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# Influence of P sources and rhizobium inoculation on growth and yield of soybean genotypes on Ferric Lixisols of Northern Guinea savanna zone of Ghana

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#### ABSTRACT

Soybean is becoming an important cash crop in northern Ghana. Yet the yields are low due to use of low yielding varieties and limited use of inputs. Greenhouse and field experiments were carried out to evaluate the effects of two phosphorus (P) sources and Rhizobium inoculation on growth, nodulation, P uptake, and yield of three soybean genotypes on Ferric Lixisols of the Guinea savanna zone of Ghana. The P sources were triple superphosphate (TSP) and Morocco phosphate rock (MPR), while the genotypes were TGx 1448-2E, TGx 1904-6F, and TGx 1955-4F. The greenhouse experiment was conducted at the University of Ghana, Legon in a completely randomized design. The field experiment which was carried out in the Upper East region of Ghana was laid out in a split-split plot design with four replicates. In both the greenhouse and field experiments, application of TSP at 30 kg P ha<sup>-1</sup> resulted in significantly higher growth and P uptake in shoot compared with MPR and control. Soybean genotypes showed significant differences in growth, nutrient uptake, and grain yield in both the greenhouse and the field experiments. Rhizobium inoculation increased nodule number and dry weight but did not increase grain yield. The genotype TGx 1955-4F appears to show greater potential for increasing productivity of soybean in low P soils in northern Ghana.

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#### **KEYWORDS**

Grain legumes; nitrogen fixation; nodulation; phosphorus fertilizer; soil fertility

# Introduction

Although global population is expected to increase by about 29% by 2050, the rise will be much greater in sub-Saharan Africa (SSA) than in other regions (United Nations, Department of Economic and Social Affairs, Population Division 2015) and this is expected to be accompanied by demand for food (FAO 2014). In areas of high population, as in the case of northern Guinea savanna zone of Ghana, the potential of expansion of agricultural land is limited making sustainable intensification a necessity (Cook et al. 2015; FAO 2014; Pretty, Toulmin, and Williams 2011; Vanlauwe et al. 2014). Integration of grain legumes, particularly soybean in the predominantly cereal-based farming systems in the northern Guinea Savanna zone of Ghana in the context of integrated soil fertility management (ISFM), thus offers a potential pathway for sustainable intensification. Soybean could be rotated with cereals with the additional benefit of reducing the need for mineral nitrogen (N) fertilizer to the subsequent cereal crop grown in rotation (Giller et al. 2011; Sanginga 2003) in the context of ISFM (Vanlauwe et al. 2015).

Soybean is increasingly becoming an important cash crop in northern Ghana. It is mostly cultivated in the three northern regions and the northern part of Volta region of the country. About 80% of national production estimated at 182,000 tons occurs in the three northern Regions of

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Ghana (USAID (United States Agency for International Development) 2014). The crop is grown mainly for food and cash income but in the northern Guinea savanna zone of Ghana, the haulms after harvest are excellent sources of quality feed for livestock. Furthermore, in the smallholder farming systems in northern Ghana, soybean contributes to soil fertility improvement as an additional benefit. Its contribution to soil fertility improvements is due to its N fixing abilities as part of the fixed N in the roots in addition to those that remain in the haulms are returned to the soil after harvest. Soybean can fix atmospheric N in the soil with estimated amounts varying from 44 kg ha<sup>-1</sup> to as high as 300 kg N ha<sup>-1</sup> (Giller et al. 2011; Sanginga, Dashiell, and Diels 2003). Cereal crops that are intercropped with or grown after soybean benefit from this N-fixing property of soybean. Potential rotational benefits of soybean on yields of cereals grown in rotation include breaking of pest and disease cycles (Francis and Clegg 1990), soil structure improvement (Peoples and Craswel 1992), enhanced phosphorus (P) availability through secretion of enzymes and acids in the legume rhizosphere (Schlecht et al. 2006), and enhanced arbuscular mycorrhizal colonization (Harinikumar and Bagyaraj 1988).

Despite its importance, the yield of soybean is far below its potential. According to SARI (2000), the average on-farm yield of soybean in northern Ghana is estimated at 1.0 t  $ha^{-1}$  which is far below the potential yield of 3 t ha<sup>-1</sup> (Alliance for a Green Revolution in Africa 2016). The low yield is attributed to several factors including the use of low-yielding varieties, poor soil fertility and particularly low P levels, high costs, or limited availability of inputs such as certified seeds, P fertilizers, and rhizobium inoculants (ACET 2013). Grain legume yields, and/or the amount of N fixed, will depend on legume genotype  $(G_L)$ , the effectiveness of rhizobium strain(s) nodulating the legume  $(G_R)$ , the biophysical environment (E), and agronomic management (M) including fertilizer application. The effect of these factors and their interactions have been expressed in literature as the relation  $(G_L \times G_R) \times E \times M$  (Giller et al. 2013). While studies have shown that rhizobium inoculation can contribute significantly to grain yield of soybeans (Ahiabor et al. 2014; Ronner et al. 2016) and genotype and P fertilizers have positive effects on the productivity of soybean (Nwoke et al. 2005), no study so far has tried to evaluate the potential contribution that improvement in the above factors can make to increasing soybean yields in smallholder systems. Furthermore, response of soybean to these inputs has been highly variable (Ronner et al. 2016; Thilakarathna and Raizada 2017; van Heerwaarden et al. 2018).

