Effect of Inoculating *Bradyrhizobium* on Phosphorus Use Efficiency and Nutrient Uptake of Soybean in Calcareous Soil, Central Rift Valley, Ethiopia

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ከ7.0 ፒ.ኤች በላይ የአፈር ፎስፈረስ ግዑዝ ስለማይነሟማ በክፍተኛ ሁኔታ ለዝግጠት ተጋላጭ ስለሚሆን ዕፅዋቶች በቀላሉ አየገኙትም፡፡ ስለዚህ ይህ የምርምር ጥናት በመተሐራ ስኳር ፋብሪካ በ2008 ዓ.ም ምርት ዘመን የሚረዲራይዞቢይም ባክቴሪያ ክትባት አስተየፅዖ አኩሪ አተርን ከሽንኩሩ አገዳ ጋር በማሰባጠር በፎስፈረስ አወሳሰድና የፎስፈረስ አወሳሰድ ብቃትን ለመገምገም ዓላማ አንግቦ በመስኖ ተተግብራል፡፡ ሙከራው ሦስት መጠን የባክቴሪያ ክትባት ማለትም ሲጊዩምፈክስ፣ SB6B1አና ያልተከተብ እንዲሁም አራት ፎስፈረስ መጠን ማለትም 0፣ 10፣ 20 እና 30 ኪ.ግ/ሄክታር ፎስፈረስ ወስደዋል፡፡ ሙከራው የተዘጋጀው በራንዶማይዝድ ኮምፕሊት ብሎክ ዲዛይን በፋክትሪየል የተደራጀ ሲሆን ሦስት ቅጃ ግልባጭ አለው፡፡ የጭብጦ (መረጃ) ትንትኔ እንዳሚያሰየው ብራዲራይዞቢይም ክትባት ትርጉም ባለው ሁኔታ የዕፅዋት ናይትሮጅንና ፎስፈረስ ይዘት ላይ ካልተከተበ አንፃር አመርኪ ውጤት አሳይቷል፡፡ የተለያዩ የፎስፈረስ መጠን እንዲሁም የፎስፈረስና ክትባት ጥምረት በናይትሮጅንና ፎስፈረስ ይዘት ላይ ትርጉም ያለው ልዩነት አሳሳዮም፣ ነገር ግን 30 ኪ.ግ/ሄክታር ፎስፈረስ በጠቅላላ ፎስፈረስ ይዘት ላይ ክፍተኛ ልዩነት አምጥቷል፡፡ በብራዲራይዞቢይም ክትባት ምክንያት ፎስፈረስን የመውሰድ ብቃት ተሻሽሏል፡፡ በዚህ መሠረት ክፍተኛ አግሮኖሚክ ኤፊሼንሲ፣ ሪከቨሪ ኤፊሼንሲና የትላይዜሽን ኤፊሼንሲ በSB6B1 ክትባት፣ ፊዚዮሎጂካል ኤፊሼንሲና አግሮፊዚዮሎጂካል ኤፊሼንሲ በሊጉምፈክስ ክትባት አማካይነት በ10 ኪ.ግ/ሄክታር ፎስፈረስ ሲገኝ አግሮፊዚዮሎጂካል ኤፊሼንሲ በ30 ኪ.ግ/ሄክታር ፎስፈረስ ተመዝግቧል፡፡ በአጠቃላይ በ10 ኪ.ግ/ሄክታር ፎስፈረስ መጠን SB6B1 ባክቴሪያ ክትባት አመርኪ ውጤት ያስገኘ ሲሆን ሊጊዩምፈክስ ደግሞ ይከተላል፡፡ የፎስፈረስ መውሰድ ብቃትን እንዲጨምር የተሻሉ አያያዝ ዘዴያችን ማለትም ፎስፌት የሚያሟሙ ረቂቅ ነፍሳት ወይም ማይኮርዛ ብራዲራይዞቢይም ጋር በማጣመር መጠቀም አስፈላጊ ነው፡፡

