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Influence of Different Nodumax Stickers on Nutrient Content of Soybean (*Glycine Max*) Varieties Grown on an Ultisol in Derived Savannah Ecology of Ishiagu, South-East Nigeria

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Abstract
The study was carried out to assess the influence of different nodumax stickers on the nutrient and mineral contents of soybean varieties grown on an ultisol. In the trial, a split plot arrangement fitted into a randomized complete block design (RCBD) was used. The three varieties (TGX 1 448-2E, TGX 1 445-2E and TGX 1 485-1 D) made up the main plots, while the six nodumax inoculant stickers (gum arabic, milk, vegetable oil, no sticker, water and control) made up the sub-plots. Pre-planting physico-chemical properties of the soils were determined to know the soil status, while inoculation was done in the field before planting. Grain nutrient and mineral contents such as crude protein, crude fat, crude fibre, ash, moisture content, dry matter, % phosphorus, potassium, calcium and magnesium were determined from the mature grain obtained at 12% moisture content. After the data analysis, the results revealed that variety significantly ($P<0.05$) affected the contents studied with variety TGX 1 448-2E giving the highest values, except for protein, fibre, phosphorous, iron calcium and magnesium content which were not significantly different from the others. The effect of sticker was also significant ($P>0.05$) on the nutrient contents studied except on iron and phosphorous which did not show any significant ($P>0.05$) difference. The values recorded in plots which had gum arabic as sticker was highest, though statistically similar to those with milk as sticker. Gum arabic, therefore, can be substituted with milk when gum arabic is not available.

Keywords: Nodumax, inoculant, Stickers, Soybean varieties

Introduction

Nodumax is a recent legume inoculation technology developed at the International Institute of Tropical Agriculture (IITA), Ibadan, Nigeria to enhance the yield of soybean, and also to replace the use of nitrogen fertilizer in soybean production (IITA, 2015). Nodumax is a type of biofertilizer that contains 50% of *Rhizobium* culture and 50% peat-based carrier. Nodumax was developed to resolve the issue of low crop productivity of most legume crops. The use of Nodumax can increase soybean yield by 30-40% (IITA, 2015). Soybean (*Glycine max*); a legume in the family *Fabaceae*, is becoming more and more accepted in the Guinea savanna of Nigeria because of its high nutrient value. It is an important oilseed crop and contributes to soil productivity through N_2-fixation (Shu-Jie *et al*., 2007). After soybean grain has been harvested, nitrogen contents of root hairs and other plant parts are made available to the soil through decomposition, leading to lower production costs and lessening problems of soil contamination caused by nitrogen fertilizers, such as nitrate (Ogoke *et al*., 2003). When soybean roots are infected by the right *Rhizobium* by inoculation, nodules are generally formed and have high prospect for N_2 - fixation (Shu-Jie *et al*., 2007).

Inoculation is the process of adding effective bacteria to the host seed or soil before planting with the purpose of providing appropriate number and type of bacteria needed for a successful establishment of legume-bacterial N_2 - fixation (Benizri *et al*., 2001). One of the issues affecting the use of inoculants is problems associated with sticking the inoculant with the seeds during the application process, as this affects the efficiency of the inoculants (IITA, 2015). Gum arabic is the sticker normally enclosed with the inoculant, but it easily gets hardened, making usage difficult. When farmers cannot overcome these problems, they see no need for inoculation, thus reducing their adoption of the technology, making both the manufacturers and the users the losers. For this reason, it is desirable that suitable and user-friendly inoculum sticker be identified and made known to the farmers. According to Ferreira and Castro (2005), some sticker materials that have been identified include; carboxymethyl cellulose, sugar solution, corn syrup, honey, powdered milk, evaporated

milk, mineral oil, or a vegetable oil such as peanut oil or soybean oil. The most excellent sticker for nodumax application is yet to be determined (IITA, 2015). The objective of this study was to determine the most effective readily available sticker that can be used in place of gum arabic; and the sticker that will best enhance the nutritional content of soybean seeds. Since, there has been development of improved varieties and production of soybean has been concentrated in the Sudan, Sahel and Guinea Savannah ecological zones of Nigeria (IITA, 1992). There is also a need to evaluate these improved varieties; the ones suited for Ishiagu in the derived savannah ecology of South-East Nigeria.