To date, little information is available on P-efficient soybean genotypes in the SSA region, even though using P-efficient soybean genotypes is a sustainable P management strategy for enhancing yield and P use efficiency (Zhou et al. 2016). To correct P deficiency and increase its status in soils of the three regions within the Guinea savanna zone, P fertilizers such as superphosphates have been recommended. However, these P soluble fertilizers are either not readily available or are expensive and therefore not attractive to the local farmers. This has caused the farmers in the northern Ghana to apply little or no mineral P fertilizers at all to their crops, leading to poor crop establishment and low yields. Meanwhile, there are indigenous phosphate rocks which are found in large deposits in Africa that can serve as a cheaper alternative P fertilizer to improve soil fertility, yield index, and enhance biological nitrogen fixation of soybean (Mokwunye and Bationo 2011).

This study was therefore carried out to evaluate the effects of P sources and rhizobium inoculation on growth and yield of three soybean genotypes on Ferric Lixisols in the Northern Guinea savanna zone of Ghana.

# Materials and methods

The experiment was carried out in the field and in the greenhouse using the same soil. The field experiment was conducted at Tilli, a village in the Bawku West District in the Northern Guinea Savanna zone of Ghana, during the 2015 farming season, while the greenhouse study was conducted at the school of Agriculture, University of Ghana, Legon, Ghana.

#### Field experiment

The field site was located within Latitude 10°53'78 N and Longitude 000°33' W with elevation of 193 m above sea level. The area receives an annual rainfall ranging between 800 and 1,100 mm, considered enough for a single farming season. The rainy season starts from May to September with peak in August/September and a long dry season from October to April. The total amount of rainfall during 2015, the year during which the field trial was carried out, was 830.3 mm with 45 rainy days while the total amount of rains during the experimental period was 583.8 mm with 31 rainy days. Temperatures are usually high, averaging 34°C. The maximum temperature could rise as high as 42° C and the minimum as low as 16°C. The low temperatures are experienced from December to late February. The soil at the experimental site is Ferric Lixisol (Asiamah and Adjei-Gyapong 2001), a representative of the study area.

# Soil sampling and analysis

Soil sampling was carried out at the start of the experiment at 0–30 cm depth using an auger for initial soil characterization. A composite was made from 20 samples collected randomly from different parts of the plot and thoroughly mixed, sub-sampled, air-dried, crushed, and sieved through a 2 mm sieve to remove twigs, plant roots, and ironstone. The sieved soil samples were brought to the laboratory for physicochemical analyses using standard protocols. The soil bulk density was determined using the core method by Blake (1965). Particle size analysis was carried out by the hydrometer method described by Bouyoucos (1962). The soil pH was determined according to the electrometric method in both distilled water and in 0.01M CaCl<sub>2</sub> solution at ratio of 1:1 and 1:2 (CaCl<sub>2</sub>). Organic carbon was analyzed by Walkley–Black procedure, N by Kjeldahl method, and available P by Bray-1 method (Bremner and Mulvaney1982).

#### Field preparation and experimental layout

The experimental plot was previously cultivated to maize. The field was ploughed with tractor and harrowed with hoe after which the field was laid out. The experiment was conducted as a split-splitplot design. The main plots were made up of three soybean genotypes while sub-plots were the two P sources, triple superphosphate (TSP) (46%  $P_2O_5$ ), Morocco phosphate rock (MPR), (30%  $P_2O_5$ ), and control (no fertilizer). The sub-sub-plots were with or without rhizobium inoculation. There were four replications giving a total of 72 experimental units (3 sources of P fertilizers applied at 30 kg P ha<sup>-1</sup> and 2 rates of inoculation (6 treatments) × 3 genotypes × 4 Reps). The soybean genotypes used were TGx 1904-6F (104–114 days maturity period) and TGx 1955-4F (105–110 days maturity period) developed by IITA (International Institute of Tropical Agriculture (IITA), Ibadan, Nigeria and TGx 1448-2E (TGx 1448-2E) (115–117 days maturity period) the local genotype developed by IITA and the Savanna Agricultural Research Institute (SARI) of the Council for Scientific and Industrial Research (CSIR), Ghana and usually cultivated by the farmers in northern Ghana. Some selected morphological and growth characteristics of the genotypes used are shown in Table 1.

The sub-plot size was 5 m  $\times$  5 m with an alley of 2 m between main plots and 1 m between sub-plots. The sub-plot consisted of 10 rows each 5 m long with 50 cm spacing between the rows and 10 cm spacing within a row at 3 seeds per stand which was later thinned to 2 seedlings per stand 3 weeks after planting

Table 1. Characteristics of soybean genotypes used in the experiments.

Breeder's code	Maturity time (days)	Maturity group	Potential yield (t ha <sup>-1</sup> )
TGx 1448-2E	115–117	Late	2.4–2.5
TGx 1904-6F	104–114	Medium	2.5–2.7
TGx 1955-4F	105–110	Medium	1.4–2.6

Source: Ronner et al. (2016)

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(WAP). The phosphorus fertilizers were applied at planting in furrows 10 cm away from the planting line. No nitrogen fertilizer was applied. However, the soybean genotypes were either inoculated with or without rhizobium inoculant (Nodumax). The inoculant contained  $10^{10}$  cells g<sup>-1</sup> of *Bradyrhizobium japonicum* strain USDA 110 manufactured by the IITA, Ibadan, Nigeria.

