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GROWTH AND YIELD RESPONSE OF SCREENED SOYBEAN (*Glycine max* L. MERRILL) GENOTYPES TO FERTILIZATION AND RHIZOBIA INOCULANTION IN NORTHERN GHANA

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Abstract

Soybean remains an important crop for the sustenance of livelihoods of resource-constrained farmers in northern Ghana. However, yield of the crop has continuously remained low due to poor soil fertility and low productivity. A two-phase experiment was carried out during the 2019 cropping season to evaluate the growth and yield response of screened soybean genotypes to nitrogen, phosphorus and rhizobia inoculation. In the first phase, growth and yield of 100 selected genotypes were evaluated under optimum phosphorus fertilization using the lattice design. In the second phase, 3 best performing genotypes (N19, N119, N135), selected in phase one together with a known variety (Jenguma) were accessed for growth and yield under phosphorus, nitrogen and inoculants using a 4 (genotype) x 7 (nutrient regime) split plot design. The nutrient regimes were sole triple super phosphate (TSP), sole inoculant, sole booster nitrogen, TSP + booster nitrogen, TSP + inoculant, TSP + booster nitrogen + inoculant and a control (no fertilizer). Each treatment was replicated three times. Data collected were subjected to analysis of variance and means separated at 5% probability using the least significant difference. Results show a significant ($P = 0.032$) interaction effect between genotype and nutrient regime on grain yield. Jenguma, treated with TSP + inoculant recorded the highest yield of 4 t/ha, followed by Jenguma variety treated with TSP + inoculant + booster nitrogen (3.9 t/ha), and genotype (N135) treated with TSP + nitrogen (3.7 t/ha), while genotype (N19) without treatment (control) recorded the least grain yield. The high yield obtained for inclusion of P, N and inoculants exceeded what is documented for northern Ghana (1.5 t/ha). Therefore, it is recommended for farmers to include P, booster N and inoculants in cultivation of the Jenguma variety, and N135 genotype. Based on economic cost analyses, farmers stand to achieve higher profit upon application of sole TSP.

Keywords: Soybean, Genotype, Phosphorus, Fertilization, Inoculation and Screened

Introduction

Soybean (*Glycine max* L. Merrill) is an annual leguminous crop that is cultivated in the tropical, subtropical and template regions. It is known that soybean is a fundamental source of high quality and cheap protein (around 40%) and contains about 20% of very edible and cholesterol-free oil. Ghana produces around 15,000 metric tons of soybean grain every year (MoFA and CSIR, 2005). The crop is cultivated mostly in the northern savannah and parts of Ashanti and Brong-Ahafo Regions. Among these Regions, Northern Region happens to be the main producer of soybeans with 70% of national soybean area, and around 77% of national soybean production (SRID, 2012). Inspite of the numerous advantages of soybean, the grain yield per unit area is still low in Ghana. MoFA (2011) revealed that farmers' average soybean yields of 1.5 t/ha is well below attainable yields of 2.3 t/ha. A number of research studies have ascribed the low soybean yields in sub-Saharan Africa to poor yielding varieties. High-yielding soybean plants require a lot of N and it is estimated that biological nitrogen fixation (BNF) can contribute 60 to 70% of the N needed by the plant (Herridge *et al.,* 2008; Salvagiotti *et al.*, 2008) through nodulation by rhizobium. Although BNF is the major source of nitrogen input in agricultural systems, the full potential of the symbiotic system may not be realized because of inefficient strains of rhizobia. The need for small amounts of N fertilizer supplied at the early stage of the crop often promote growth and N_2 fixation in legumes (Tahir *et al.,* 2009). Phosphorus unavailability is one of the most important nutrient elements that limit plant growth (Fernandez *et al.*, 2007). Phosphorus plays a significant role in nodule formation and fixation of atmospheric nitrogen, hence nodulating legumes requires higher P compared to non-nodulating crops. A few variables including: declining soil fertility, utilization of poor yielding varieties, inadequate utilization of rhizobium inoculant, poor utilization of mineral fertilizer and absence of or low use of booster nitrogen influence the grain yield of soybean in northern Ghana (Ahiabor *et al.,* 2014). High yielding varieties in combination with rhizobia inoculant and fertilization are essential to increase soybean yield (Alam *et al.,* 2015). Salvagiotti *et al.* (2008) reported the potential to improve soil fertility and soybean yields per unit area if the farmer use high yielding variety in addition to adoption of appropriate rhizobium inoculant, booster nitrogen and phosphorus fertilization. Although, numerous studies have been conducted on one or a combination of two and/or three of these yield enhancing factors, data remains relatively unavailable. Lack of knowledge on soybean performance in combination with these factors limits soybean productivity by resource-poor smallholder farmers. There is need therefore to investigate the combined effect of enhanced genotype, booster nitrogen, phosphorus fertilization and inoculation on productivity of soybean.