Abstract

At a soil pH value of above 7.0, inorganic phosphorus (P) is highly susceptible to precipitation as insoluble form that is unavailable to plants. Hence, a field experiment was conducted at Metehara Sugar Estate under irrigation during the 2014/15 cropping season to evaluate the effect of inoculating Bradyrhizobium on P uptake and P use efficiency of soybean intercropped with sugarcane. The treatments consisted of three levels of inoculation (Legume fix, SB6B1 and uninoculated) and four rates of P (0, 10, 20 and 30 kg Pha[-1]). The experiment was laid out in a randomized complete block design (RCBD) in a factorial arrangement and replicated three times. Analysis of the data indicated that Bradyrhizobium inoculation significantly increased plant N concentration and P uptake compared to the uninoculated treatment. The effect of P rates and its interaction with inoculation was not significant on N concentration and P uptake, but significantly increased total P uptake at the application of 30 kg Pha[-1]. Phosphorus use efficiency indices were improved in response to inoculating the crop with Bradyrhizobium. The highest AE (13.6 kg kg^{-1}), PRE (31.8%) and PUE (10.6 kg kg^{-1}) were obtained by SB6B1 inoculation and the highest PE (117.2 kg kg^{-1}) and APE (161.7 kg kg^{-1}) were obtained by Legumefix inoculation all at 10 kg P ha^{-1}except PE which recorded the highest at 30 kg P ha^{-1}.Thus, it can be concluded that SB6B1 isolate can be used as the best inoculant followed by Legumefix isolate with 10 kg P ha^{-1}of P fertilizer. However, strategies for increasing P use efficiency by adopting best management practices like co-inoculation of phosphate solubilizing microorganism or mycorrhiza with these Bradyrhizobium inoculants should be adopted to enhance P use efficiencies.

Introduction

Phosphorus (P) is the most essential element for plant growth and development next to nitrogen (N) (Vance et al., 2000). It is one of the most important nutrients for crop productions on more than 30% of the world's arable land. Some estimates, world resources of inexpensive P may be depleted by 2050 (Yan et al., 2009). Phosphorus has significantly positive effect on nodulation nitrogenase activity and the yield of pulse crops (Sepetoglu, 1992). However, more than 80% of the added P is fixed or precipitated, and only a part of it goes to soil solution which may be taken up by crops (Leytem and Mikkelsen, 2005) because it rapidly forms insoluble complexes with cations and is incorporated into organic matter by microbes (von Uexkuill and Mutert, 1995; Vance, 2001). Sustainable management of P in agriculture that enhances P acquisition and exploits these adaptations to make plants more efficient at acquiring the nutrient is very important. Therefore, in areas with low soil fertility, with a poor supply and/or high cost of fertilizers, cultivating legumes with high efficiencies of P uptake and P use would be very useful. A two site experiment conducted at Metahara Sugar Estate showed that P application (0 to 60 kg P ha^{-1}) had no significant effect on sugarcane yield (Agricultural Services, 1974). This might be ascribed to the high P precipitation caused by presence of high calcium carbonate (BAI, 2009).

Several key processes which affect the availability of P to plantsin the P cycle are mediated by different types of microbial processes (Richardson and Simpson, 2011). Most researches in microbial inoculants to enhance P availability and root uptake have centered on soil microorganisms capable of solubilizing sparingly-available P (Leggett *et al.*, 2007). Qin *et al.* (2011) demonstrated that soil beneficial microorganisms including rhizobia can solubilize the insoluble form of organic and inorganic P. Increase in productivity of wheat by 30-40% was due to inorganic P application with inoculation, as compared to P alone (Afzal and Asghari, 2008). In addition, enhancement of P utilization from insoluble P through inoculation of rhizobia has been demonstrated in lettuce (Chabot *et al.*, 1996). The objective of this study was thus to evaluate the effect of inoculating selected *Bradyrhizobium* isolates on N and P uptake and P use efficiency of soybean intercropped with sugarcane at Metahara Sugar Estate in the Central Rift Valley of Ethiopia.

Materials and Methods

Description of Experimental Site
A field experiment was conducted at Metahara Sugar Estate under irrigation during 2014/15. The estate is located at 8° 53' N latitude and 39° 52' E longitude

at an altitude of 950 meters above sea level in the Eastern Shewa Administrative Zone, Oromia Regional State, 200 km south-east of the capital city, Addis Ababa, Ethiopia.