The soybean seeds were moistened with Gum Arabica solution in a basin and the inoculant was added at the rate of 7 g per kg of seeds. The mixture was stirred thoroughly and uniformly with wooden spatula until even coating was attained. The seeds were then spread on a sack under a shade and allowed to air dry for 30 min to enable the inoculant to stick well enough onto the surface of the seeds before planting. The treated seeds were sown early in the morning to avoid its exposure to direct sun rays that might affect the quality of the inoculant. With these treatments, the uninoculated seeds were sown before the inoculated ones to avoid contamination. Weed control was done twice manually using hoe. All cultural practices recommended for growing soybean were applied equally to all the plots. The crops were grown for a maximum of 110 days.

# Greenhouse experiment

The experimental design for the greenhouse experiment was completely randomized design using the same soil type used for the field experiment. The same treatment combinations used in the field experiment was also used in the greenhouse experiment. These included the three soybean genotypes, the rhizobium inoculation and the two P sources, TSP (46% P<sub>2</sub>O<sub>5</sub>), and MPR  $(30\% P_2O_5)$  at the application rate of 30 kg P ha<sup>-1</sup> which is equivalent to 251 mg MPR and 164 mg TSP 2.5 kg<sup>-1</sup> soil and a control. The experiment was replicated four times, giving a total of (18 treatments  $\times$  4 Reps) 72 pots. Equal amount (2.5 kg) of the soil was weighed carefully into plastic pots and the P fertilizers, TSP and MRP, were added at the rate of 258 and 168 mg  $2.5 \text{ kg}^{-1}$  soil, respectively. To ensure uniform application of the P sources to the pots, the soil in each pot was transferred into a large plastic basin and the weighed amount of the P source was added, thoroughly mixed, and returned to the respective pots. Three seeds of each soybean genotype were sown in each pot and distilled water added to bring the soils to 60% field capacity. To minimize uneven environmental effects within the greenhouse, the pots were rotated weekly. The plants were thinned to two per pot 3 days after germination to ensure adequacy of nutrients other than P. The plants were sprayed with Aceta Star 46EC insecticide against white flies and leaf miners 2 weeks after germination. The plants were grown for 110 days. The plants were immediately weighed after cutting them at the soil surface for fresh weight. The plants were dried in an oven at 70°C for 48 h to a constant weight. The dried matter was then ground to pass through a 1 mm sieve and later stored in small plastic bags for subsequent P and N analyses. P was determined by molybdenum blue calorimetric method and N by micro-Kjeldahl method (IITA 1981).

# Data collection

# Growth and nodulation assessment

At 10 weeks after sowing, 10 plants were randomly selected from each plot for nodulation and growth measurements (plant height assessment); the plant height was measured from the ground level to the apex of the plant using graduated measuring pole both in the greenhouse and in the field and the average was calculated for each plot/pot. In the field experiment, the plants were carefully uprooted and cut at the soil level; the root zone was gently washed on a 2 mm mesh sieve under tap water. The nodules removed were weighed freshly, counted, and oven-dried at 65°C for 48 h to determine their dry weights. Data on nodulation were not taken for the greenhouse experiment due to limited replication of pots. However, all other data taken for the field study were also taken for the greenhouse study.

#### Yield and yield components

Harvesting was done at physiological maturity. Physiological maturity was considered to have taken place when 95% of the plants had turned golden yellow (Tukamuhabawa et al. 2002) and 75% of the plants had their pods filled with seeds and hardened (Masumba 1984). At harvest, the plants within an area of 4 m  $\times$  3 m = 12 m<sup>2</sup> (6 rows of 4 m length) were harvested from the 25 m<sup>2</sup> plot leaving 50 cm from the outer ends of the plots. The plants were cut using a sharp knife at about 5 cm above the soil level.

Ten plants were randomly picked after harvest from the harvested plants to determine their podding capacities. All the pods were counted and the average number of pods per plot was calculated. The harvested plants from each plot were bulked and weighed with a digital scale after which the pods were removed shelled, and both fresh and oven dry weights were recorded. The fresh weight of the seeds was subtracted from initial weight (whole plant with pods) to obtain the haulm dry weight per plot. The oven dry weight was used to estimate the grain yield and harvest index. Grain yield per hectare was determined by threshing the harvested plants from the harvested area (4 m  $\times$  3 m) central 12 m<sup>2</sup> of each plot. The threshed seeds were dried in the sun for 3 days and weighed. The resulting weights, in kg per harvest area, were then extrapolated to kg  $ha^{-1}$  basis to get the average grain yield per hectare. For the greenhouse experiment, at harvest all the whole plants with pods per pot were weighed after which the pods from the plants were detached, counted shelled, and weighed. The fresh weight of the seeds was subtracted from initial weight (whole plant with pods) to obtain the actual weight of shoot biomass/pot. The shoot biomass for both experiments was then oven-dried at 70°C for 48 h to a constant weight and their weights were also recorded after which they were grounded into powder for plant analysis. The N was determined using micro-Kjeldahl method and P by molybdenum blue calorimetric method (IITA 1981).

#### Data analysis

Data collected from the two experiments were subjected to analysis of variance (ANOVA) using GENSTAT software version 9. When ANOVA indicated statistical significance of a treatment effect (p < 0.05), the means were separated using Duncan Multiple Range Test.