The aim in this study was to evaluate soybean genotypes for improved yield under booster nitrogen fertilization, phosphorus application and rhizobium inoculation and to determine the most cost effective technology to be used in soybean production to maximize profit for the resource-poor farmers.

Materials and Methods

Site description

The research was carried out in Nyankpala from July to October 2019. Nyankpala is located about 16 km west of Tamale and lies on latitudes N 09°24′ 15.9″ and longitude W 01°00′ 12.1″ of the interior Guinea Savanna agro-ecological zone of Ghana. Rainfall across the region is unimodal and starts from April – May; builds up slowly to a height in August-September before declining sharply in October-November. The total precipitation is about 1,100 mm per annum, with a range of 800 mm to about 1,500 mm. Average ambient temperatures are high year round (about 28°C), but the harmattan months of December and January are characterized by minimum temperatures that may decrease to 13°C at night. Geologically, the area consists of proterozoic rocks which differ in lithology and degree of metamorphism. Granite and metamorphic rocks are the main rock types and include: biotiteschists, biotite-hornblande gneisses, garnet-hornblende and garnet-biotite gneisses and schists. Others include: Albite-chlorite, sericite-quartz schists with inter-bedded acid tuffs, manganiferous phyllites and sandstones. The soil consist of laterites and are mostly Savannah Ochrosols and Luvisols/Lixisols (World Reference Base for Soil Resource; Freddy *et al*., 1998).

Land preparation

Land was ploughed and harrowed at the commencement of the rains. Lining and pegging was done according to crop spacing of 60 cm x 10 cm for soybean. Plot size was 4 m by 3.5 m. The spacing between blocks was 2 m and within blocks 1 m.

Experimental design and treatments

The study was in two stages: Selection of high-yield genotypes from a pool of available germplasm, followed by evaluation of inoculants and mineral fertilization effect on the selected high-yield genotypes.

a. Selection of high yielding genotypes from germplasm pool

Hundred soybean genotypes, selected from soybean germplasm pool of Savannah Agriculture Research Institute (SARI) were screened in 2019 under irrigation. The experiment was laid out in latice design, planted at a spacing of 60 cm by 10 cm, and plot sizes of 3 m by 4 m were used. Seeds were sown at a rate of four seeds per hill, and later thinned to two plants per stand. Triple super phosphate (TSP) was applied at 60 kg/ha two weeks after planting. Weeds were controlled by hoeing at 6 and 10 week after panting (WAP). Growth of genotypes was observed and yield recorded at harvest. The best three high-yield genotypes (3% genotypes) were selected and used to assess the impact of inoculants and mineral fertilization on their productivity.

b. Effect of fertilization and inoculation on high-yield soybean genotypes

The experiment was laid out in a split plot design and replicated three times. The three selected-soybean genotypes and a released variety served as sub-plot factor, while mineral fertilizer and inoculant served as main plot factor. Based on result of the genotype-screening experiment, the three soybean genotypes selected and used were N19, N135, and N119. The Jenguma variety was used as a check in the study. The planting distance was 60 cm by 10 cm, with 2 seeds per stand. The size of each main plot was 4 m by 3 m. The four cultivars (N19, N135, N119 and Jenguma variety) were planted in line on each main plot at 1 m interval.

c. Treatment details

Fertilization regime was used as the main factor at seven levels [Sole soybean (S), triple superphosphate (TSP), nitrogen (N), inoculant (I), triple superphosphate + inoculant (T+I), triple superphosphate + nitrogen (TSP+N), and triple superphosphate + inoculant + nitrogen (TSP+I+N)], and genotypes as a sub-factor at four levels (N19, N135, N119 and Jenguma). Detailed fertilization combination is presented in Table 1. The seed of the soybean genotypes used were obtained from the SARI germplasm collection of the Council for Scientific and Industrial Research (CSIR) at Nyankpala, Northern Region.