Materials and Methods

Location of Experimental Site

The experiment was conducted at the Research and Teaching Farm of the Federal College of Agriculture, Ishiagu (FCAI), South-east Nigeria. The area lies between latitude 5º 55´ N and 6º 00´ N, and longitudes 7º 30´ E and 7º 35´ E in the derived savannah agro-ecology. The mean annual rainfall of the area is 1350 mm, spread from April to October with average air temperature of 29ºC. The underlying geological material is Shale formation with sand intrusions locally classified as the 'Asu River' group. The soil is hydromorphic and belongs to the order Ultisol. It has been classified as Typic Haplustult (Onunwa *et al.*, 2016).

Experimental design and layout

The experiment was a split plot arrangement laid out in a randomized complete block design (RCBD) replicated three (3) times. There were three main plots each containing six sub-plots. The three main plots contained cultivars V1= TGX 1 448-2E, V2= TGX 1 445-2E and V3= TGX 1 485-1 D; while the sub-plots were the six stickers namely T_1= Gum Arabic, T_2 = Liquid milk, T_3= vegetable oil, T_4= no sticker, T_5= Water and T_6= Control (No Inoculation). Each main plot measure 15.5 m by 2 m (31.0 m^2) and distance between main plots was 1m. Each sub plot measure 2m by 2m (4m^2) and distance between sub-plots was 0.5m.

Source of Experimental Materials

The *Rhizobium* inoculant (NODUMAX) and the three varieties of soybean seeds were obtained from the International Institute of Tropical Agriculture (IITA), Ibadan in Nigeria. Gum Arabic, one of the stickers used, was enclosed in the Nodumax pack, while the other stickers such as milk, vegetable oil, liquid milk and water were obtained from the open market in Ishiagu.

Inoculation Procedures

Preparation of Stickers- Clean lukewarm water measuring 5mls was poured into a 30 ml plastic bottle and 5 g (half tablespoon) of each sticker was added and shaken vigorously.

Inoculation-The inoculation was done in the field just before planting using the Slurry method outlined by Woomer *et al*. (2011). In the case of no sticker, the inoculant was sprayed on the seeds.The inoculated seeds were put in a container covered with cloth to protect it from sunlight.

Agronomic Practices

Sowing was done after the process of inoculation was carried out. Three seeds were planted per hole at a depth of 4 cm. The seedlings were later thinned down to two seedlings per hole at 2 weeks after sowing (WAS). Before sowing, a basal application of phosphorus fertilizer (P_2O_5) at 30kg/ha was done. Soybean seeds were sown in rows using plant spacing 50 cm x 50 cm apart. Weeding was done manually by hand using a hoe at 3 and 6 WAS.

Plant Sampling

The harvested seeds were collected per treatment, rinsed with distilled water, oven-dried at 70°C for 48 hours and ground using the Thomas stainless steel milling machine. Nutritional contents of the seeds (crude protein, fat, crude fiber, total ash and moisture contents) were determined according to AOAC (1990). The concentrations of the selected heavy metals were determined using atomic absorption spectrophotometer (Unicam Solar 32 model) following the standard procedures presented in AOAC (1999). The rating for adequacy or otherwise of concentrations was done according to Landen (1991). Total nitrogen was determined by the modified micro Kjeldahl distillation methods (Bremner, 1993).

Data Analysis

The data collected were subjected to analysis of variance (ANOVA) according to the procedure for split-plot in randomised complete block design using GENSTAT 3 7.2 Edition. Significant treatment means were separated and compared using fishers Least Significant Difference FLSD at 5% probability level.

Results and Discussion

Soil properties

Pre-planting Physico-chemical Properties of the Soil

The pre-planting physical and chemical properties of the soil are presented in Table 1. The soils were texturally sandy loam with 13% clay, 20% silt and 67% sand contents. The bulk density was 1.39 mg/m^3 with soil porosity of 47.57%, saturated hydraulic conductivity of 0.36 cm/h and particle size of 2.56 g/cm^3 Soil organic carbon concentration recorded was 6.47 g/kg, whereas the total soil nitrogen was 1.35 g/kg. The pH measured in water was 5.2 and the value of 10.60 cmol/kg was recorded for cation exchange capacity. The analysis also indicated that the exchangeable sodium, potassium, calcium and magnesium were; 0.11 cmol/kg, 0.17 cmol/kg, 3.37 cmol/kg and 2.40 cmol/kg, respectively. In addition, the exchangeable acidity and available phosphorous in the studied soil had values of 1.10 cmol/kg and 4.31 mg/kg, respectively. Nutrient and mineral composition of soybean seeds as influenced by variety and nodumax stickers. The results of the nutrient composition of soybean seeds from the different varieties and stickers are presented in Table 2.