#### Results

# General characteristics of the soil from the study site

The soil analysis also showed that the soil was moderately acidic with pH values of 5.55 (water 1:1) and 5.05 (Cacl<sub>2</sub>, 1:2). The organic carbon content and total N of the soil which were 2.7 and 0.5 g kg<sup>-1</sup>, respectively, were low. Phosphorus availability as determined by Bray-1 was 1.12 mg P kg<sup>-1</sup> was very low while that of total P soil was not high either (18.28 mg P kg<sup>-1</sup>) (Table 2).

the study site.	
Properties	Mean values
Sand (g kg <sup>-1</sup> )	700
Silt (g kg <sup><math>-1</math></sup> )	150
Clay (g kg <sup><math>-1</math></sup> )	150
Bulk density (Mg m <sup>3–1</sup> )	1.70
pH (Water) (1:1)	5.55
pH (0.01M (CaCl <sub>2</sub> ) (1:2)	5.05
Total N (g kg $^{-1}$ )	0.5
O C (g kg <sup>-1</sup> )	2.7
Total P (mg kg <sup><math>-1</math></sup> )	18.28
Available P (mg kg <sup><math>-1</math></sup> )	1.12
CEC (cmol(+) kg <sup>-1</sup> )	4.03

Table 2. Physicochemical	properties	of	the	soil	at
the study site.					

# The effect of genotype, P source, and rhizobium inoculation on growth, yield, and yield components of soybean

# Greenhouse experiment

The soybean genotypes TGx 1904-6F and TGx 1955-4F produced significantly taller plants than TGx 1448-2E. TGx 1448-2E however had significantly larger shoot dry matter and pod number at harvest than the other two genotypes. The air-dried pod weight and oven-dried grain yield were significantly greater in TGx 1904-6F than TGx 1955-4F and TGx 1448-2E (Table 3). Harvest Index (HI) varied (p < 0.05) among the genotypes with the genotype TGx 1955-4F having the highest value.

Plant height, pod weight, and HI were significantly affected by rhizobium inoculation with the inoculated plants having significantly taller plants but smaller pod weight and HI than the un-inoculated plants. There was no significant difference between the inoculated and un-inoculated treatments with respect to shoot dry matter, pod number, and grain yield (Table 3).

P application significantly (p < 0.05) influenced plant height, shoot dry matter, and HI with plants that received TSP producing significantly taller plants and greater shoot dry matter but lower HI (Table 3). P application however did not influence grain yield and yield components.

#### Field experiment

The results of the effect of genotype, P source, and rhizobium inoculation on growth, nodulation, and yield from the field experiment showed that plant height, nodule dry weight, grain yield, and HI significantly (p < 0.05) varied among the soybean genotypes (Table 4). Plant height was significantly greatest for TGx 1955-4F and least for TGx 1448-2E (Table 4). As was found in the greenhouse experiment, HI was significantly greatest in TGx 1955-4F and least for TGx 1448-2E. The genotype TGx 1955-4F produced the highest grain yield and TGx 1448-2E the lowest yield (Table 4).

Rhizobium inoculation positively influenced nodulation, podding, biomass yield, and HI. Inoculated plants produced greater number of nodules and nodule dry weight as well as greater number of pods and higher HI but lower haulm yield (Table 4). Inoculation did not significantly influence pod weight and grain yield.

P application significantly influenced plant height, pod number, pod weight, grain yield, haulm yield, and HI (Table 4). Although application of TSP resulted in greater biomass yield, greater number of pods, and large pod weight, these did not translate into grain yield. The grain yield of the control plot was significantly higher than that of the TSP-treated plots. The interaction between rhizobium inoculation and P application significantly affected plant height, grain yield, and biomass yield.

	Plant height (cm pot <sup>-1</sup> )	Dry matter (g	Pod No	Pod wt. (g	Grain yield (g	Harvest
Treatment	10 WAP	pot <sup>-1</sup> )	(pot <sup>-1</sup> )	pot <sup>-1</sup> )	$pot^{-1}$ )	index
Genotype						
TGx 1448-2E	41 ± 6.14b	4.77 ± 0.68a	12 ± 1.06a	$3.28 \pm 0.35a$	2.31 ± 0.32a	0.29 ± 0.34b
TGx 1904-6F	44 ± 5.97a	4.35 ± 0.84b	10 ± 2.21b	2.88 ± 0.59b	2.01 ± 0.46b	0.28 ± 0.06b
TGx 1955-4F	42 ± 3.70ab	4.10 ± 0.87b	10 ± 1.56b	$3.19 \pm 0.78a$	2.35 ± 0.40a	0.33 ± 0.06a
Rhizobium inocula	tion					
Un-inoculation	41 ± 4.83b	4.33 ± 0.95a	11 ± 1.55a	$3.27 \pm 0.41a$	2.30 ± 0.39a	0.32 ± 0.06a
Inoculation	43 ± 5.93a	4.48 ± 0.71a	10 ± 2.10a	2.97 ± 0.55b	2.14 ± 0.44a	0.29 ± 0.05b
P sources						
Control	38 ± 5.39c	3.79 ± 0.71c	11 ± 2.30a	$3.07 \pm 0.45a$	2.09 ± 0.37a	0.32 ± 0.05a
MPR	42 ± 3.10b	4.29 ± 0.47b	10 ± 1.40a	3.14 ± 0.44a	2.30 ± 0.37a	0.31 ± 0.04a
TSP	46 ± 4.48a	5.15 ± 0.67a	10 ± 1.79a	3.15 ± 0.62a	2.27 ± 0.50a	0.27 ± 0.05b

Table 3. Plant height, yield, and yield components as affected by genotype, P sources, and rhizobium inoculation in the greenhouse experiment.

