Journal of Advances in Microbiology

3(3): 1-11, 2017; Article no.JAMB.33084
ISSN: 2456-7116

Influence of *Bradyrhizobium* and Mycorrhiza on Growth, Yield and Phosphorus Use Efficiency on Soybean under Manure Application

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Authors' contributions

This work was carried out in collaboration between both authors. Author OAB designed the study, performed the statistical analysis, wrote the protocol and author MOA wrote the first draft of the manuscript. Authors OAB and MOA managed the analyses of the study. Author MOA managed the literature searches. Both authors read and approved the final manuscript.

Article Information

DOI: 10.9734/JAMB/2017/33084
Editor(s):
(1) Pongsak Rattanachaikunsopon, Department of Biological Science, Faculty of Science, Ubon Ratchathani University, Ubon Ratchathani 34190, Thailand.
Reviewers:
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(3) Arun Karnwal, Lovely Professional University, India.
Complete Peer review History: http://www.sciencedomain.org/review-history/19356

Original Research Article

Received 30th March 2017
Accepted 13th May 2017
Published 5th June 2017

ABSTRACT

A field experiment was conducted at the Federal University of Agriculture, Abeokuta, South West Nigeria, to determine the influence of *Bradyrhizobium* and mycorrhiza on growth, yield and phosphorus use efficiency of soybean (*Glycine max* L.) under manure application. The treatments consisted of three factors, *Bradyrhizobium* (with or without), Phosphorus source (Single super phosphate) and poultry manure (0, 5, 10 ton/ha). The variety of soybean worked on was TX 1448-2E. The influence of these treatments on plant height shows, poultry manure of 0, 5 and 10 ton/ha had no significant difference at 4 – 12 week after planting (WAP) but at 14 WAP plots which received 0 ton/ha had highest values. Also, for Phosphorus source, they shows no differences at 6 WAP while at 4, 8, 10, 12 and 14 WAP plants that received mycorrhiza shows a greater influence on the plant height. Plot with *Bradyrhizobium* show no differences at 4 and 14 WAP. The

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application of mycorrhiza as a P source had great influence on soybean production under manure application. Also, the use of poultry manure had an influence on soybean at 5-10 ton/ha. Similarly, on Phosphorus use efficiency, the application of the treatments shows significant differences at 6 WAP where plots that received mycorrhiza with 10 ton/ha of poultry manure showed higher values at p <0.05. However, there were no significant differences on the treatments at 8 WAP, but plots that received *Bradyrhizobium*, mycorrhiza with 10 tons of poultry manure showed higher values at p<0.05.

Keywords: Bradyrhizobium; mychorriza; single super phosphate; poultry manure; soybean.

1. INTRODUCTION

Soybean [*Glycine max* (L.)] (Faba beans) is a globally important oilseed crop and source of high quality protein for human consumption, used as fodder for animal and is also important in improved crop rotation systems when in symbiotic association with *Bradyrhizobium* species [1]. Soybean is one of the most valued oilseed and protein rich crops in the world. The crop is known by its highest protein content (approximately 40%) and the highest gross output of vegetable oil (approximately 20%) among the cultivated crops in the world [2]. The grain is also a good source of calcium, phosphorus, copper, potassium, magnesium and thiamine and used for biodiesel and glycerol production [3]. Besides its stated purposes, it improves soil properties and soil biological health through its proliferated and deep tap-root system, soil nitrogen enrichment through biomass addition and N_2 fixation [4]. World soybean productivity reached approximately 2.5 ton ha^{-1} with an area coverage and total production of 111.5 million hectares and 276.03 million tons, respectively [5]. In soils where the soybean crop has not been grown previously comparable populations of *Bradyrhizobium* are seldom available [1]. Soybean has been cultivated all over the world since old times for its high protein and lipid content. Its earlier uses have varied from a green manure crop to a forage crop and a N_2 fixing crop due to its ability to fix substantial quantities of atmospheric nitrogen in association with nodule-forming bacteria (*Bradyrhizobium*) [4]. The nitrogen demand of soybean can be supplied via biological nitrogen fixation through the inoculation with selected *Bradyrhizobium japonicum/ B. elkanii* strains. Biofertilizers such as *Rhizobium* and mycorrhiza, are steadily receiving increased attention and recognition from scientists. This could be attributed to the fact that they pose no ecological threats, usually have a longer lasting effect and if properly managed can out-yield recommended doses of chemical fertilizers [6]. It has been found that inoculation of soybean with *Bradyrhizobium japonicum* significantly increased nodulation, yield and seed quality [7,8]. Efforts throughout the world are directed towards improving the quality of food crops by increasing the nutritional value of grains and decreasing the anti-nutrients level. Breeding, fertilization program and genetic engineering are directed towards improving seed quality. *Rhizobium* inoculation of Faba beans was reported to increase yield and protein content [9]. The presence of indigenous Rhizobium, soil nitrogen availability, soil physicochemical constraints and climatic conditions were reported to influence the ability of the crop to achieve maximum nitrogen fixation through inoculation [10] and consequently improved the crop yield. *B. japonicum* has the ability to enhance the growth of *G. max* in that soil where bacteria are not available for nitrogen fixation [11]. Inoculation of seeds of *G. max* with *B. japonicum* favours to survive bacteria on the seeds surface before planting. *B. japonicum* infection starts in the root of the *G. max* after the emergences of radicles. Vesicular-arbuscular mycorrhizal (VAM) fungi form symbiotic association which enhances water and nutrient transport particularly phosphorus (P) and thereby increase growth and yield of many crop plants. In recent years the effect of combined inoculation with VAM-fungi and Rhizobia have been reported to further increase the growth and yield of some crops including soybean [12]. The tripartite association of symbiotic VAM-fungi, root nodule bacteria, *B. japonicum* was investigated to find its effect on the promotion of growth and yield of soybean var. William 82 and NARC-II. Application of poultry litter as a source of N and P has been shown to increase yields of crops such as corn and pasture [13]. Therefore, the objective of this work is to examine the effects of *Bradyrhizobium* and mycorrhiza on growth, yield and phosphorus use efficiency on soybean under manure application.