Table 1: Detailed treatment description of main plot (fertilization regime)

Detailed treatment combination	Treatment code
TSP = 60 kg/ha, Booster Nitrogen = 20 kg/ha and 5 g of legume fix inoculant per 1 kg of seed	
No fertilization	Control
Triple superphosphate	TSP
Inoculant	I
Nitrogen	N
Triple superphosphate + Nitrogen	TSP+N
Triple superphosphate + Inoculant	TSP+I
Triple superphosphate + Inoculant + Nitrogen	TSP+I+N

Seed inoculation
Inoculation of seeds was done using the slurry method described by Woomer et al. (1994). Inoculation was done with water as adherent. The seeds and inoculums were placed in a bowl and mixed thoroughly until seeds were covered with black film of inoculants. The treated seeds were dried for a few minutes after which they were planted. The inoculation was done at the rate of 5 g of legume fix inoculant per 1 kg of seed.

Cultural practices

a. Thinning
Thinning was done to ensure 10 cm between plants in a row, 20 days after sowing, when the soil was moist and seedlings well established.

b. Weeding
Weeding was done manually by hand using hoe, on the sixth and tenth week after sowing to control weeds. Each weeding activity was done the same day for all every blocks.

Nodule count and effectiveness
Six sampled plants from each plot were taken 35 days after sowing to determine nodulation. The samples were carefully dug out to retrieve nodules and each nodule was plucked out from the root. The nodules were kept in labeled polythene bags and sent to the laboratory and washed, counted and the fresh weight taken. The nodules were cut opened using a knife and hand lens to determine apparent nodule effectiveness. Nodules with pink or reddish color were considered effective in fixing nitrogen, while those with green, colorless or no color were considered to be ineffective (Gwata *et al*., 2003).

Biomass dry weight
The biomass dry weight was taken at 10WAP. Six sampling plants from every plot were placed in envelopes which were labeled and dried in the oven to steady weight at 84°C for 24 hours, and afterward weighed and recorded in grams.

Pods weight per plant
For pods number, six plants were taken from each plot, and every one of the pod detached and placed in envelopes which were labeled and dried in the oven to stable weight at 84°C for 24 hours, and afterward weighed and recorded in grams.

Seed weight
The 100 seed weight was recorded by counting 100 seeds from the lot per plant and oven drying at 82°C for 24 hours. These were weighed to determine the mean seed weight in grams.

Grain yield (tons per hectare)
Grain yield per hectare was estimated by threshing the harvested plants from the middle, one square meter of each plot. These were placed in named envelopes and oven dried to a steady weight at 60°C for 48 hours, and after that the weight were recorded. The resulting weights, in grams (g) per meter square were then scaled up to kilogram per hectare basis to get the average grain yield per hectare

Economic cost analysis
From the various technologies, the economic cost benefit was determined by quantifying the various technologies in terms of monetary values and comparing the cost to determine which has the highest benefit in terms of cost ratio (Das *et al.,* 2010).

Data analysis
Data were subjected to analysis of variance (ANOVA) model using GenStat statistical package edition 12. Means were separated using Least Significant difference (LSD) at 5% probability level. Results are presented in graphs and tables.

Results and Discussion

Grain yield response to genotypes
The grain yield was significantly ($P < 0.001$) affected by soybean genotypes. The grain yield ranged from 33.7 kg/ha to 1238 kg/ha (Figure 1a and 1b). Genotype N119 had the highest grain yield of 1238 kg/ha which was not significantly different from genotype N19 (1223.8 kg/ha), which was statistically different from N135 (1056.9 kg/ha). Genotype N210 recorded the least grain yield of 33.7 kg/ha which was statistically different from the grain yield of other genotypes.