TSP: Triple superphosphate; MPR: Morocco phosphate rock.

For each factor, figures with the same letters within a column are not statistically different.

			Nodule dry					
Treatment	Dlant hairdht (cm) at 10 MAD	Nodula No. 10 nlante <sup>-1</sup>	weight (mg) 10	Pod No. 10 nlante <sup>-1</sup>	Pod wt. (ba ba <sup>-1</sup> )	Grain yield	Haulm weight (الم المع	Haniact indev
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uenotype TC 111025								
1GX 1448-2E	$69 \pm 13.05$	$24 \pm 10.32a$	$0.18 \pm 0.10b$	483 ± 128a	1,160 ± 295.40a	$694 \pm 1450$	$2,408 \pm 539.3a$	$0.23 \pm 0.04b$
TGx 1904-6F	75 ± 16.26ab	27 ± 9.52a	0.20 ± 0.11ab	393 ± 98a	1,095 ± 303.00a	754 ± 249 b	2,406 ± 749.7a	$0.24 \pm 0.05ab$
TGx 1955-4F	82 ± 10.14a	31 ± 9.58a	0.23 ± 0.09a	397 ± 137a	1,160 ± 393.50a	1,034 ± 262a	2,574 ± 673.7a C	0.29 ± 0.04a
Rhizobium inoculation	ntion							
Un-inoculation	76 ± 17.10a	$22 \pm 6.00b$	$0.17 \pm 0.09b$	420 ± 127a	1,080 ± 289.7a	852 ± 250.5a	2,685 ± 632.4a	$0.24 \pm 0.06b$
Inoculation	74 ± 10.87a	33 ± 10.78a	0.23 ± 0.11a	429 ± 129a	1,197 ± 361.1a	803 ± 284a	$2,240 \pm 608.1b$ 0.26 $\pm$ 0.05a	0.26 ± 0.05a
P Sources								
Control	$71 \pm 12.30b$	27 ± 10.29a	0.21 ± 0.08a	$385 \pm 88b$	$1,075 \pm 246.4b$	933 ± 261a	2,558 ± 430.8a	0.27 ± 0.05a
MRP	69 ± 13.48b	27 ± 8.71a	0.18 ± 0.14a	$417 \pm 140b$	$1,075 \pm 313.5b$	739 ± 239b	$2,050 \pm 580.7b$ 0.27 $\pm$ 0.05a	0.27 ± 0.05a
TSP	85 ± 11.49a	29 ± 11.47a	0.22 ± 0.07a	471 ± 138a	1,265 ± 390.8a	$810 \pm 274b$	2,781 ± 713.9a	$0.23 \pm 0.05b$

P sources in the field experiment.	
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# Effect of genotype, source of P, and rhizobium inoculation on plant nutrient uptake

# Greenhouse experiment

Soybean genotype influenced N and P uptake both in the shoot and in the grain. Uptake of N and P in the shoot was significantly higher in TGx 1448-2E than in TGx 1904-6F and TGx 1955-4F, while in the grain, uptake of N and P was highest in TGx 1955-4F.

P uptake in the shoot was significantly higher with inoculated treatment than the un-inoculated treatment. However, N uptake in both shoot and grain and P uptake in the grain were not significantly affected by rhizobium inoculation (Table 5).

Nutrient uptake in both the shoot and the grain varied significantly among the P sources with TSP-treated plants recording the greatest N and P uptake in the shoot while the control plants had the smallest uptake (Table 5). In the grain while the control plants had the smallest nutrient uptake there were not significant differences between the MPR and the TSP-treated plants.

#### Field experiment

Although the uptake of N and P in the shoot was not statistically different among the genotypes, TGx 1955-4F showed tendency for a higher uptake (Table 6). However, in the grain TGx 1955-4F had significantly higher N and P uptake than TGx 1448-2E and TGx 1904-6F.

Rhizobium inoculation had negative effect on N and P uptake in both grain and shoot (Table 6). Uptake of N and P in the shoot and grain was higher in the un-inoculated plots than in the inoculated plots.

Shoot uptake of N was significantly influenced by source of P with the MRP treatment having significantly higher N uptake while the TSP-treated plants had the least N uptake (Table 6). P uptake

	Shoot uptak	e (mg pot <sup>-1</sup> )	Grain uptake	(mg pot <sup>-1</sup> )
Treatment	N	Р	Ν	Р
Genotype				
TGx 1448-2E	90 ± 13.07a	33 ± 8.51a	77 ± 12.38a	14 ± 2.84a
TGx 1904-6F	82 ± 14.91b	30 ± 9.78b	68 ± 17.13b	12 ± 3.88b
TGx 1955-4F	78 ± 16.26b	29 ± 9.76b	80 ± 16.00a	15 ± 3.59a
Rhizobium Inoculation				
Un-inoculation	81 ± 17.66a	29 ± 10.93b	77 ± 16.42a	14 ± 4.05a
Inoculation	85 ± 12.84a	32 ± 7.56a	74 ± 15.41a	13 ± 3.00a
P Sources				
Control	72 ± 13.77c	20 ± 5.60c	65 ± 11.81b	10 ± 2.02b
TSP	96 ± 9.33a	40 ± 3.47a	80 ± 12.95a	15 ± 2.39a
MRP	82 ± 11.89b	32 ± 5.13b	81 ± 17.48a	15 ± 3.34a

Table 5. N and P uptake as affected by genotype, P sources, and rhizobium inoculation in the greenhouse experiment.