The long term mean (LTM) annual rainfall is 551 mm with the LTM annual maximum and minimum air temperatures of 33.0 and 17.5 °C, respectively. According to meteorological information recorded in the last five decades, the rainfall period ranged from April to October albeit the maximum rainfall was recorded in the months of July (127.4 mm) and August (140 mm) in Metahara Sugar Estate.

Most soils of the experimental site are developed under tropical hot condition from alluvium-colluvium parent materials which include basic volcanic rocks such as basalt, limestone, acidic volcanic rocks such as granite, sandstone as well as recent and ancient alluvial soils (Ambachew and Abiy, 2009; BAI, 2009). Soils of Metahara Sugar Estate are classified as Calcaric Cambisols (BAI, 2009).

Experimental Procedures

Carrier based *Bradyrhizobium* inoculants, namely, indigenous isolate (SB6B1) and exotic UK-isolate (Legumefix) were obtained from the Soil Microbiology Laboratory of Holetta Agricultural Research Center and used for seed inoculation at planting. Soybean variety 'Williams' was obtained from Hawassa Agricultural Research Center and intercropped with high yielding and widely cultivated sugar cane variety 'B52-298'.

The experiment consisted of four rates of phosphorus (0, 10, 20 and 30 kg Pha^{-1}) in the form of triple super phosphate (TSP) (0:19:0%; N: P:K) and three types of inoculant inoculation, *i.e.* SB6B1 (local isolate), Legumefix (UK isolate) and uninoculated control. The experiment was laid out in a randomized complete block design (RBCD)in a factorial arrangement and replicated three times per treatment.

Carrier-based inoculants of each isolate were applied at the rate of 10 g inoculant/kg seed (Rice et al., 2001). To ensure that the applied inoculants stick to the seed, the required quantity of inoculants was suspended in 1:1 ratio in 10% sugar solution for 10 minutes.

Land preparation was done by a tractor (ripping, uprooting of old cane stools, disking, leveling and furrowing) and a selected portion of land was then divided into blocks and plots for this experiment. Sugarcane was planted on 21st November 2015 in the furrow trench with end-to-end sett position and 145 cm inter-row spacing. Soybean seed was also sown in the following day at one side of the ridge with the spacing of 10 cm between plants and similar row spacing as

sugarcane on 8.7 m x 5.0 m (43.5 m²) gross plot size, which holds 6 rows of both soybean and sugarcane but data were collected from four central rows. There was a 1 m space between each plot and two furrow (2.90 m) path between blocks, in which no cane was planted.

The experiment was carried out using an irrigated field with furrow irrigation method with an irrigation interval of seven days which was recommended for soybean cultivation. Nitrogen fertilizer at the rate of 20 kg N ha^{-1} was applied as urea (46% N).

Plant Tissue Sampling and Analysis

At physiological maturity, five randomly selected soybean plants were harvested from the four central rows and partitioned into grain and straw. The grain and straw samples were separately oven-dried at 70 $^{\circ}$C to a constant weight, ground to pass through 1 mm sieve and saved for tissue analysis of grain and straw N and P. Total N in grain and straw subsamples were quantitatively determined by a kjeldahl procedure (Bremner and Mulvarey, 1982). Nitrogen content of the grain and straw was determined by multiplying the N concentrations in the dry matter of the tissues by the respective grain and straw dry yields. Phosphorus in grain and straw subsamples were determined by using Meta vanadate method (NSL, 1994). Phosphorus uptake in the grain and straw of soybean was determined from the phosphorus content of the respective parts after multiplying with the grain and straw yields, respectively.

Phosphorus Use Efficiency

Based on the results of plant tissue analysis, phosphorus use efficiency indices were computed (Albrizio et al., 2010).

Agronomic efficiency(AE): is defined as the quantity of grain yield per unit of nutrient applied.

$$AE \ (kg \ kg^{-1}) = \frac{G_f - G_u}{N} \qquad (1)$$

Where:G_f is the grain yield of the fertilized plot (kg), G_U is the grain yield of the unfertilized plot (kg), and N_a is the quantity of P applied (kg).