2. MATERIALS AND METHODS

2.1 Experimental Location

The experiment was carried out in an experimental field in the Federal University of Agriculture, Abeokuta.

Single super phosphate and Soybean were obtained from FUNAAB, the variety is TGx 1448-2E, *Bradyrhizobium japonicum* (HISTICK) was obtained from IITA and Mycorrhiza was obtained from IITA but was cultured in FUNAAB. Poultry Manure was obtained from FUNAAB poultry unit.

The experimental design was a Randomized Complete Block Design (RCBD), replicated three times. The treatments consisted of combination of three (3) factors – *Bradyrhizobium* (with or without) P source (SSP, Mycorrhiza) and poultry manure (0, 5, and 10 kg/ha).

2.2 Planting and Cultural Practices

The land was 25 m by 25 m and divided into 36 sub-plots of dimension 3 m by 3 m each. Manual clearing and stumping was carried out on the land. The stumping was done to ensure that, there were no roots inside the soil which can stunt the growth of the crop planted. Planting was done by drilling two weeks after the incorporation of the poultry manure on the field prepared.

2.3 Determination of Soil Physical and Chemical Properties

Soil sample of 5 kg was air dried at room temperature, ground and passed through a 2 mm sieve for soil physical and chemical analysis. Particle size analysis was determined using the hydrometer method as described by [14], pH was determined in a 1:1 soil to water suspension using a pH meter with glass electrode; total N was determined using macro-Kjedhal digestion technique of [15]; Organic C using wet oxidation method. Available P was extracted using Bray-1 method [16] and determined colorimetrically. Exchangeable acidity was determined by titrimetry [16]. Cation Exchange Capacity (CEC) was determined by summation of total exchangeable bases and total acidity while exchangeable bases were extracted with 1 N ammonium acetate buffer at pH 7.0. Exchangeable Na^+ and K^+ in the extract were determined by flame photometry while Ca^{2+} and Mg^{2+} were determined using AAS [16].

2.4 Statistical Analysis

Data collected was subjected to analysis of variance (ANOVA) using the SAS package (Statistical Analysis System). The differences of means were identified by least significant differences (LSD) at $p < 0.05$.

3. RESULTS AND DISCUSSION

3.1 Physical and Chemical Properties of Soil

Table 1 showed some physical properties of the soil used for this experiment. The soil pH was neutral and it has low N, Mg, K, Na and organic carbon and organic matter. The exchangeable acidity was 1.31.