For each factor, figures with the same letters within a column are not statistically different.

	Table 6. Effect of genotype,	rhizobium inoculation, and P	sources on N and P	uptake in the field experiment.
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	Shoot upta	Shoot uptake (kg ha <sup>-1</sup> )		e (kg ha <sup>-1</sup> )
Treatment	Ν	Р	Ν	Р
Genotype				
TGx 1448-2E	70.51 ± 15.46a	36.00 ± 12.57a	27.23 ± 5.69b	4.17 ± 0.83b
TGx 1904-6F	69.66 ± 21.43a	36.35 ± 16.63a	29.54 ± 9.53b	4.60 ± 1.42b
TGx 1955-4F	74.23 ± 18.56a	39.20 ± 13.66a	41.01 ± 10.72a	6.42 ± 1.71a
Rhizobium Inoculation				
Un-inoculation	75.07 ± 18.26a	41.24 ± 17.56a	32.86 ± 10.04a	5.23 ± 1.69a
Inoculation	67.86 ± 18.25b	33.13 ± 8.38b	32.33 ± 11.41a	4.90 ± 1.66b
P Sources				
Control	74.30 ± 13.94a	29.58 ± 6.37b	34.47 ± 10.82a	4.87 ± 1.46b
TSP	59.79 ± 15.77b	49.93 ± 15.61a	30.51 ± 9.10a	4.77 ± 1.61b
MRP	80.31 ± 19.48a	32.05 ± 9.27b	32.79 ± 11.25a	5.56 ± 1.86a

For each factor, figures with the same letters within a column are not statistically different.

in shoot was also significantly higher in the MPR-treated plots and lowest in the control plots. Uptake of N in the grain was not significantly influenced by source of P, while P uptake was significantly higher in the MRP-treated plots.

# Discussion

## Characteristics of soil used in the study

The soil used in this experiment was moderately acidic with pH of 5.55 in water. This may be due to the nature of inherent parent material. Soybean does well in soils of pH between 4.5 and 8 (Dugje et al. 2009) and hence soil acidity will not constrain production since the soil pH was within pH range adequate for soybean production. The available P level of the soil used for this experiment was below the critical value of 10 mg kg<sup>-1</sup> needed for crop growth in SSA (Fairhurst 2012). The low level of available phosphorus of this soil may be because of the nature of parent materials that formed the soil and the presence of iron pans (concretions) in these soils in the savanna zones of Ghana (Abekoe 1989).

# Effects of genotype, inoculation and P source on growth, nodulation, and yield of soybean

The soybean genotypes showed high variability in growth, nodulation, and yield and yield components indicating variability in the genotypes studied. The significant differences in plant height of the soybean genotypes in both the greenhouse and the field experiments may primarily be due to differences in their growth habit and their ability to adapt to the soil, and other environmental conditions. Differences among genotypes in plant height among genotypes have been reported by El Naim and Jaberereldar (2010). In a similar study, Talaka, Rajab, and Mustapha (2013) also reported significant difference in growth of five different soybean varieties grown under rain-fed condition at 6 WAP and no significant difference at 8 WAP.

Although the results of the field experiment did not show significant differences among the three genotypes with respect to the number of nodules, there were significant differences among the genotypes with respect to nodule dry weight; the genotype TGx 1955-4F produced the greatest nodule dry weight while the genotype TGx 1448-2E produced the smallest nodule dry weight (Table 4). The differences in the nodule dry weight among the different genotypes could be attributed to differences in their ability to tolerate adverse environmental condition, particularly temperature and soil pH. According to Lie (1974) and Lee and Lee (1998), the environment has a profound impact on nodulation that can be confirmed by comparing the nodule number and weight of the genotype. According to Serraj and Adu-Gyamfi (2004), acid tolerant host genotypes and inoculants strains have been used as strategies for reducing the negative effects of environmental stress on nodulation and nitrogen fixation in legumes. The result showed that the genotype TGx 1955-4F which had the highest nodule number also had the highest nodule dry weight, while TGx 1448-2E which had the lowest nodule number had the least nodule dry weight confirming earlier report by Oti and Agbim (2000) that suggests a simple relationship between nodule number and nodule dry weight both of which are indices of nitrogen fixation. The high nodule number and greater nodule dry weight obtained in TGx 1955-4F suggests its potential to fix more atmospheric nitrogen in this soil than the other genotypes. Effective nodulation has been suggested to be crucial for a functioning legume-rhizobium symbiosis and so plants most susceptible to infection and capable of producing high effective nodules have the utmost capacities to fix more atmospheric nitrogen (Kellman 2008).

Genotypic differences affected pod numbers and pod weight. The number of pods per plant which is also the main component of yield in the end determines the potential productivity of soybean. TGx 1448-2E, the most widely cultivated genotype in northern Ghana, produced the highest pod number in both greenhouse and field experiments. This was, however, statistically significant only in the greenhouse experiment. This agrees with the assertion of Graham (1992) that selection of genotypes is one of the essential factors for increasing pod yield in soybean. The

non-significant difference in pod number and pod weight among the genotypes in the field experiment may be due to the reduction in soil moisture during flowering and pod filling because of late planting. Masoumi et al. (2011) and Behtari and Abadiyyan (2009) reported that soybean yield could significantly be reduced by soil moisture during flowering and pod filling periods.