Physiological efficiency (PE): is defined as the aboveground biomass yield obtained per unit of nutrientuptake.

$$PE \ (kg \ kg^{-1}) = \frac{BY_f - BY_u}{N_f - N_u} \qquad (2)$$

Where: BY_f is the aboveground biomass yield (grain plus straw) of the fertilized plot (kg), BY_u is the aboveground biomass yield (grain plus straw) of the unfertilized plot (kg), N_f is the nutrient uptake (grain plus straw) of the fertilized

plot (kg) and N_u is the nutrient uptake (grain plus straw) of the unfertilized plot (kg).

Agro-physiological efficiency (APE): is defined as the grain yield obtained per unit of nutrient uptake.

$$APE \text{ (kg kg}^{-1}) = \frac{G_f - G_u}{N_f - N_u} \qquad (3)$$

Where: G_f and G_u are grain yieldsfrom fertilized and unfertilized plots (kg), respectively; N_f and N_u are P uptakes (grain plus shoot) from fertilized and unfertilized plots (kg), respectively.

Phosphorus recovery efficiency (PRE): is defined as the quantity of nutrient uptake per unit of nutrient applied.

$$PRE \text{ (\%)} = \frac{N_f - N_u}{N_a} \times 100 \qquad (4)$$

Where: N_f and N_u are nutrient uptakes (grain plus straw) from fertilized unfertilized plots (kg), respectively, and N_a is the quantity of nutrient applied (kg).

Phosphorus Utilization efficiency (PUE): is defined as the product of physiological efficiency and recovery efficiency.

$$PUE \text{ (kg kg}^{-1}) = PE \times PRE$$

Data analysis
Data were subjected to analysis of variance using SAS version 9.1.3 GLM procedure (SAS Institute Inc., 2004). Comparison among treatment means with significant difference for measured and scored characters were made using Least Significant Difference (LSD) at 5% level of significance.

Results and Discussion

Selected soil physical and chemical properties
Analysis of the soil of the experimental field indicated a clayey texture with a clay content of 70%. The soil pH could be rated as moderately alkaline according to the rating of Tekalign (1991) (Table 1). The organic matter content of the soil is low according to the rating of Tekalign (1991). The low organic matter content of the soil might be attributed to the intensive cultivation underway for a long time and continuous removal of crop residues through sugarcane burning. In line with this result, BAI (2009) reported soil organic matter content of Metahara Sugar Estate ranges from low to very low.

Table 1. Physical and chemical properties of soils of the experimental site before planting

Soil property	Value	Soil property	Value
Depth (cm)	20	OM (%)	1.70
Particle size (%)		EC (dSm^{-1})	0.37
Sand	12	Exchangeable Na (cmol$_{(+)}$kg^{-1})	1.89
Silt	18	Exchangeable K (cmol$_{(+)}$kg^{-1})	3.33
Clay	70	Exchangeable Ca (cmol$_{(+)}$kg^{-1})	49.0
Textural class	clay	Exchangeable Mg (cmol$_{(+)}$kg^{-1})	11.0
pH (1:2.5 H$_2$O)	7.70	CEC (cmol$_{(+)}$kg^{-1})	67.0
TN (%)	0.12	PBS (%)	97.3
Avail. P (ppm)	5.60	CaCO$_3$ (%)	7.00

Note: P: Available phoshorus, CEC: Cation exchange capacity, EC: Electrical conductivity, OM: Organic matter, PBS: Percent base saturation, TN: Total nitrogen

The analysis further indicated that the soil has low contents of total nitrogen (Tekalign, 1991) and available phosphorus (Marx et al., 1996) (Table 1). The low nitrogen content could be attributed to the low soil organic matter content. The low available P could be ascribed to the precipitation of phosphorus into unavailable forms of calcium and magnesium carbonates. Consistent with this result, BAI (2009) reported low available phosphorus in the Estate because of high P precipitation. Beside, the cation exchange capacity (CEC) of the soil was rated in the range of very high as reported by Landon (1991) with the dominant cation being calcium in the exchange site. The high CEC of this soil might be attributed to the high clay content in the soil. Percent calcium carbonate (CaCO$_3$) content (7.0%) was moderate according to the rating of Nachtergaele *et al*. (2009)