Table 1. The physical and chemical properties of the soil used

Properties	Values
Sand	91.0 %
Silt	4.6 %
Clay	4.4 %
Textural class	Sandy
pH	6.8
Nitrogen	1.4 %
Available P (mg/kg)	17.73
Organic carbon	0.6 %
Organic matter	1.03 %
Ca (cmol/kg)	0.64
Mg (cmol/kg)	0.58
K (cmol/kg)	0.18
Na (cmol/kg)	0.21
Exchangeable acidity (mol/kg)	1.31
Cation exchange capacity (cmol/kg)	1.61

3.2 Some Chemical Composition of Poultry Manure Used

Table 2 showed some chemical composition of poultry manure used for the experiment before planting. This has high nitrogen and phosphorus percentage but low potassium percentage. However, the pH of the poultry manure was neutral.

3.3 Effect of the Treatments on Plant Height from 4 – 14 WAP

Table 3 showed the effect of poultry manure, P source and *Bradyrhizobium* on plant height from

4 – 14 WAP. Poultry manure at 0, 5 and 10 ton/ha had no significant difference on plant height at 4 – 12 WAP while 0 ton/ha have higher value at 14 WAP.

Table 2. Chemical properties of poultry manure used

Properties	Values
N	3.57%
K	1.70%
P	1.91%
pH	6.8

However, P source had no significant difference at 6 WAP but at 4, 8, 10, 12 and 14 WAP plots treated with mycorrhiza had significantly higher values than those treated with single super phosphate (SSP). Also, plots treated with *Bradyrhizobium* and those without *Bradyrhizobium* had no significant differences at 4 and 14 WAP but at 6, 8, 10 and 12 WAP differences occurred, where plots without *Bradyrhizobium* had higher values.

3.4 Interactive Effects of Treatments on Plant Height from 4 – 12 WAP

Table 4 showed the effect of poultry manure, P source and *Bradyrhizobium* on plant height from 4 – 14 WAP. The application of the treatments shows significant differences ($p<0.05$) on plant height from 4 to 14 WAP.

3.5 Effect of Treatments on Stem Girth

Table 5 showed the effect of poultry manure, P source and *Bradyrhizobium* on the stem girth of the soybean planted. Application of poultry manure at 0, 5, and 10 ton/ha had no significant difference at 4, 6, 8, 10, and 14 WAP on the plant stem girth, while 10 ton/kg significant difference on the plant stem at 4 – 14 WAP and also, plots treated with *Bradyrhizobium* and those without *Bradyrhizobium* had no significant difference on plant stem girth at all at 4 – 14 WAP.

3.6 Interactive Effects of Treatments on Stem Girth from 4 – 12 WAP

Table 6 showed the interaction between poultry manure, P source and *Bradyrhizobium* on the stem girth of the soybean planted. The application of the treatments had no significant differences at 4 – 8 WAP. However, at 10 and 12 WAP treatments showed significant differences ($p<0.05$).

3.7 Effects of the Treatments on Leaf Area

Table 7 showed the main effect of poultry manure, P source and *Bradyrhizobium* on leaf area at 4 – 14 WAP. Plots that was amended with poultry manure at 0, 5, and 10 ton/ha had no significant difference at 4 and 12 WAP but at 6, 8, 10 and 14 WAP there was differences in their leave area with the plot amended with 10 ton/ha of poultry manure having significantly lower values. In addition, for P sources, there were no significant differences between them at 4, 6, 10, 12 and 14 WAP statistically whereas there was a significant difference at 8 WAP, but plots amended with mycorrhiza have higher values throughout the 4 – 14 WAP.

However, at 4, 6, 12, and 14 WAP plots treated with *Bradyrhizobium* and those without *Bradyrhizobium* had no significant differences but

Table 3. The main effect of poultry manure, P source and *Bradyrhizobium* on plant height from 4 – 14 WAP (cm)

Treatments	4 WAP	6 WAP	8 WAP	10 WAP	12 WAP	14 WAP
Poultry manure source						
0 (ton)	12.22^a	21.55^a	29.58^a	35.20^a	42.78^a	47.99^a
5 (ton)	12.39^a	22.12^a	30.54^a	36.22^a	42.89^a	46.34^a
10 (ton)	12.94^a	21.29^a	29.24^a	34.15^a	40.85^a	44.13^b
P value	0.18	0.37	0.39	0.19	0.34	0.04
Phosphorus source						
Mycorrhiza	13.23^a	22.09^a	30.69^a	36.32^a	43.56^a	48.11^a
SSP	11.80^b	21.21^a	28.89^b	34.07^b	40.79^b	44.19^b
P value	0.00	0.10	0.02	0.01	0.03	0.00
***Bradyrhizobium* source**						
Bradyrhizobium-	12.28^a	22.24^a	31.03^a	36.42^a	44.99^a	47.00^a
Bradyrhizobium+	12.75^a	21.06^b	28.50^b	33.97^b	36.35^b	45.30^a
P value	0.20	0.02	0.00	0.01	0.00	0.18