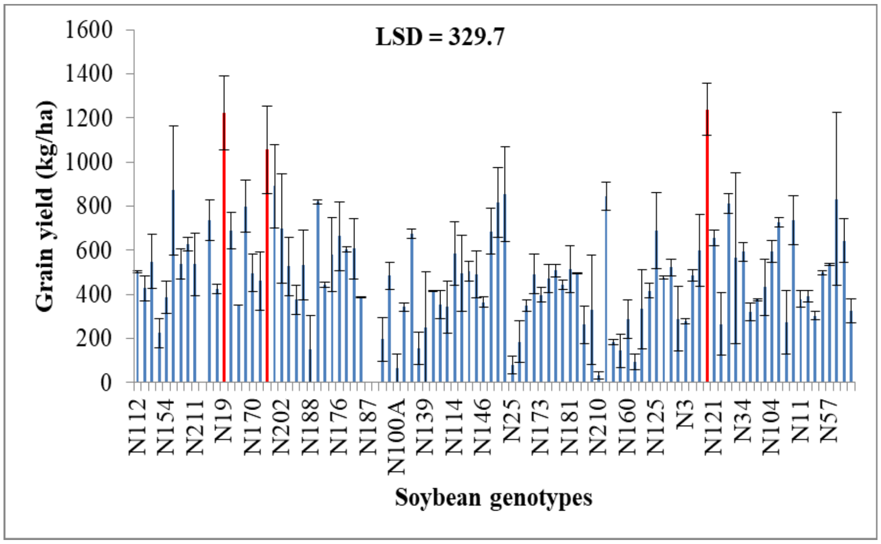

Figure 1a: Effect of soybean genotypes on grain yield. Error bars represent standard error of means

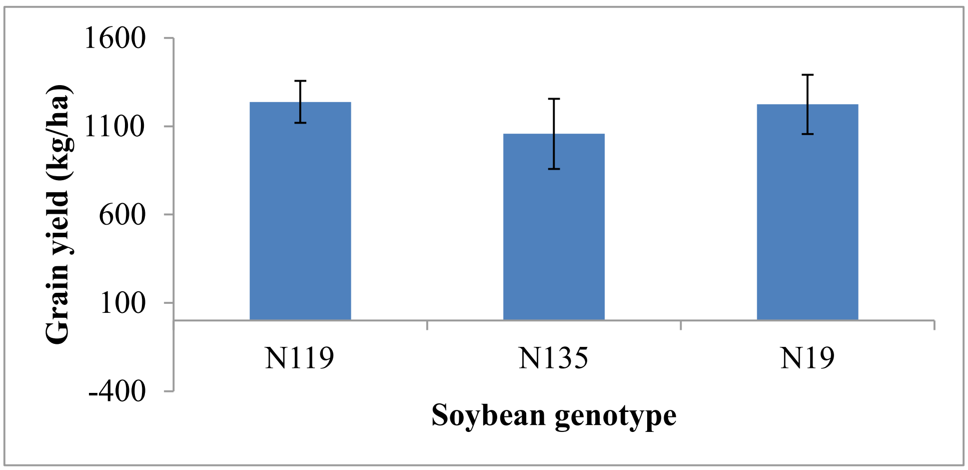

Figure 1b: Three best performing genotypes under optimum phosphorus fertilization. Error bars represent standard error of means (SEM)

Fertilization and genotype effect on growth and yield
Number of nodules per plant

There was no significant interaction (P=0.182) between fertilization regime and soybean genotype. However, fertilization and inoculant had a significant (P=0.003) effect on number of nodules in which Tsp + inoculants produced the highest number of nodules per plant (108) which is not significantly different from sole inoculants (96), followed by the control (no fertilizer) and sole Tsp, while sole nitrogen and Tsp + N recorded statistically similar number of nodules (37 and 40 respectively) (Figure 2).

Though genotype had no significant (P=0.226) effect on number of nodules per plant, N119 recorded the higher number of nodules (87.0) compared to Jenguma (83.0), which was significantly similar to N135 (74), while genotype N19 recorded the least number of nodules (63) per plant.