Nitrogen content and P uptake of soybean

Nitrogen contents of grain, straw and total biomass

Inoculation with *Bradyrhizobium* significantly (P <0.01) influenced the N contents of the grain and straw as well as the total biomass of soybean (Table 2). *Bradyrhizobium* inoculation alone improved the whole N content of soybean regardless of P application (Table 2). Both SB6B1 and Legumefix inoculations significantly increased grain, straw and total N contents over the uninoculated control. The highest mean N contents of grain, straw and total biomass yield were obtained from inoculation with SB6B1 isolate albeitno significant variation was observed between the two *Bradyrhizobium* inoculants. SB6B1 increased grain, straw and total biomass contents of N by 6, 14 and 8%, respectively compared to Legumefix while the respective increase over uninoculated control was higher by 147, 97 and 130%. An increase in N contents due to *Bradyrhizobium* inoculation could be related to the significant increase in nodulation resulting in higher accumulation of N through biological N$_2$ fixation (Siczek and Lipiec, 2011).

This current result is in agreement with the findings of Tahir et al. (2009) who reported that soybean N accumulation in grain, straw and total biomass was increased by 9, 122 and 76% over the control due to inoculation with *Bradyrhizobium*. This result is also in accord with the finding of Tajini *et al.* (2011) who reported that inoculation with rhizobia improved symbiotic N_2 fixation even under phosphorus deficiency. In line with this result, Tufenkci *et al.* (2006) reported that *Rhizobium* inoculation improved NPK uptake.

Table 2. Effect of *Bradyrhizobium* inoculation and P application on N content and P uptake in grain and straw (kg ha⁻¹) of soybean intercropped with sugarcane at Metahara Sugar Estate.

Treatment	Nitrogen			Phosphorous		
	Grain N	Straw N	Total N (grain + straw)	P uptake by grain	P uptake by straw	Total P uptake
P rate (kg ha⁻¹)						
0	71	35.2	102.2	11.5	5.3	16.8[b]
10	82.9	36.6	119.5	13.8	5.9	19.7[ab]
20	85.2	31.1	116.3	11.5	6	17.4[b]
30	84.1	37.3	121.4	15.8	6.9	22.7a
Significance	NS	NS	NS	NS	NS	*
LSD (0.05)	NS	NS	NS	NS	NS	3.93
Inoculation						
Uninoculated	41.8[b]	22.4[b]	64.2[b]	10.0[b]	5.1[b]	15.0[b]
SB6B1	103.2[a]	44.1[a]	147.4[a]	16.1[a]	6.7[a]	22.8[a]
Legumefix	97.4[a]	38.7[a]	136.1[a]	13.34[a]	6.3[a]	19.6[a]
Significance	**	**	**	**	*	**
LSD (0.05)	11.7	6.7	11.52	3.31	1.12	3.4
CV (%)	17	22.5	11.7	29.8	22	21

*Where: NS, * and **: Non significant, significant at 5 and 1%, respectively; CV: Coefficient of variation, LSD: Least significant difference. Means within the same factor and column followed by the same letter are not significantly different at 5% level of significance.*

Phosphorus fertilizer rate and its interaction with inoculants did not significantly affect the N contents of the grain and straw as well as the total biomass at physiological maturity although soil available P was low (Table 2). This might be due to the high alkalinity of the soil which predisposes the available P in the soil to precipitation into unavailable forms (BAI, 2009). However, a slight increase in total plant N content was obtained due to small P application (10 kg Pha⁻¹) with SB6B1 and Legumefix inoculation as compared to uninoculated treatment (Figure 1a).This shows that application of 10 kg P ha⁻¹ is optimum for sufficient uptake of nitrogen by the crop. Consistent with this result, Tekle and Walelign (2014) reported that P significantly increased the soybean grain N and straw N contents at lower P rate (25 kg P ha⁻¹) than at higher level (50 kg P ha⁻¹).
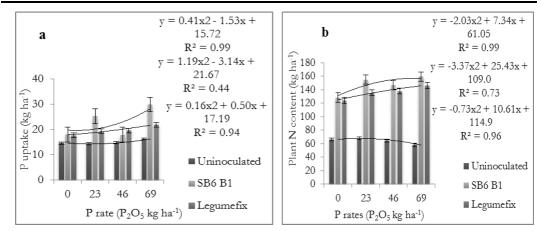
Figure 1. The regression analysis of the response of (a) P uptake (kg ha^{-1}) and (b) Plant N content (kg ha^{-1}) to P application rates (kg P$_2$O$_5$ ha^{-1})