Table 4. Interaction effect of factors on plant height of soybean (cm)

Treatments	4 WAP	6 WAP	8 WAP	10 WAP	12 WAP
Brad/Myc/10PM	19.27^{e}	12.84^{ef}	36.99^{abc}	38.47^{cd}	41.73^{cd}
Brad/SSP/10PM	22.62^{abc}	31.97^{abc}	36.99^{abc}	38.53^{cd}	44.49^{bc}
Myc/10PM	24.31^{a}	34.76^{a}	40.93^{a}	52.15^{a}	53.32^{a}
SSP/10PM	19.27^{e}	24.40^{f}	28.29^{c}	27.06^{d}	37.05^{d}
Brad/Myc/5PM	22.11^{abcd}	30.81^{bcd}	37.87^{abc}	38.47^{cd}	45.65^{bc}
Brad/SSP/5PM	21.90^{bcd}	28.65^{cd}	34.11^{abc}	45.70^{b}	46.71^{bc}
Myc/5PM	22.85^{ab}	32.19^{abc}	36.67^{bc}	47.05^{ab}	48.67^{ab}
SSP/5PM	21.62^{bcd}	30.52^{bcd}	36.25^{c}	40.34^{cd}	44.03^{bc}
Brad/Myc	20.02^{de}	27.38^{def}	32.18^{de}	38.77^{cd}	45.65^{bc}
Brad/SSP	20.41^{cde}	26.61^{ef}	32.30^{de}	39.00^{cd}	47.29^{bc}
Myc	23.99^{ab}	33.16^{ab}	39.90^{ab}	49.32^{ab}	53.67^{a}
SSP	21.76^{bcd}	31.18^{abc}	36.47^{bc}	40.32^{bc}	45.35^{bc}

Table 5. The main effect of poultry manure, P source and *Bradyrhizobium* on stem girth at 4 – 14 WAP (cm)

Treatments	4 WAP	6 WAP	8 WAP	10 WAP	12 WAP	14 WAP
Poultry manure source						
0 (ton)	0.10^{a}	0.15^{a}	0.21^{a}	0.28^{a}	0.37^{a}	0.46^{a}
5 (ton)	0.10^{a}	0.27^{a}	0.23^{a}	0.27^{a}	0.36^{a}	0.44^{a}
10 (ton)	0.10^{a}	0.74^{a}	0.22^{a}	0.27^{a}	0.34^{a}	0.45^{a}
P value	0.16	0.18	0.20	0.51	0.10	0.10
Phosphorus source						
Mycorrhiza	0.10^{a}	0.37^{a}	0.22^{a}	0.27^{a}	0.36^{a}	0.46^{a}
SSP	0.10^{a}	0.40^{a}	0.22^{a}	0.28^{a}	0.35^{a}	0.44^{a}
P value	0.16	0.90	0.20	0.77	0.85	0.10
***Bradyrhizobium* source**						
Bradyrhizobium-	0.10^{a}	0.15^{a}	0.21^{a}	0.28^{a}	0.35^{a}	0.46^{a}
Bradyrhizobium+	0.10^{a}	0.40^{a}	0.21^{a}	0.27^{a}	0.36^{a}	0.44^{a}
P value	0.16	0.94	0.10	0.38	0.25	0.10

Table 6. Interactive effect of treatment on girth of soybean (mm)

Treatments	4 WAP	6 WAP	8 WAP	10 WAP	12 WAP
Brad/Myc/10PM	0.1^{a}	0.1^{a}	0.21^{a}	0.27^{ab}	0.35^{ab}
Brad/SSP/10PM	0.1^{a}	0.1^{a}	0.15^{a}	0.26^{b}	0.35^{ab}
Myc/10PM	0.1^{a}	0.1^{a}	0.17^{a}	0.26^{b}	0.35^{ab}
SSP/10PM	0.1^{a}	0.1^{a}	0.22^{a}	0.28^{ab}	0.32^{b}
Brad/Myc/5PM	0.1^{a}	0.1^{a}	0.14^{a}	0.26^{b}	0.35^{ab}
Brad/SSP/5PM	0.1^{a}	0.1^{a}	0.22^{a}	0.28^{ab}	0.37^{ab}
Myc/5PM	0.1^{a}	0.1^{a}	0.22^{a}	0.26^{a}	0.35^{ab}
SSP/5PM	0.1^{a}	0.1^{a}	0.16^{a}	0.28^{ab}	0.36^{ab}
Brad/Myc	0.1^{a}	0.1^{a}	0.17^{a}	0.27^{ab}	0.36^{ab}
Brad/SSP	0.1^{a}	0.1^{a}	0.17^{a}	0.27^{ab}	0.36^{ab}
Myc	0.1^{a}	0.1^{a}	0.14^{a}	0.26^{b}	0.40^{a}
SSP	0.1^{a}	0.1^{a}	0.15^{a}	0.27^{ab}	0.33^{ab}