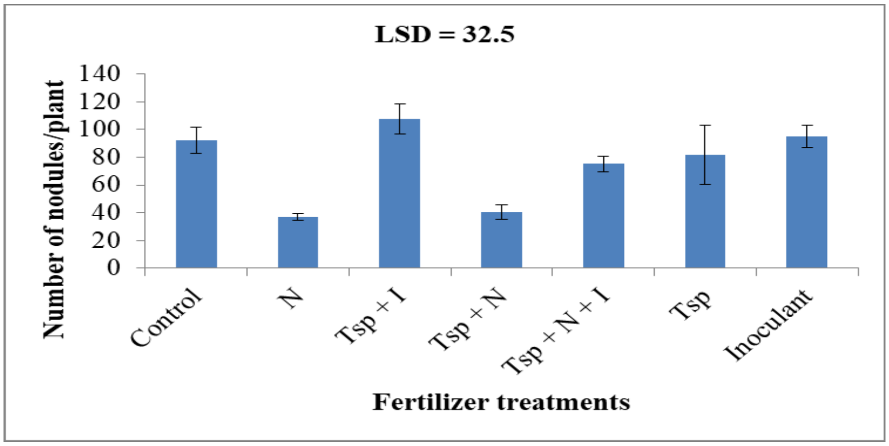

Figure 2: Effect of fertilizer regime on number of nodules per plant. Error bars represent standard error of means

Effective nodules percentage

There was no significant interaction (P = 0.141) between fertilizer regime and soybean genotype on effective nodule percentage. Fertilization had a significant (P = 0.045) effect on effective nodules percentage (Figure 3). Triple superphosphate + inoculant (Tsp +I) recorded the greatest effective nodule percentage (81.2%) which is not significantly different from TSP (72.5%) followed by I (68.5%) while N recorded the least percentage of effective nodules (31.5%).

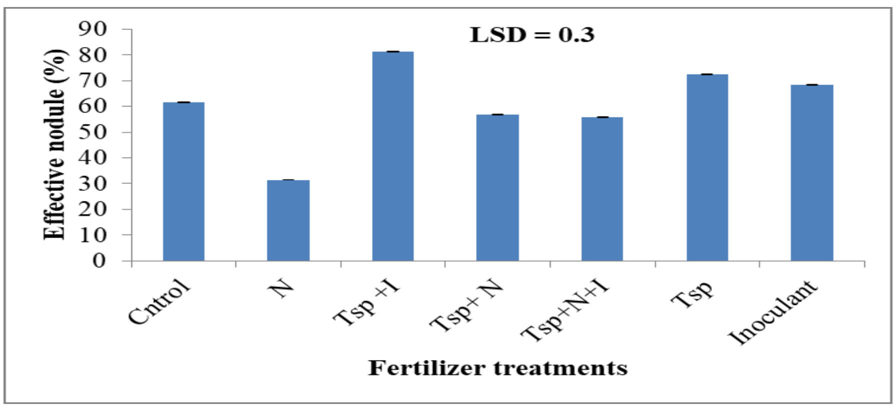

Figure 3: Effect of fertilizer regime on effective nodules percentage of soybean. Error bars represent standard error of means

Biomass dry weight

There was no interaction (P > 0.05) effect between fertilizer regime and Soybean genotypes on biomass dry weight at 6, 8 and 10WAP. Fertilizer regime significantly (P<0.001) influenced biomass dry weight at 10WAP (Figure 4). Tsp + nitrogen + inoculant recorded the highest biomass dry weight (18 g/plant), which is significantly similar to Tsp + inoculant (15 g/plant) followed by Tsp, with sole N recording the least biomass dry weight (8 g/plant). Soybean genotype had no significant (P=0.827) influence on biomass dry weight at 10WAP but N119 recorded the greatest biomass dry weight (13 g/plant) followed by N135 and Jenguma recording the least biomass dry weight (12 g/plant).