Uptake of P in Grain, Stover and Total Biomass of Soybean

Inoculation with *Bradyrhizobium* had significant (P<0.01) effect on the uptake of P by grain, straw, and total soybean biomass compared to the uninoculated treatment. Phosphorus uptake by grain, straw, and total biomass in response to inoculation with SB6B1 as well as in response to relative to inoculation with Legumefix was significantly higher than the uptake observed in response to no inoculation. Thus, the total P uptake that resulted from inoculation with SB6B1 and Legumefix exceeded the P total uptake obtained in response to no inoculation by about 52 and 31%, respectively (Table 2). The higher P uptake due to inoculation with SB6B1and Legumefix could be attributed to the fact that some rhizobia have the ability to solubilize precipitated P components, thereby increasing the uptake in plants (Qin *et al*., 2011). Consistent with the results of this study, the finding of Taye (2006) showed that except P uptake in the straw, inoculation of pea by *Rhizobium* significantly increased both grain and total P uptake. Similarly, Tahir *et al*. (2009) reported that *Rhizobium* inoculation increased total P uptake by 79%. Havlin *et al*. (1999) also indicated that large quantities of P are found in seed and P is considered to be essential for seed formation.

Improved N status in soybean plants due to better root growth might be the mechanism by which soybean P uptake was increased in plants inoculated with the effective *Rhizobium* strains on low-P acid soils (Neila *et al.,* 2014). Cheng et al. (2008) found that inoculating soybean with effective rhizobial inoculants significantly improved root growth as well as N and P contents in low-Pacidic soils. In addition, Tang *et al.* (2007) found that total P uptake from sparingly

soluble P correlated highly with plant biomass production, N$_2$ fixation and nodulation, and seed P concentrations. Singh et al. (2005) found that inoculation

of P solubilizing bacteria increased P content in grain and straw by 10.72 and 31.94%, respectively, over the uninoculated treatment.

In contrast to the main effect of inoculation, P rates and its interaction with inoculation had no significant effect on grain and straw P uptake at physiological maturity (Table 2). However, total P uptake was significantly ($P \leq 0.05$) increased in response to the increase in the rate of phosphorus application. The maximum total P uptake was recorded due to the applications of 10 and 30 kg P ha^{-1}). Similarly, Egamberdiyeva *et al*. (2004) confirmed that P uptake by soybean increased with the increase in the rate of phosphorus application in N-deficient calcareous soils. Apparently, a similar trend was also reported by BAI (2009) who found that soils of Metahara Sugar Estate were alkaline and strongly calcareous and that the organic matter and total N contents were low. Among the tested isolates, SB6B1 inoculation showed significantly higher total P uptake at 30 kg P ha^{-1} than the other rates of P application though no interaction effect was observed (Figure 1b).

Phosphorus use efficiency

Agronomic efficiency

The higher the rates of P application, the lower were the agronomic efficiency in all observed treatments. Across P rates, a 2.8, 8.4 and 7.7kg soybean grain yield was produced per unit of P applied by un-inoculated, SB6B1 and Legumefix inoculation, respectively. The highest agronomic efficiency (AE) of 3.9,13.6 and 11.5 kg kg^{-1}was obtained at 10 kg Pha^{-1} application coupled with un-inoculated, SB6B1 and Legumefix inoculation (Table 3, 4 and 5). However, the least AE value was noted at 30 kg Pha^{-1} in all treatments. Application of P fertilizer above 10 kg P ha^{-1} had no appreciable effect on soybean grain yield. Nonetheless, the AE was more influenced by soybean inoculation than un-inoculated treatments. This might be due to the fact that rhizobial symbiosis requires large amounts of P to meet the high energy costs for adenosine triphosphate (ATP) synthesis (Tang et al., 2001)in order to produce higher grain yield. This agrees with Gifole *et al*. (2011) who found a declining trend of AE from 69.8 to 9.3 kg kg^{-1} at the P rates ranging from 10 to 60 kg P ha^{-1} on haricot bean. This might be due to small amounts of applied fertilizer optimized nutrient use efficiency (Bationo and Buerkert, 2001).Similar to this result, the combined application of phosphorus and inoculation enhanced agronomic efficiency of soybean and common bean over the un-inoculated control (Devi et al., 2012; Anteneh, 2014).