Significant differences in grain yield was observed among the soybean genotypes with TGx 1955-4F producing the greatest yield both in the greenhouse (Table 3) and in the field (Table 4) experiments. The significant differences in the grain yield of these soybean genotypes in this study may be due to differences in their growth habit which is related to genetic potentials of the different genotypes. While TGx 1955-4F and TGx 1904-6F are both medium maturing genotypes with maturity periods ranging between 104 and 110 days, TGx 1448-2E is a late maturing genotype with maturity period of between 115 and 117 days (Table 1). It is possible that TGx 1904-6F and TGx 1955-4F flowered and podded earlier than the TGx 1448-2E and therefore slightly escaped the drought which occurred during the pod filling period. Other workers (Alam et al. 2009; Malik et al. 2007; Rahman et al. 2011) have reported of significant yield differences among soybean genotypes. The grain yields reported in this study are however lower than what have been reported in recent times in similar environments in northern Ghana (Adjei-Nsiah, Alabi, and Kanampiu 2018) and northern Nigeria (Ronner et al. 2016). This might be attributed to drought that set in during pod filling as explained earlier.

The results from the greenhouse experiment reveal that soybean genotype TGx 1448-2E produced the highest biomass yield (4.77 g/pot) and this may be due to its longer growth period than the genotypes TGx 1955-4F and TGx 1904-6F which had caused it to produce higher biomass under controlled environment (greenhouse). In the field experiment, biomass production did not differ much, although TGx 1955-4F appears to have produced the largest biomass (2,573.96 kg ha<sup>-1</sup>). Study has shown that soybean straw (biomass) yield is dependent on the variety and the environment (Martinov 2008).

The results show a positive response of soybean to the application of TSP at 30 kg P ha<sup>-1</sup> which significantly influenced the plant height. In both the field and greenhouse experiments, plants grown with TSP at 30 kg P ha<sup>-1</sup> were taller than plants grown with MPR at 30 kg P ha<sup>-1</sup> and the control treatment. This indicated that TSP was readily available for plants' uptake and hence reflected in early plant growth and development as observed in both the field and the greenhouse experiment. The increase in plant growth in response to phosphorus supply has been reported in several studies (Kwari 2005; Rani 1999; Rebafka, Ndunguru, and Marchner 2003; Tomar, Singh, and Singh 2004).

Although the effect of P application on nodulation was not statistically different between the different P sources, TSP had the tendency to produce greater number of nodules and greater nodule dry weight than MRP and the control. Similar results have been reported by Jemo et al. (2010). Studies by Tagoe, Horiuchi, and Matsui (2008) and Waluyo, Lie, and Mannetje (2004) have shown that phosphorus helps to initiate nodule formation as well as the development and functioning of formed nodules. According to Miller and Roy (1982), the weight of nodule is a factor contributing to N<sub>2</sub> fixation activity while nodule number is important in relation to nodule weight. It may be possible that the quantity of P applied (30 kg P ha<sup>-1</sup>) in this experiment was not adequate to allow sufficient nodulation.

Phosphorus application influenced shoot biomass at harvest both in the field and in the greenhouse with the plots receiving TSP having the largest biomass. Several studies (Asia, Safder, and Shahzad 2005; Lamptey et al. 2014; Roughley et al. 1993) have reported of similar increases in shoot biomass yield of soybean with the application of TSP fertilizer. The poor influence of MPR on the growth and yield parameters of the soybean in both the field and the greenhouse experiments may be due to its low solubility compared with TSP. Pod number and pod weight were also positively influenced by the application of phosphorus with TSP having significantly greater number of pods and greater pods weight than the MRP and the control in the field experiment. Similar results have been reported by Rani (1999).

In the field experiment, grain yield was statistically greater in the control plots than in the plots treated with TSP or MRP contrary to what has been reported in the literature (Lamptey et al. 2014; Malik et al. 2006; Ronner et al. 2016; Sabir et al. 2001), although TSP-treated plots tended to produce

larger pod number, pod weight, and shoot biomass yield. It has been reported that soybean has high requirement for phosphorus, and seed and yield quality can be improved by phosphorus fertilizer in soils with low phosphorus levels (Shahid et al. 2009) as those found in northern Ghana. In our present study, the failure of the TSP-treated plots to translate the higher growth exhibited during the vegetative phase as well as the larger pod number and pod weight into grain yield was probably due to the drought that occurred during the pod filling period.

Rhizobium inoculation positively improved nodule number and nodule dry weight in the field experiment although this did not have any significant effects on plant height, shoot biomass, grain yield and its components suggesting that increased nitrogen fixation may not necessarily result in higher growth and yields. The effect of inoculation on nodulation has been reported in other studies (Elkoca, Turan, and Donmez 2010; Kumaga and Etu-Bonde 2002). In the present study, the larger number of nodules and the greater nodule dry weight associated with the inoculated treatment suggest that the introduced rhizobium strain was able to outcompete the native rhizobia in the soil (Okereke et al. 2000). The failure of inoculation to significantly influence shoot biomass at harvest is, however, contrary to the findings of Alam et al. (2015) who reported increase in biological yield (biomass) of soybean with rhizobium-inoculated seeds. Although inoculation did not significantly influence plant height in the field experiment, it significantly affected plant height in the greenhouse experiment confirming earlier observation made by Hernandez and Cuevas (2003) that *Rhizobium* inoculation has positive effect on plant height.