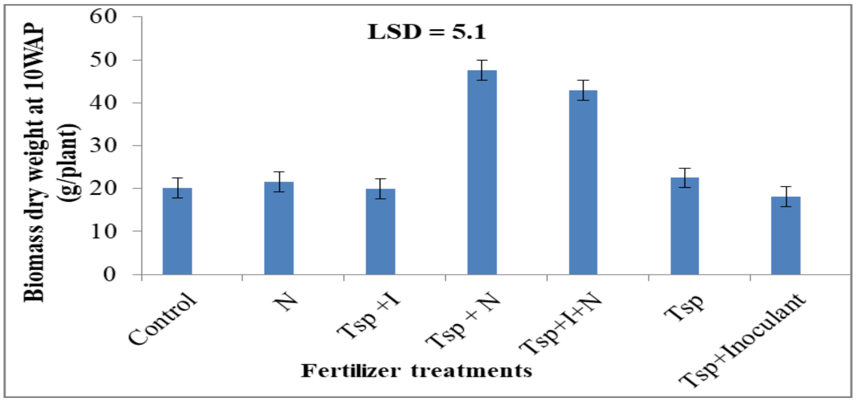

Figure 4: Effect of fertilizer regime on biomass dry weight at 10 weeks after planting. Error bars represent standard error of means

Pod weight per plant

There was no interaction (P = 0.517) effect between fertilizer regime and genotype on weight of pods. Pod weight was not significantly (P = 0.0683) affected by soybean genotypes. However, Jenguma recorded the highest pod weight (36.2 g) followed by N135 (35.1 g), while N119 and N19 recorded same pod weight (32.8 g) (Figure 5). Fertilization had significant (P=0.042) influence on pod weight with Tsp + inoculant recording the greatest pod weight (43.2 g), which is significantly similar to Tsp (42.9 g) and Tsp+ nitrogen + inoculant (40.7 g), whereas the control recorded the least (23.7 g).

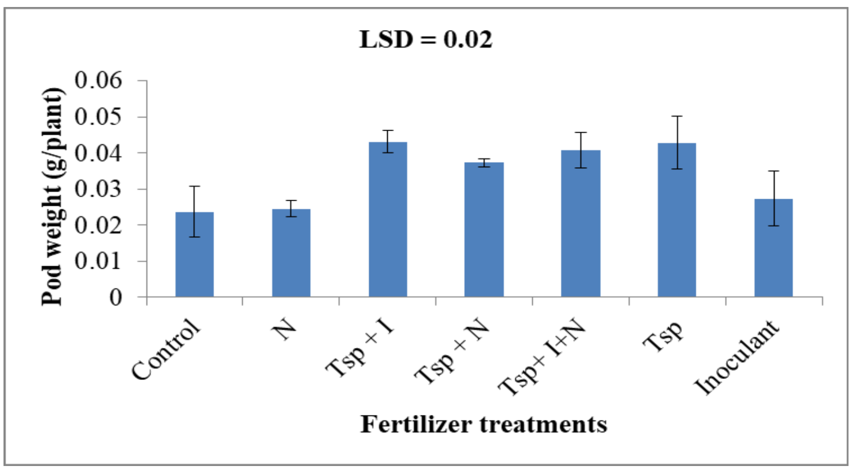

Figure 5: Effect of fertilizer regime on pod weight per plant in g. Error bars represent standard error of means

Grain weight

There was no interaction (P = 0.920) effect between fertilization and genotype on 100 grain weight (Figure 6). Hundred grain weight significantly (P < 0.001) differed by soybean genotype. Genoype N135 recorded the highest 100 seed weight (13.9 g) which did not statistically differ from Jenguma (13.7 g) and N119, while genotype N19 had the least 100 seed weight (8.8 g). There was no significant difference (P=0.405) among the fertilizer treatments. However, Tsp + inoculant gave the highest (13.9 g), followed by Tsp, and the control had the least 100 seed weight (11.0 g).

