

Research Article

Evaluation of a New Rhizobial Inoculant for Soybean (*Glycine max***) Production in Semi-Arid Zone of Nigeria**

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ABSTRACT

An investigation was carried out in a pot experiment to access the effect of a rhizobial inoculant on the performance of soybean varieties. This study was conducted at the Teaching and Research Farm of the University of Abuja, Nigeria. Three varieties of soybean (TGm 263, TGm 295 and TGm 307) and 2 rhizobial inoculation levels (with and without) were set up as a factorial arrangement in a completely randomized design. Plant height, number of leaves, shoot and root weight, number and weight of nodules were among the parameters evaluated. Significant interaction was observed between soybean varieties and rhizobial inoculation. Shoot dry weight under non-inoculated TGm 307 (TGm307*NRh) increased by 49.5% as compared to the inoculated TGm 307 (TGm307*Rh). However, with rhizobia inoculation, number of nodules for TGm 263 (TGm263*Rh) significantly increased by 61% over that of uninoculated TGm 263 (TGm263*NRh). The study indicated inoculants interaction with soybean varieties differed across parameters.

Key words: Soybean varieties, Inoculation, Biomass, Interaction

INTRODUCTION

For Africa, Nitrogen still remains one of the most limiting nutrients for the growth and production of crops despite its abundance in the atmosphere. Increasing cost of inorganic fertilizer has forced farmers in sub-Saharan region to embrace biological nitrogen fixation as a complementary nitrogen source (Bala *et al.*, 2003; Peoples *et al.*, 1995). The benefits of Soybean to human, animal nutrition and soil fertility enhancement through biological N fixation are well known and appreciated (Mpepereki *et al* 2000; Graham and Vance 2003; Osunde *et al.*, 2003; Vanlauwe *et al.*, 2014).

Yield decline and poor nodulation of soybean in African soils have been linked to the absence of effective bradyrhizobial strains and unavailability of rhizobial inoculants. Besides, awareness of farmers on inoculants, their uses are limited and, as such, most farmer cultivate without inoculation (Abaidoo *et al.*, 1999). Seed inoculation with relevant strains of bacteria is necessary prior to sowing except for fields where soybean has been recurrently cultivated over years (Okereke and Onochie, 1996). Rhizobial inoculation helps in crop yield and soil fertility enhancement and with reduced soil nitrogen demand, the soil pollution effects from chemical fertilizers is consequently minimized (Graham and Vance, 2003; Brutti *et al.*, 2001).

Nitrogen fixation in soybean could vary among genotypes and rhizobial strains and as well, their interaction (Okogun and Sanginga 2003; Sameh *et al.*, 2014). The unity and selectivity between rhizobial strain and a variety of soybean is necessary for a favorable nodulation, thus suitable rhizobial strains must be sourced. A study was, thus, setup to determine the effectiveness of a new rhizobial inoculant on the growth, biomass yield and nodulation of promiscuous soybean varieties.

MATERIALS AND METHODS

Site description and soil preparation

This experiment was carried out in pots at the University of Abuja Teaching and Research Farm ($08^{\circ} 40'$ N, $07^{\circ} 12'$ E). Soil was randomly sampled at a depth of 0 – 15 cm and bulked to a single composite sample. A representative sub-sample was taken after a thorough mixing and prepared for a routine soil physical and chemical property analysis. Thereafter, 5 kg of soil were weighed into 18 pots.

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Experimental Design and Treatment application

treatments were laid out as factorial The combinations in complete randomized design and replicated three times. The factors were soybean varieties (TGm 263, TGm 295, TGm 307) developed by IITA Ibadan and rhizobial inoculation (with and without inoculant). Rhizobia inoculant used was a new peat based carrier inoculant containing bradyrhizobial strains applied at 100 g per 10-20 kg of soybean. Inoculation was done by mixing 5 g of inoculant into 5% gum arabic solution. The formed slurry was then mixed with dry seeds until an even coating was achieved. Coated seeds were then air dried under shade for 30 minutes prior to planting. Uninoculated seeds were first planted to avoid contamination. Two seeds were planted in each pot at 2 cm depth and then thinned to 1 plant per pot 2 weeks after planting (WAP). Weeding was done regularly by hand pulling during the period of the experiment.

Data collection and Analysis

Parameters measured were plant height and numbers of leaves per pot assessed fortnightly with meter rule and counts respectively; shoot and root biomass per plants, weight and number of nodule per plant. Plant shoots were cut from the base and each pot emptied onto a mesh on a plastic bag and sieved. Detached root and nodules were picked from soil prior to washing off adhering soil under tap water. Nodules were carefully detached from the root and counted. Fresh root, shoot and nodules were oven dried at 80°C to a constant weight and their dry weight recorded. All the data collected were subjected to analysis of variance using statistical package SAS Version 9.2. Means were separated using Duncan multiple rang test at $P \leq 0.05$.

RESULTS

Physical and Chemical properties of soil before planting

The soil physico-chemical properties showed that soil was of clay loam texture, slightly acidic (pH = 5.8), with a moderately available P (18 mg kg⁻¹). The organic carbon, total nitrogen and exchangeable cation of the soil were low.