at 8 and 10 WAP they shows differences where plots without *Bradyrhizobium* had higher values.

3.8 Interactive Effects of the Treatments on Leaf Area

Table 8 showed the interaction between the poultry manure, P source and *Bradyrhizobium* on leaf area at 4 – 12 WAP. The application of the treatments showed significant differences at 4 – 12 WAP.

3.9 Influence of the Treatments on Plant Phosphorus Concentration

Table 9 showed the influence of poultry manure, P source and *Bradyrhizobium* on plant phosphorus concentration at 6 and 8 WAP. Application poultry manure at 0, 5, and 10 ton/ha showed no significant differences at 6 and 8 WAP but higher values were observed with 10 kg/tons at 6 and 8 WAP. There were no significant differences in P source but SSP shows higher values at 6 WAP and mycorrhiza gives higher value at 8 WAP. Similarly, higher values were noticed at 6 and 8 WAP in plots that had application of *Bradyrhizobium* over those without *Bradyrhizobium* but statistically there was no significant difference.

Table 7. The main effect of poultry manure, P source and *Bradyrhizobium* on leaf area at 4 – 14 WAP (cm^2)

Treatments	4 WAP	6 WAP	8 WAP	10 WAP	12 WAP	14 WAP
Poultry manure source						
0 (ton)	5.58^{a}	13.81^{a}	23.69^{ab}	29.78^{ab}	38.32^{a}	41.83^{a}
5 (ton)	5.47^{a}	13.41^{ab}	25.77^{a}	32.64^{a}	36.69^{a}	43.43^{a}
10 (ton)	5.58^{a}	13.81^{a}	23.69^{ab}	29.78^{ab}	38.32^{a}	38.28^{b}
P value	0.97	0.12	0.03	0.00	0.26	0.02
Phosphorus source						
Mycorrhiza	5.58^{a}	13.34^{ab}	25.13^{a}	30.89^{a}	38.57^{a}	41.91^{a}
SSP	5.45^{a}	12.93^{ab}	23.39^{b}	30.78^{a}	38.05^{a}	40.45^{a}
P value	0.73	0.55	0.02	0.12	0.09	0.36
***Bradyrhizobium* source**						
Bradyrhizobium-	5.73^{a}	13.65^{a}	22.02^{a}	31.30^{a}	37.65^{a}	41.99^{a}
Bradyrhizobium+	5.31^{a}	13.61^{a}	12.50^{b}	28.38^{ab}	35.96^{a}	40.36^{a}
P value	0.25	0.13	0.00	0.03	0.24	0.30

Table 8. Interactive effect of treatment on leaf area of soybean (cm^2)

Treatments	4 WAP	6 WAP	8 WAP	10 WAP	12 WAP
Brad/Myc/10PM	4.09^{d}	9.57^{d}	19.39^{d}	21.48^{d}	28.70^{b}
Brad/SSP/10PM	7.31^{ab}	13.45^{bcd}	22.67^{abcd}	30.16^{abc}	35.33^{a}
Myc/10PM	6.45^{abc}	15.01^{ab}	28.44^{a}	33.64^{ab}	41.91^{a}
SSP/10PM	4.17^{d}	10.67^{dc}	17.82^{d}	23.00^{d}	35.71^{a}
Brad/Myc/5PM	4.84^{cd}	13.71^{abc}	27.94^{a}	34.86^{a}	36.10^{a}
Brad/SSP/5PM	5.73^{abcd}	14.01^{abc}	22.28^{abcd}	30.72^{abc}	40.91^{a}
Myc/5PM	5.49^{bcd}	14.44^{ab}	27.89^{a}	33.66^{ab}	42.00^{a}
SSP/5PM	5.81^{abcd}	11.48^{bcd}	24.66^{abcd}	30.28^{ab}	26.89^{b}
Brad/Myc	4.36^{d}	10.84^{dc}	20.67^{bcd}	22.07^{bcd}	37.75^{a}
Brad/SSP	5.50^{bcd}	14.11^{abc}	20.19^{cd}	25.96^{d}	36.10^{a}
Myc	7.50^{a}	16.45^{a}	27.46^{a}	34.63^{a}	41.91^{a}
SSP	4.98^{cd}	13.85^{abc}	26.44^{ab}	31.52^{abc}	37.75^{a}