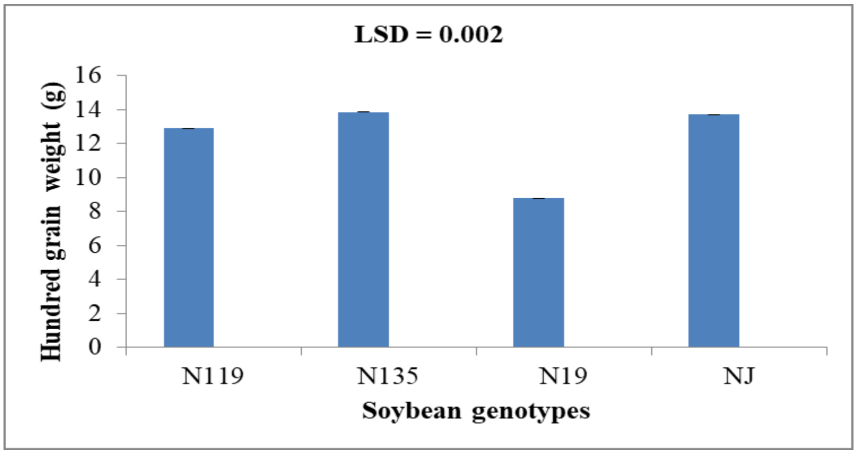

Figure 6: Effect of high-yield soybean genotypes on 100 grain weight. Error bars represent standard error of means (SEM). NJ = Jenguma variety

Grain yield

There was significant interaction (P = 0.039) effect between genotype and fertilization regime on grain yield (Table 2). Jenguma treated with Tsp + inoculant recorded the highest grain yield (4.06t/ha), which was significantly similar to Jenguma treated with Tsp + nitrogen + inoculant (3.86t/ha), followed by genotype N135 treated with Tsp + nitrogen (3.74 t/ha). The least grain yield (0.01 t/ha) was recorded by genotype N19 without treatment (control).

Table 2: High-yield soybean genotype and fertilization regime effect on grain yield (t/ha)

Fertilizer Regime	**N119**	**N135** (← Genotypes →)	**N19**	**NJ**
Control	1.428	2.325	0.010	1.685
Nitrogen	1.174	1.905	0.325	1.549
Tsp + I	2.127	3.295	0.678	4.062
Tsp + N	1.756	3.743	1.089	1.956
Tsp + I + N	3.251	3.301	0.261	3.862
Tsp	2.657	2.781	0.690	3.435
Inoculant	1.959	1.930	0.941	2.300
LSD (5%)	**1.66**			
CV (%)	**28**			

NJ = Jenguma variety

Economic analysis

Based on the results on comparative analyses of the economic productivity of soybean production, soybean treated with sole Tsp gave the highest benefit cost ratio, followed by the soybean with Tsp + nitrogen + inoculant. Treatment with Tsp + nitrogen and Tsp + inoculant recorded the same benefit cost ratio, followed by soybean treated with sole inoculant (Table 3). The control treatment (sole soybean) gave a higher benefit-cost than Soybean with sole nitrogen, which gave the least benefit cost ratio.

Table 3: Benefit and cost analysis for production of high yield soybean genotypes as influenced by fertilization

Technology	Cost of production	Benefit (GHC)	Benefit/Cost ratio
Control	1245	2828.5	2.3
Tsp+soybean	1695	4325.2	2.5
I+soybean	1470	2602.5	1.7
N+soybean	1445	1919.2	1.3
Tsp+N+I+soybean	1870	4858.4	2.5
Tsp+I+soybean	1795	5287.6	2.9
Tsp+N+soybean	1770	4781.4	2.7

Yield and yield components

Improved nodule count led to increase N fixed into the soil; this improves soil fertility and increase crop development and growth (Bogino *et al.,* 2011). The greatest nodule number recorded by Tsp + inoculants might be that phosphorus played an important function in the formation of nodule and atmospheric nitrogen fixation. Tagoe *et al*. (2008) showed that the availability of P helps to initiate the formation of nodules as well as growth, development and functioning of nodules formed. This might explain why all the treatment combinations with Tsp produced higher number of nodules (Figure 2), and percentage of effective nodule than the other treatments (Figure 3). The non-significant effect of nitrogen application on number of nodule is in line with the finding of Seneviratne *et al*. (2000) that the presence or absence of nitrogen in the soil does not significantly impact nodulation of the crop. The non-significant differences in biomass weight among soybean genotypes are in agreement with (Lambon *et al.,* 2018) Who indicated no significant difference in biomass among varieties. The trend in biomass weight was Tsp + inoculant + nitrogen >Tsp + nitrogen >Tsp+I> and shows that nitrogen, and/or Tsp improved vegetative growth. This implies that omission of N and Tsp from soybean production could reduce biomass yield as suggested by Bekere and Hailemariam (2012) (Figure 4).