Varietal and inoculation effects on soybean growth and nodulation

At 6 and 7 WAP, a significant varietal effect was observed on number of leaves of soybean plant. TGm 295 produced the highest number of leaves with 38.17 and 49.5 at 6 and 7 weeks, respectively (Figure 1). Plant height was also influenced significantly by soybean variety, from the fourth to the ninth week. TGm 295 had the highest plants height, from the fourth week to the end of the experiment (Figure 2).

A varietal effect was also observed on fresh shoot and root weights. TGm 295 had the highest fresh shoot and root weight of 170.33 g and 21.95 g, respectively. It was followed by TGm 307 and TGm 263 with 13.78 and 10.45 g, respectively. There was however no significant difference between soybean varieties for nodulation (nodules number, fresh and dry weights), root length, dry shoot and root weights (Table 2).



Fig. 1: Effect of soybean varieties on number of leaves (bars with same letter, within the same period, are not significantly different).



Fig. 2: Effect of soybean varieties on plant height (ns = non-significant, *, ** and *** = significant at P \leq 0.5, P \leq 0.01 and P \leq 0.001 at indicated growth period).

Гab	ole 1	l: Soil	physica	l and c	hemical	characteristics
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Parameters	Values	Parameters	Values
pH (H ₂ O)	5.8	Exchangea	ble cations
-		(Cmo	l kg ⁻¹)
$OC (g kg^{-1})$	1.8	Ca ²⁺	2.5
$N(g kg^{-1})$	0.01	Mg^{2+}	3.11
Available P (mg kg ⁻¹)	18	K^+	0.35
Exchangeable Acidity	1.11	Na ⁺	0.4
Sand (g kg ⁻¹)	610	ECEC	7.47
Silt (g kg ⁻¹)	130		
Clay (g kg ⁻¹)	260		
Textural class	Clay loam		

The inoculation of soybean showed some significant effects on growth parameters, especially from the sixth WAP on the number of leaves produced by the plant. As well, there was a significant difference on plant height due to inoculation as observed at 6, 7 8 and 9 WAP. Furthermore, inoculation effects on other parameters such as nodulation, shoot and root biomass did not differ significantly.

Interaction effect between varieties and rhizobial inoculation on soybean development

From the results, soybean development was significantly influenced by the inoculation treatment. The non-inoculated TGm 295 variety (TGm295*NRh) had the highest number of leaves (46 leaves per plant) compared with other interaction treatments (Figure 3). This was followed by the uninoculated TGm 263 variety (TGm263*NRh).



Fig. 3: Interaction effect between soybean varieties and rhizobial inoculation on number of leaves and plant height (ns = non-significant, *, ** and *** = significant at $P \le 0.5$, $P \le 0.01$ and $P \le 0.001$ at indicated growth period).

Table 2: Ef	fect of soybean	varieties on	the shoot an	nd root bion	nass and nod	ulation variables
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Variation	Shoot (g)		Root				Nodules		
varieties	FW	DW	FW (g)	DW (g)	Length (cm)	Number	NFW (g)	NDW (g)	
TGm 263	112.22 a	18.78 a	10.45 a	3.2 a	39.43 a	93.17 a	2.87 a	0.35 a	
TGm 295	170.33 a	32.52 a	21.95 a	2.67 a	34.25 a	109.67 a	3.62 a	0.46 a	
TGm 307	132.00 a	25.90 a	13.78 a	3.38 a	37.55 a	70.42 a	2.75 a	0.39 a	

FW = fresh weight, DW = dry weight, NFW = nodules fresh weight, NDW = nodules dry weight. Means with same letter are not statistically different according to Tukey test at P ≤ 0.05 .

Table 3: Rhizobial inoculation effects on soybean number of leaves and plant height

	Time of measurement (weeks)										
	1	2	3	4	5	6	7	8	9		
	Number of	leaves									
Inoculated	3.89 a	6.22 a	7 a	10.56 a	15.89 a	29.56 b	41.33 b				
Uninoculated	5.22 a	6.67 a	6.78 a	9.89 a	20.56 a	36.67 a	48.56 a				
	Plant heigh	t (cm)									
Inoculated	21.11 a	24.44 a	33.22 a	39.78 a	48.56 a	58.22 a	69.44 b	78.11 b	91.33 b		
Uninoculated	21.67 a	25.11 a	33.33 a	41.78 a	51.56 a	65.11 a	76.00 a	91.67 a	104.11 a		
Means with same	e letter are not	statistically o	lifferent acco	rding to Tuk	ev test at P<	0.05.					

Table 4: Inoculation effects on soybean nodulation, shoot and root biomass.

Inoculation	Shoot* (g)		Nodules*			Root*		
liloculation	FW	DW	Number	NFW (g)	NDW (g)	FW (g)	DW (g)	Length (cm)
Inoculated	126.78	23.19	106.89	3.2	0.43	12.52	3.31	39.34
Uninoculated	149.59	28.28	75.28	2.96	0.37	18.27	2.86	34.81
FW – fresh weight	DW - dry	weight N	FW – nodules	fresh weight	NDW – nodules	dry weight	* – no signifi	cant difference was

FW = fresh weight, DW = dry weight, NFW = nodules fresh weight, NDW = nodules dry weight, * = no significant difference was observed on the variable.