Table 9. Influence of poultry manure, P source and *Bradyrhizobium* on plant phosphorus concentration (%), phosphorus uptake (mg/kg)

Treatments	4 WAP	6 WAP	8 WAP	10 WAP	12 WAP	14 WAP
Poultry manure source						
0 (ton)	8.10^{a}	9.29^{a}	44.51^{a}	43.10^{a}	8.86^{a}	17.15^{a}
5 (ton)	7.93^{a}	9.22^{a}	37.33^{a}	37.42^{a}	9.39^{a}	17.56^{a}
10 (ton)	8.14^{a}	9.36^{a}	37.20^{a}	36.01^{a}	29.98^{a}	29.66^{a}
P value	0.95	0.98	0.57	0.67	0.30	0.64
Phosphorus source						
Mycorrhiza	7.97^{a}	9.29^{a}	41.32^{a}	40.10^{a}	21.70^{a}	28.10^{a}
SSP	8.11^{a}	9.28^{a}	38.05^{a}	37.62^{a}	10.46^{a}	14.47^{a}
P value	0.79	0.98	0.61	0.72	0.38	0.27
***Bradyrhizobium* source**						
Bradyrhizobium+	8.23^{a}	9.48^{a}	39.64^{a}	39.30^{a}	11.84^{a}	25.43^{a}
Bradyrhizobium-	7.85^{a}	9.08^{a}	39.72^{a}	38.38^{a}	20.32^{a}	17.41^{a}
P value	0.48	0.45	0.26	0.94	0.50	0.51

Also, on P uptake by plant at 6 and 8 WAP the application of poultry manure had no significant effect at 0, 5, and 10 ton/ha on phosphorus uptake in plants at 6 and 8 WAP but poultry manure with 0 kg/ton shows higher values than other treatments, P – Source showed no significant difference, but higher values were observed in plots treated with mycorrhiza. There were no significant differences on the phosphorus uptake between plants that has *Bradyrhizobium* and those without. However, on P- use efficiency at 6 and 8 WAP, there was no significant difference in the treatments, but higher values were observed in 10 tons of poultry manure 6 and 8 WAP, mycorrhiza supply phosphorus more effectively than SSP 6 and 8 WAP. Similarly, the plot where *Bradyrhizobium* was not applied had higher value at 6 WAP whereas at 8 WAP plots with *Bradyrhizobium* have higher values. Therefore, 10 tons of poultry manure, mycorrhiza and *Bradyrhizobium* helps in phosphorus use efficiency on soybean.

3.10 Interaction of Treatments on Plant Phosphorus Concentration, P-uptake

Table 10 showed the interaction between poultry manure, P source and *Bradyrhizobium* on plant phosphorus uptake at 6 and 8 WAP. Statistically, the interaction of these treatments shows no significant differences. However, plots where *Bradyrhizobium*, mycorrhiza and 5 ton/ha of poultry manure were applied had higher values at 6 WAP. Similarly, at 8 WAP plots that received SSP and 10 ton/ha of poultry manure had higher values.

3.11 Influence of Treatments on Yield Parameter

Table 11 showed the interaction of Poultry Manure, P source and *Bradyrhizobium* on Yield Parameter. The application of poultry manure showed no significant differences at 0, 5 and 10 ton/ha, but higher values were observed on the plots that received 10 ton/ha of poultry manure. However, the application of P source showed significant differences ($p < 0.05$) with mycorrhiza having the higher value. *Bradyrhizobium* showed no significant difference, higher values were observed on the plots without *Bradyrhizobium*.

3.12 Interactive Effects of Treatments on Yield Parameter

Table 12 showed the interaction between poultry manure, P-source and *Bradyrhizobium* on yield parameter. The application of the treatments showed significant differences where the plots that received *Bradyrhizobium,* Single Super Phosphate and 5 ton/ha of poultry manure had the higher values.