The pod weight was influenced statistically (P = 0.042) by mineral fertilization and inoculant application (Figure 5). The greatest pod weight by Tsp + inoculant could be attributed to the presence of Tsp in the treatment combination as the treatments which contained Tsp produced heavier pods weight than the other treatment combinations. Nitrogen application did not positively influence dry weight and number of pods per plant. Khan (2000) reported that Phosphorus application and Rhizobium inoculation significantly increased pod formation, grain yield and dry matter production as compared with un-inoculated treatments.

The least 100 seed weight recorded by N19 though it produced the highest number of pods could be attributed to the fact that N19 is a late maturing genotype and was affected by climatic conditions. There was water stress during pod filling stage and N19 could not fill its pods before the draught set in due to lateness in its maturity (Figure 6). The results also agree with the work of Masoumi *et al*. (2011) who reported that soybean yield could significantly be reduced by reduced soil moisture during flowering and pod filling period.

Genotypic difference played a significant role on the grain yield in the first phase since the only variable factor was the different genotypes, while every other factor was the same. The greatest grain yield by N119 during the screening phase could be attributed to its ability to utilize environmental conditions as an early maturing genotype (Figure 1a). This is in consonance with the report by Bouquet (1998), that genotype selection is one of the most important factors for increasing pod yield in soybean.

The significant interaction between soybean genotypes and fertilization in the second phase could be attributed to differences in response of genotype to fertilizer regimes (Table 2). Jenguma gave the highest grain yield when treated with Tsp combined with inoculants (4.062 t/ha), which was statistically similar to Jenguma treated with Tsp + inoculant + nitrogen (3.862 t/ha). The result of this experiment is in agreement with the findings of Ebenezer *et al*. (2019) in relation to Jenguma who reported that soybean variety treated with Tsp combined with inoculant recorded higher grain yield per hectare than other treatment combinations. The findings of Ebenezer *et al*. (2019) contradict this result in relation to N19, N135 and N119, because these genotypes combination with Tsp + N and Tsp + N +I each, gave the highest grain yield. However, generally, Tsp, N or I, a combination of two or three enhanced grain yield. Similar research that supports findings of this study is Jalaluddin (2005). Khan (2000) stated that pod formation and grain yield are significantly enhanced with the application of phosphorus and rhizobium inoculant as compared with un-inoculated treatments.

Economic cost analysis

Information on cost and benefit of treatments is a prerequisite for adoption of technical innovation by farmers (Das *et al.,* 2010). The use of booster N in soybean production was associated with the least production cost among the technologies used except for the control. Tsp + inoculant + N recorded the highest revenue generation and also the highest cost. Rhizobia inoculants + Tsp treatment on soybean produced higher yields and is comparatively cheaper than Tsp+ inoculant + booster N application, while the control gave the least cost ratio but a higher benefit than sole N fertilizer (Table 3). From the observations on comparative analyses of the technologies used in soybean production, it is recommended for farmers to go for Tsp + inoculants as soybeans treatment since this will give a higher yield at minimum cost. The findings of this research are contrary to Adegeye and Dittoh (2011), who stated that the higher the benefit-cost ratio, the higher the gain derived from the use of the given production system.

Conclusion

The interaction effect is an indication that improved varieties treated with mineral fertilizer with inoculant will increase yield. The study also shows that inoculation of soybean seeds with P leads to increase in almost all yield and growth parameters of soybean. Resource poor farmers in Africa can also opt for sole P if inoculation and booster N fertilizers becomes expensive to come by, since it is economically beneficial. It is recommended that soybeans producers should inoculate seeds and also apply or fertilize their soil with P and booster N to increase soybean yield.

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