Table 3. Phosphorus use efficiencies of soybean intercropped with sugarcane as affected by P application

P (kg ha⁻¹)	Uninoculated				
	AE (kg kg⁻¹)	PE (kg kg⁻¹)	APE (kg kg⁻¹)	PRE (%)	PUE (kg kg⁻¹)
0	-	-	-	-	-
10	3.9	21.2	21.2	3.1	0.7
20	2.6	25.7	25.7	3.2	0.8
30	2.0	28.8	28.9	2.6	0.8
Mean	2.8	25.2	25.3	3.0	0.8

Note: AE: Agronomic efficiency, PE: Physiological efficiency, APE: Agro-physiological efficiency, PRE: Phosphorus recovery efficiency and PUE: Phosphorus utilization efficiency

Table 4. Phosphorus use efficiencies of soybean intercropped with sugarcane as affected by SB6B1 inoculation

P_2O_5 (kg ha⁻¹)	SB6B1				
	AE (kg kg⁻¹)	PE (kg kg⁻¹)	APE (kg kg⁻¹)	PRE (%)	PUE (kg kg⁻¹)
0	-	-	-	-	-
10	13.6	33.2	42.8	31.83	10.58
20	6.7	35.5	37.99	17.74	6.30
30	4.8	40.9	27.83	17.36	7.10
Mean	8.4	36.6	36.2	22.3	8.0

Table 5. Phosphorus use efficiencies of soybean intercropped with sugarcane as affected by Legumefix inoculation.

P_2O_5 (kg ha⁻¹)	Legumefix				
	AE (kg kg⁻¹)	PE (kg kg⁻¹)	APE (kg kg⁻¹)	PRE (%)	PUE (kg kg⁻¹)
0	-	-	-	-	-
10	11.5	98.2	161.7	7.1	7.0
20	7.1	114.5	140.2	5.1	5.8
30	4.5	117.2	75.0	6.1	7.1
Mean	7.7	110.0	125.6	6.1	6.6

Physiological Efficiency

The physiological efficiency (PE) indicates the biological yield obtained per unit of nutrient uptake. Along P rates slight increase in biomass accumulation was observed and maximum biomass yield was obtained at application rate of 30 kg P ha⁻¹ in all treatments (Tables 3, 4 and 5).Across P rates on average 25.2, 36.6 and 110.0 kg biomass yields were noted per 1 kg of applied P with respect to un-inoculated, SB6B1 and Legumefix. Legumefix inoculation led to higher biomass yield and P uptake than SB6B1 throughout P application rates, whereas the lowest PE was recorded by un-inoculated control. The higher PE fraction obtained due to Legumefix inoculation might indicate its tendency to accumulate relatively higher biomass yield as P fertilizer rates increase with small amounts of

increase in total P uptake. This could also be due to better symbiotic N_2 fixation with Legumefix inoculation thereby increasing the response of soybean to P application (Singleton *et al.,* 1984). Moreover, it might have produced hormones and solubilizing insoluble P from the soil (Sobral *et al*., 2004; Singh *et al*., 2005).The slight increase in dry biomass yield at higher P fertilizer application rate indicated that the plants grown at the lowest P level were the most efficient in using P for the production of dry matter (Win *et al*., 2010).