Table 10. The effect of interaction between poultry manure, P source and *Bradyrhizobium* on plant phosphorus uptake at 6 and 8 WAP

Treatments	P- uptake	
	6 WAP	8 WAP
Myc/10PM	54.67[a]	32.29[a]
Brady/Myc/10PM	24.67[a]	37.70[a]
SSP	41.94[a]	46.50[a]
Brady/SSP/10PM	33.36[a]	33.13[a]
Brady/SSP	53.77[a]	28.93[a]
Brady/Myc/5PM	55.55[a]	39.03[a]
Myc/5PM	30.67[a]	34.42[a]
Brady/SSP/5PM	40.74[a]	45.43[a]
SSP/10PM	36.11[a]	46.56[a]
SSP/5PM	22.36[a]	25.15[a]
Myc	52.58[a]	45.38[a]
Brady/Myc	29.75[a]	51.58[a]
P value	0.510	0.694

Table 11. Influence of Poultry Manure, P source and *Bradyrhizobium* on Yield Parameter

Treatments	Yield
Poultry manure source	
0 (ton)	100.1[a]
5 (ton)	103.5[a]
10 (ton)	106.8[a]
P value	0.01
Phosphorus source	
Myc	109.8[a]
SSP	97.1[b]
P value	0.01
***Bradyrhizobium* source**	
Bradyrhizobium -	106.1[a]
Bradyrhizobium +	100.1[a]
P value	0.01

Table 12. Interactive effect of the treatments on yield (g)

Treatments	Yield
Brad/SSP/5PM	50.89[a]
Myc/5PM	39.58[ab]
Brad/Myc/5PM	31.74[ab]
SSP/5PM	27.73[ab]
Brad/SSP/10PM	27.09[ab]
Myc/10PM	20.74[ab]
Brad/Myc/10PM	18.41[ab]
SSP/5PM	12.28[b]

3.13 Analysis on Weight of One Thousand Seeds and 100 Seed Weight

Table 13 showed significant differences on the seed weight of one thousand seeds and 100 seed weight. However, plots that received mycorrhiza showed higher values at one thousand seed weight and in 100 seeds weight.

3.14 Influence of the Treatments on Soil Phosphorus and the Percentage of Mycorrhiza Colonization

Table 14 showed the main influence of poultry manure, P source and *Bradyrhizobium* on soil phosphorus at 14 WAP. Statistically, the treatments had no significant differences over other on soil phosphorus but 5 kg/ton of poultry manure, SSP, and the plot with *Bradyrhizobium* had higher values that other treatments. In addition, the influence of the treatments on percentage of mycorrhiza colonization at 6 WAP shows no significant differences on the percentage of colonization, but higher values were shown in plots treated with 5 ton/ha of poultry manure, mycorrhiza and *Bradyrhizobium*. Inoculation of soybean with *B. japonicum* significantly increased nodulation, yield and seed quality.

3.15 Interaction of Treatments on Soil Phosphorus and the Percentage of Mycorrhiza Colonization

Table 15 showed the interaction between poultry manure, P source and *Bradyrhizobium* on the soil phosphorus at 14 WAP, the application of these treatments shows no significant differences, but plots where *Bradyrhizobium*,

Table 13. One thousand seeds weight and 100 seed weight (kg/plot)

Treatments	One thousand seeds weight (kg/plot)	100 seeds weight (kg/plot)
Myc/10PM	90.97[b]	106.3[a]
Brady/Myc	85.93[a]	89.73[cde]
Brady/Myc/5PM	84.83[ab]	93.57[c]
Myc/5PM	83.5[abc]	102.17[ab]
Brady/SSP/10PM	81.37[abc]	91[cde]
SSP	80.6[abc]	82.6[e]
SSP/5PM	78.57[abc]	84.37[de]
Myc	78.43[abc]	104.77[a]
Brady/SSP/5PM	76.23[abc]	92.5[cd]
Brady/SSP	73.1[abc]	83.63[de]
SSP/10PM	64.57[bc]	90.5[cde]
Brady/Myc/10PM	63.5[c]	94.8[bc]

Table 14. Influence of poultry manure, P source and *Bradyrhizobium* on soil phosphorus at 14 WAP and % mycorrhiza colonization at 6 WAP