Crude Protein
TGX 1448-2E variety presented the highest protein content of 37.58% which did not significantly (P>0.05) differ from that of TGX 1445-2E (36.79%) and TGX1485-1D (36.65%) varieties (Table 2). Seeds harvested from inoculated plots with gum arabic as sticker had the highest protein content of 37.21% which significantly (P<0.05) differed from values obtained from water (35.14%) and control (35.02%), but similar to the rest stickers. However, the result indicated a significant difference in the interaction. The highest value was recorded on the plots with TGX 1448-2E and gum arabic as sticker.

Crude Fat
Fat content was significantly (P< 0.05) affected by variety. The highest fat value of 28.01% and 27.92% were exhibited by TGX 1448-2E and TGX 1445-2E, respectively while TGX1485-1D variety had the least value of 25.54%. Gum Arabic and milk obtained the same fat value of 28.11% which was significantly higher than fat contents of the other stickers. The interaction of the two factors was significant, with the highest interaction recorded in TGX 1445-2E with milk.

Crude fibre
The crude fibre of all the soybean varieties were not significantly (p>.0.05) different. However, the effect of stickers was significant as values ranged from 5.49% of control to 8.84% of gum arabic. More so, the interaction of the variety and stickers indicated no significant (P<0.05) effect on crude fiber content.

Ash content
TGX 1448-2E variety with ash content of 4.12% had the highest value, and was significantly (P< 0.05) higher than the fat content of 3.60, and 3.51% of TGX 1445-2E and TGX1485-1D varieties, respectively. As for the stickers, gum arabic with ash content of 4.28% was significantly (P< 0.05) higher than the ash contents of other stickers used in the field experiment. The interaction between variety and sticker was not significant.

Moisture Content
The moisture contents of the evaluated soybean varieties significantly varied and ranged from 0.71% (TGX1448-2E) to 1.32% (TGX1485-1D). Seeds harvested from the control plots recorded much more moisture content (1.13%) which was significantly (P< 0.05) higher than the values obtained from the rest stickers. The result aso indicated a significant difference due to interaction. The highest interaction value was obtained between TGX 1445-2E and milk.

Dry matter
There was significant (P< 0.05) difference on the dry matter content due to varieties used, the highest dry matter of 12.02% recorded in TGX 1448-2E was statistically similar to 11.74% obtained from TGX 1445-2E, but differed from 8.77% gotten in TGX1485-1D. On stickers, dry matter content ranged 8.72% - 11.87% with seeds from control treatment having the least dry matter, while those from gum arabic exhibited the highest dry matter content. On interaction, the result indicated a significant difference among the treatments, with the highest value recorded between TGX 1448-2E and milk.

The results of the mineral composition of soybean seeds from the different varieties and stickers are presented in Table 3.

Sodium (Na) and Zinc (Zn)
Table 3 shows that Sodium and Zinc contents were significantly (P< 0.05) improved by both varieties and stickers. TGX 1448-2E had the highest Na and Zn values of 3.0 mg/100g and 2.7 mg/100g, respectively. Seeds harvested from inoculated plots having gum arabic as sticker also had the highest values of Na and Zn of 3.3 mg/100g and 3.0 mg/100g, respectively. The effect of interaction was significant in both Na and Zn contents.

Phosphorus (P) and Iron (Fe)
Unlike NA and Zn, the effect of varieties and stickers on percent P and Fe were not significant (P >0.05).The values ranged from 460.22 mg/100g to 460.81 mg/100g and 15.87 mg/100g to 15.99 mg/100g for P and Fe as obtained in TGX1485-1D and TGX 1445-2E varieties respectively. For stickers, values ranged from 450.12mg/100g to 451.01 mg/100g and 15.11 mg/100g to 15.35 mg/100g for P and Fe respectively. Similarly the effect of interaction was not significant for both P and Fe contents.

Calcium (Ca) and Magnesium (Mg)
The different varieties of soybean did not show any significant (P> 0.05) effect on the Ca and Mg contents of the harvested seeds. However, the Ca content of plots inoculated with gum arabic gave the highest significant (P< 0.05) value of 300.51 mg/100g over other stickers. The Mg content of milk (257.39mg/100g) was significantly (P< 0.05) higher than those from other stickers except gum arabic which was statistically similar.The interaction effect was negative.