Agro-physiological Efficiency

Agro-physiological efficiency (APE) is the economic production (grain yield) obtained per unit of nutrient uptake. Along P application rates, APE drastically decreased in the inoculated treatments but showed slight increment in the un-inoculated control albeit it scored the lowest APE compared to the inoculated ones. Across P application rates on average 25.3, 36.2 and 125.6 kg grain yield was obtained per unit of nutrient absorbed in un-inoculated, SB6B1 and Legumefix, respectively. The highest agro-physiological efficiency of 42.8 and 161.7 kg kg-1 was noted at the lowest P rate of 10 kg P ha-1with SB6B1 and Legumefix, respectively (Table 3, 4 and 5).The higher APE by Legumefix inoculation might be due to the presence of plant growth promoting characteristics in addition to N_2 fixation which enabled to produce relatively higher nutrient uptake at lower P rate fertilizer. Similar results were reported by Singh *et al.* (2005) in lentil. Contrary to this result, Abbasi *et al*. (2010) found the highest (51 kg ha-1) APE for soybean at lower P fertilizer application rate (50 kg P_2O_5 ha-1) than at higher rate of 100 kg P_2O_5 ha-1which produced APE of 42 kg kg-1. In alkaline soil pH, the availability of some essential nutrients for plant is reduced (Maschner, 2011).

Phosphorus Recovery Efficiency

Phosphorus recovery efficiency (PRE) provides the quantity of nutrient uptake per unit of nutrient applied. The mean recovery efficiency of P by soybean treated with un-inoculated, SB6B1 and Legumefix were3.0, 22.3 and 6.1%, respectively. The highest recovery efficiency of 31.8 and 7.1% were noted due to SB6B1and Legumefix inoculation at the lowest P rate of 10 kg ha-1. However, the lowest recovery efficiency was noted at 30 kg P ha-1 with SB6B1 and un-inoculated, and at 20 kg P ha-1 with Legumefix inoculation (Tables 3, 4 and 5). The higher recovery efficiency by SB6B1 inoculation might be due to the fact that large number of strains of *Rhizobium* and *Bradyrhizobium* could solubilize inorganic phosphate through the enzymatic action of acid and alkaline phosphatase (Halder and Chakrabartty, 1993) and assimilate the soluble P in plants and prevent it from adsorption or fixation (Khan and Joergensen, 2009).

The lower recovery efficiency was also reported by Abbasi *et al*. (2010) who found PRE of 12.1% in soybean at lower P rate(10kg Pha-1) and 10.2% when dose

increased to20kg Pha⁻¹. Syers *et al*. (2008) also reported that in the year of fertilizer application P fertilizer used by plants ranged from 10–30%. The low P recovery efficiency in the present study might be associated with high P fixation property of the soil due to the presence of Ca compounds and clay minerals. Besides, P sorption increases at higher fertilizer rates than at lower application (Chaudhary et al., 2003). Kumar and Kairon (1980) also determined an apparent P recovery of 4.7% by field grown cotton in alkaline soils. Beside this, Fixen (2004) concluded that first year recovery of P is low, not only because the P is immediately "fixed" into plant unavailable forms but also because it moves so little in soils that crop roots are too far from much of the fertilizer-soil reaction zones to be accessed.

Phosphorus utilization efficiency (PUE)

As shown in Table 3, 4 and 5 the efficiency of soybean in P utilization inconsistently decreased as the P fertilizer rate increased. On the average, every kilogram of P applied to the un-inoculated, SB6B1 and Legumefix treated soybean produced 0.8, 8.0 and 6.6 kg of grain yield respectively. The highest P utilization efficiency was observed at 10 kg P ha⁻¹due to SB6B1 and at 30 kgP ha⁻¹with Legumefix inoculation. However, the lowest PUE was noted at 20 kg P ha⁻¹with SB6B1 and Legumefix, and at 10 kg P ha⁻¹with un-inoculated treatments. It is evident from the result that inoculation enhanced PUE of soybean where better numerical values were attained from SB6B1, followed by Legumefix. This could be due to the fact that symbiotic N_2 fixation is an energy consuming process with a high (16) ATP demand for the reduction of one molecule of N_2 into $2NH_3$ (Schulze *et al.*, 2006).Phosphorus application also enhanced growth of rhizobial strains and host plants (Munns et al., 1981; Leung and Bottomley, 1987). Win et al. (2010) reported the declining trend in PUE as P rate increased from 0.5 to 2 m*M*P, which is in in agreement with the results of this study. by . Singh et al. (2005) also reported that the highest PUE was obtained from the lowest P rate with inoculation of P solubilizing bacteria. y. However, P application did not improve PUE significantly in uninoculated treatment.

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