Treatments	Soil P (mg kg^{-1}) 14 WAP	Mycorrhiza colonization (%) 6 WAP
Poultry manure source		
0 (ton)	10.68[a]	34.33[ab]
5 (ton)	10.82[a]	38.33[a]
10 (ton)	10.74[a]	37.00[a]
P value	0.99	0.74
Phosphorus source		
Mycorrhiza	10.66[a]	38.89[a]
SSP	10.84[a]	34.22[ab]
P value	0.82	0.27
***Bradyrhizobium* source**		
Bradyrhizobium-	10.59[a]	34.89[ab]
Bradyrhizobium+	10.91[a]	38.22[a]
P value	0.99	0.62

Table 15. The interaction between poultry manure, P source and *Bradyrhizobium* on soil phosphorus at 14 WAP and % mycorrhiza colonization at 6 WAP

Treatments	Soil phosphorus (mg kg^{-1})	Mycorrhiza colonization (%)
	14 WAP	**6 WAP**
Myc/10PM	9.91^a	44.33^a
Brady/Myc/10PM	11.31^a	45.33^a
SSP	11.07^a	37.33^a
Brady/SSP/10PM	10.61^a	30.67^a
Brady/SSP	10.30^a	26.27^a
Brady/Myc/5PM	11.21^a	29.33^a
Myc/5PM	10.18^a	41.33^a
Brady/SSP/5	10.64^a	44.00^a
SSP/10PM	11.13^a	28.00^a
SSP/5PM	11.26^a	38.67^a
Myc	9.96^a	40.00^a
Brady/Myc	11.41^a	33.33^a
P value	0.82	0.73

mycorrhiza and also plots with *Bradyrhizobium*, mycorrhiza and 10 tons of poultry manure had higher values respectively. In addition, on the percentage of mycorrhiza colonization on soybean there were no significant differences statistically on the treatments, however higher values were showed on the plots, which had *Bradyrhizobium*, mycorrhiza and 10 ton of poultry manure.

4. DISCUSSION

It was seen that the effect of mycorrhiza had higher values over other phosphorus source (SSP) on growth, yield and phosphorus use efficiency, so also is the inoculation of poultry manure at 5 – 10 ton/ha. Mycorrhiza, 5 – 10 ton/ha of poultry manure, and *Bradyrhizobium* are very good biofertilizer for the production of soybean. This positive effect of inoculation by *B. japonicum* was accompanied with increase in seed yield of soybean [17-20,12,21].

According to [22], nutrient uptake by plants depends largely in the amount, concentration and activities in the rhizosphere as well as the capacity of the soil to replenish the nutrients in the soil solution. Soil microorganisms such as *B. japonicum* inoculants and other plant growth promoting rhizobacteria are reported to influence the chemistry of soils nutrients in many ways and enhancing nutrients uptake by plants [23]. In the last few years, the numbers of *Rhizobium* inoculants have been developed and primarily used for enhancing N_2-fixation by legume plants [24]. For example *Bradyrhizobium* are reported to establish symbiosis relationship with legume where they fix nitrogen that is important for plant growth and in turn, the plant provide them with carbohydrates as their source of energy [23]. On top of atmospheric nitrogen fixation, rhizobia inoculation have been reported to improve plant nutrient such as P by mobilizing inorganic and organic P from organic and inorganic sources in the soil rhizosphere [23,25,22] in their study conducted at green house and field experiments also reported that *Rhizobium* inoculation significantly increased the uptake of P, K, Ca, and Mg in the plant parts attributed to increased soil pH.

Similarly, [26] reported that *Rhizobium* inoculation significantly increased hundred seeds weight of soybean by 91%. Likewise, [27] also indicated that inoculation of soybean seeds increased hundred seeds weight. This could be due to significant contribution of N_2 fixation which supplied extra N for the crop as it is a major constituent of amino acids and many biological compounds that play major roles in photosynthesis which eventually increased seeds weight.

5. CONCLUSION

- Inoculation of mycorrhiza and 5 ton/ha of poultry manure should be applied for optimum growth of soybean.
- Mycorrhiza, *Bradyrhizobium* and 10 ton/ha of poultry manure is recommended for increase in plant phosphorus concentration in soybean.

- Mycorrhiza inoculation should be applied for maximum P – uptake by soybean.
- Single super phosphate, *Bradyrhizobium* and 5 ton/ha of poultry manure is recommended for soil phosphorus uptake.
- Inoculation of 10 ton/ha poultry manure and mycorrhiza should be applied for increase in grain yield of soybean.
- *Bradyrhizobium*, mycorrhiza and 10 ton/ha of poultry manure is recommended for optimum P-use efficiency on soybean.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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http://sciencedomain.org/review-history/19356