Inoculation of soybean varieties with the newly manufactured rhizobial inoculant revealed that non-inoculated TGm 295 (TGm295*NRh) produced the highest shoot fresh weight with 179.8 g. The non-inoculated TGm 307 (TGm307*NRh) produced significantly higher shoot dry weight than the inoculated TGm 307 (TGm307*Rh) with 175.47 g and 88.53 g, respectively (Table 5).

On the number of the nodules, there was significant interaction between soybean varieties and rhizobial inoculation. An increase of 61% was obtained for number of nodules in the inoculated TGm 263 (TGm263*Rh) over the uninoculated TGm 263 (TGm263*NRh) with 52.33 nodules. The root fresh weight was also significantly, influenced by the interaction between soybean varieties and rhizobial inoculation. The non-inoculated TGm 295 (TGm295*NRh) had the highest root fresh weight of 28.9 g (Table 5).

DISCUSSION

Soybean is an important grain legume playing key roles in SSA. The satisfaction of the roles of soybean are

based only on its capacity to nodulate profusely and fix abundantly atmospheric nitrogen (Okogun and Sanginga 2003, Tefera, 2011). Inoculation of promiscuous soybean varieties, sometimes, result in failure which translates the non-competitiveness of introduced rhizobial strains (Thies et al., 1991; Ojo et al, 2014). This fact could explain the insignificant effect of inoculation observed in this experiment on soybean nodulation, shoot yield and root attributes. Many factors are responsible for the competitiveness between introduced and local rhizobial strains (Mårtensson et al., 1989; Thies et al., 1991; Brutti et al., 1999). Furthermore, an incompatibility factor between soybean varieties and the rhizobial inoculant could also, be involved. In fact, it was observed that the non-inoculated soybean had significantly high number of leave at 6 and 7 WAP and plant height from 7 to 9 WAP.

There was an interaction between soybean and rhizobial inoculation. However, the non-inoculated TGm 295 (TGm295*NRh) had the highest number of leaves at 6 and 7 WAP, and plant height at 6, 7 and 8 WAP. This observation emphasized the point that the rhizobial strain might not have been compatible with this variety. However, on nodulation, TGm 263 responded positively

Table 5: Comparison of soybean variety and rhizobial inoculant effect on shoot fresh weight, number of nodules and root fresh weight
Least Squares Means for effect Variety*Inoculation

Pr > t for H0: LS Mean (i) = LS Mean (j)										
Dependent Variable: Shoot fresh weight										
i/j	TGm263*Rh	TGm263*NRh	TGm295*Rh	TGm295*NRh	TGm307*Rh	TGm307*NRh				
TGm263*Rh		0.57	0.7591	0.31	0.4474	0.3987				
TGm263*NRh			0.0883	0.0214	0.9999	0.0297				
TGm295*Rh				0.9521	0.0612	0.984				
TGm295*NRh					0.0147	1				
TGm307*Rh						0.0204				
TGm307*NRh										
Means	130.93	93.5	160.87	179.8	88.53	175.47				
		Dep	endent Variable: N	lumber of nodules						
i/j	TGm263*Rh	TGm263*NRh	TGm295*Rh	TGm295*NRh	TGm307*Rh	TGm307*NRh				
TGm263*Rh		0.029	0.8482	0.8964	0.1956	0.0626				
TGm263*NRh			0.1912	0.1592	0.8415	0.9968				
TGm295*Rh				1	0.7524	0.3636				
TGm295*NRh					0.6861	0.3103				
TGm307*Rh						0.9765				
TGm307*NRh										
Means	134	52.33	108.33	111	78.33	62.5				
		Dep	endent Variable: I	Root fresh weight						
i/j	TGm263*Rh	TGm263*NRh	TGm295*Rh	TGm295*NRh	TGm307*Rh	TGm307*NRh				
TGm263*Rh		0.9972	0.9036	0.0045	1	0.7476				
TGm263*NRh			0.6926	0.0022	0.9969	0.4966				
TGm295*Rh				0.0244	0.9067	0.9992				
TGm295*NRh					0.0045	0.0436				
TGm307*Rh						0.7524				
TGm307*NRh										
Means	11.27	9.63	15	28.9	11.3	16.27				

to the rhizobial inoculation: inoculated TGm 263 had significant number of nodules as compared with noninoculated TGm 263. Furthermore, a higher symbiotic performance between TGm 295 and the native rhizobial strains was revealed. Preferences of native rhizobial strains were also observed by Shutsrirung *et al.* (2002) during a large screening for soybean varieties symbiotic efficiency with native rhizobia.

Conclusion

The present study has shown that rhizobia inoculants differ with soybean varieties. The study satisfied the objective of breeding promiscuous varieties which is to increase the gain of soybean from indigenous rhizobia. Through the evaluated parameters, TGm 295 development was generally improved by native rhizobia. Further research activities are consequently needed to study the response of some other soybean varieties to this newly introduced inoculant.

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