Pod number and pod weight were not, however, significantly affected by rhizobium inoculation while inoculation seemed to have negatively affected grain yield significantly in spite of the large number of nodules as well as the greater nodule dry weight observed with the inoculated treatment. The failure of the higher nodulation in the inoculated plots to be translated into yield could probably be due to the extreme drought condition experienced during the pod filling period which occurred at the terminal part of the season. Schultz and Thelen (2008) reported of similar negative yield response from inoculation under extreme drought condition during pod filling stage.

Results of both field and greenhouse experiments showed significant interaction effect between P sources and rhizobium inoculation on growth and yield of soybean. In similar studies, Khan, Mehmood, and Aslam (2000) and Amos, Ogendo, and Joshua (2001) reported that P application with rhizobium inoculation significantly increased pod number, grain yield, and dry matter yield when compared with un-inoculated treatments. Gunarto (2000) attributed increased plant growth with inoculation and P application to improved soil productivity because of the increased supply of P for enhanced N fixation by the bacteria.

# Effect of genotype, source of P, and rhizobium inoculation on plant nutrient uptake

Uptake of N and P in both shoot and grain was significantly influenced by phosphorus alone and its interaction with rhizobium ( $P \times I$ ) in the field experiment. This may be due to the supply of P needed for rhizobium to fix more N for plant use resulting in the enhanced growth, yield, and N uptake by roots to the shoot. This shows the importance of P in soybean production and supports the conclusions of earlier workers (Kamara et al. 2007; Lamptey et al. 2014; Okugun, Otuyem, and Sanginga 2004) that P has significant effect on growth, nodulation, and yield of soybean. The accumulation of P in the soybean resulted in corresponding improvement in growth and shoot biomass yield (Tables 5 and 6) although these effects did not translate into grain yield due possibly to drought during the pod filling period. In both the field and the green house experiments, uptake of N and P in the grain was highest in the genotype TGx 1955-4F. The uptake of nutrients in the grain of this genotype reflected significantly in the growth and yield parameter suggesting that this genotype may be more efficient in utilizing P which could be attributed to its root morphology although this was not studied in the present study. According to Zhao, Fu, and Liao (2004), plants with shallow root architecture had better spatial configuration in the higher P cultivated soil surface layer and hence had higher P uptake efficiency and yield. Also, soybean genotypes are known to

differ in their ability to grow under low P conditions and the more efficient genotypes may exhibit internal or external mechanisms that allow greater P uptake and grain yield (Kirk et al. 1998). Our finding thus agrees with the reports of Solomon, Pant, and Angaw (2012) that varietal differences exist in soybean with respect to P uptake. The high performance of TGx 1995-4F in terms of growth, nodulation, and yield showed its high affinity for P in this soil.

The greenhouse experiment showed that Rhizobium inoculation had significant effects on N and P uptake in shoot. According to Biswas, Ladha, and Dazzo (2000), rhizobium inoculation may induce an increase in number of root hairs and thereby favors nutrient uptake by exploring a greater soil volume.

The combination of P application and rhizobium inoculation had positive effect on the uptake of N and P in both experiments. Rodelas et al. (1999) made similar observation when they applied Rhizobium inoculant and P fertilizer to faba beans and attributed the increased uptake of the nutrients to enhanced root development which resulted in increased mineral uptake

The results indicate that soybean genotype influenced N and P uptake in grain. The highest uptake of both N and P was found in TGx 1955-4F. This may be because this genotype was able to take up nutrient efficiently and transported it from other plant parts to the seed at the start of seed filling as confirmed by Greenwood et al. (1991). Genetic factors of the soybean genotypes may have contributed to the differences in their accumulation of N and P in their grains. Rhizobium inoculation did not statistically influence total N and P in grain although it enhanced N and P uptake in shoot. The inability of the rhizobium inoculation to significantly influence N and P uptake in grain of the soybean could be due to the drought that occurred during the grain filling period which consequently affected grain yield.

Addition of P tended to increase total N and P uptake both in the shoot and in the grain of the soybean plant with TSP-treated plants showing the highest uptake. This was, however, only statistically significant in the greenhouse experiment. However, P concentration in both the shoot and the grain was statistically (p < 0.05) highest in the TSP-treated plants and lowest in the control treatment both in the green house and in the field experiments (data not shown). The limited effect of P application on P uptake in the grain in the field experiment was due to lower grain dry matter production due to the drought that occurred during the grain filling period which affected pod filling and grain yield.

# Conclusion

The soybean genotypes used in our study showed high variability in growth, grain yield, and nutrient uptake in grain. The higher grain yield and higher P uptake in grain of the genotype TGx 1955-4F in comparison with the current genotype (TGx 1448-2E) being cultivated widely by farmers suggest that TGx 1955-4F has the potential to increase the productivity of soybean in P-deficient soils as those found in the northern Guinea Savanna zone of Ghana. However, since this study was conducted for only one season and because the terminal phase of the trial coincided with a severe drought, further studies are needed to determine if the yield of this genotype is stable enough to form the basis of recommendation of this genotype to farmers.

TSP appears to be a better source of P than MRP as it promoted better plant growth and greater pod number than the MRP and the control treatment, although these did not translate into higher grain yield due to the drought that occurred during the pod filling period. Although TSP has the potential to increase soybean productivity in P-deficient soils as those found in the northern Guinea Savanna zone of Ghana, the availability of TSP in rural areas remains a challenge. The importation and distribution of TSP in the areas where soybean is produced therefore require urgent attention to ensure its sustainable supply.

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