available N, P K (kg ha^{-1} soil)			O C (%)
	Nitrogen	Phosphorus	Potassium	
T_1	188.19	13.15	181.37	0.53
T_2	199.95	16.29	185.52	0.54
T_3	198.36	15.56	208.18	0.54
T_4	210.24	19.25	210.07	0.54
T_5	197.24	17.25	183.14	0.54
T_6	204.18	18.16	184.33	0.54
T_7	204.52	18.45	184.57	0.54
T_8	202.13	14.25	200.14	0.54
C.D.(P=0.05)	0.422	0.209	0.232	NS
C.V.	0.12	0.722	0.069	-

Note: T_1-RDF (120:60:40), T_2- T_1+PSB @ 20 g kg^{-1} of seed, T_3-T_1+AM fungi@5.0 g pot^{-1}, T_4-T_1+PSB@20 g kg^{-1} seed+AM fungi @ 5.0g pot^{-1}, T_5-75% RDF of P +PSB@ 20 g kg-^{1} seed, T_6-75% RDF of P +AM fungi @ 5.0g pot^{-1}, T_7-75% RDF of P +PSB @ 20g kg^{-1} of seed+AM fungi@5.0 g pot^{-1}, and T_8-50% RDF of P +PSB @ 20gkg^{-1} seed+AM fungi @5.0 gpot^{-1}.

Table 3: Effect of co-inoculation of PSB and AM fungi on N P K content (%) and N, P, K uptake (g pot^{-1}) by wheat plant

Treatment	N, P, K content (%) in plant			N, P, K uptake(g pot-^{1}) by plant		
	N	P	K	N	P	K
T_1	0.140	0.189	1.00	0.0360	0.0490	0.264
T_2	0.142	0.242	1.05	0.0410	0.0710	0.308
T_3	0.145	0.240	1.15	0.0380	0.0711	0.340
T_4	0.149	0.245	1.16	0.0460	0.0760	0.362
T_5	0.141	0.228	1.03	0.0398	0.0630	0.285
T_6	0.142	0.234	1.04	0.0399	0.0657	0.292
T_7	0.144	0.235	1.05	0.0380	0.0626	0.279
T_8	0.148	0.198	1.07	0.0392	0.0534	0.289

C.D.(P=0.05)	0.001	0.008	0.008	0.001	0.005	0.004
C.V.	0.402	0.936	0.426	0.395	0.456	0.389

385 Note: T_1-RDF (120:60:40), T_2- T_1+PSB @ 20 g kg^{-1} of seed, T_3-T_1+AM fungi@5.0 g pot^{-1}, T_4-
386 T_1+PSB@20 g kg^{-1} seed+AM fungi @ 5.0g pot^{-1}, T_5-75% RDF of P +PSB@ 20 g kg-^{1} seed, T_6-
387 75% RDF of P +AM fungi @ 5.0g pot^{-1}, T_7-75% RDF of P +PSB @ 20g kg^{-1} of seed+AM
388 fungi@5.0 g pot^{-1}, and T_8-50% RDF of P +PSB @ 20gkg^{-1} seed+AM fungi @5.0 gpot^{-1}.

389