Discussion
The values of protein, fiber, fat, ash and moisture contents of the seeds of the inoculated plots were enhanced when compared with the control. This could be as a result of increase in nitrogen fixation as a result the inculation, which increased biological activities and subsequently biosynthesis of nutrients. Zhang *et al.* (2002) reported that seed inoculation with *Rhizobium* increases nitrogen uptake and could be responsible for increased protein, carbohydrate and ash content including higher fatty acid content of soybean seeds. The higher values of sodium, zinc, phosphorous, iron, calcium and magnesium recorded from harvested from the inoculated plots particularly those with gum arabic and milk as stickers could be attributed to the fact that both stickers were more tenacious and gave the best adhesion more to seed surfaces than other stickers, which lead to increased root hair infection of the

Rhizobia, and increased N_2- fixation and nutrient availability. Ferreira and Castro (2005) reported that gum arabic is more tenacious and always provides good adhesion which increases N_2- fixation. Materials that however leave a sticky coating on the seed are better, such as oils or gum arabic. The larger nutriet and mineral values gotten from TGX 1448-2E variety can be attributed to the genetic makeup of the variety which is characterized by higher vegetative growth and resulted to higher nutrient and mineral accumulation when compared to other varieties. Aditya *et al.* (2011) observed that higher nutrient and mineral accumulation including different growth attributes shown by soybean varieties may vary under similar growth conditions, because they have different genetic makeup potential.

Conclusion

The use of TGX 1448-2E variety, gum arabic and milk as stickers proved to be effective in improving the nutrient value of soybean in the studied area, except protein and fiber which failed to respond to the differences in the soybean varieties. Gum arabic can therefore be substituted with milk, especially where gum ararbic is not readily available.

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Table 1: Pre-planting physical and chemical properties of the soil (0-20cm depth)

Soil properties	Value
Clay(%)	13
Silt (%)	20
Sand (%)	58
Textural class	Sandy loam
Bulk density (mg/m^3)	1.39
Porosity (%)	47.57
Saturated hydraulic conductivity (cm/h)	0.36
Particle size (mg/m^3)	2.56
pH (H_2O)	5.2
Organic Carbon (g/kg)	6.47
Total nitrogen (gkg^{-1})	1.35
Sodium (Na^+)(cmol/kg)	0.11
Potassium(K^+)(cmol/kg)	0.17
Calcium(Ca^{2+})(cmol/kg)	3.37
Magnesium(Mg^{2+})(cmol/kg)	2.40
Cation exchange capacity (cmol/kg)	10.10
Exchangeable acidity (cmol/kg)	1.10
Available phosphorous (mg/kg)	4.31

Table 2: Effect of varieties and nodumas stickers on some nutrient content Soybean evaluated in derived savannah ecology of Ishiagu

Treatments	Crude Protein %	Crude Fat%	Crude Fibre%	Crude Ash%	Moisture Content%	Dry matter%
Variety						
TGX 1448-2E	37.58	28.01	8.04	4.12	0.71	12.05
TGX 1445-2E	36.79	24.92	7.87	3.60	0.72	11.78
TGX1485-1D	36.65	25.54	7.80	3.51	1.32	8.77
$LSD_{0.05}$ variety	NS	1.24	NS	0.23	0.41	1.20
Stickers						
Gum Arabic	37.02	28.11	8.84	4.28	0.29	11.87
Milk	37.21	28.11	6.62	3.12	0.42	10.62
Vegetable oil	36.84	25.44	6.57	3.18	0.43	10.45
No sticker	36.74	25.98	6.71	3.01	0.40	10.51
Water	35.14	23.31	6.24	3.01	0.31	10.20
Control	35.02	23.11	5.49	2.54	1.13	8.72
$LSD_{0.05}$ sticker	0.83	1.76	1.98	1.01	0.24	0.85
$LSD_{0.05}$ variety vs sticker	0.54	1.01	NS	NS	0.23	0.94

Table 3: Effect of varieties and nodumas stickers on some mineral content Soybean evaluated in derived savannah ecology of Ishiagu

Treatments	Sodium mg/100g	Zinc mg/100g	Phosphorus mg/100g	Iron mg/100g	Calcim mg/100g	Magnesium mg/100g
Variety						
TGX 1448-2E	3.0	2.7	460.81	15.99	300.64	248.44
TGX 1445-2E	2.4	2.0	460.45	15.98	300.59	251.56
TGX1485-1D	2.2	1.8	460.22	15.87	300.55	248.10
$LSD_{0.05}$ variety	0.5	0.1	NS	NS	NS	NS
Stickers						
Gum Arabic	3.3	3.0	451.32	15.34	300.51	257.37
Milk	2.3	2.1	451.55	15.35	300.40	257.39
Vegetable oil	2.3	2.1	451.01	15.01	300.31	250.30
No sticker	2.1	2.0	451.33	15.24	300.32	250.35
Water	2.1	2.0	450.56	15.21	300.01	249.88
Control	1.9	2.0	450.12	15.11	300.01	248.21
$LSD_{0.05}$ sticker	0.2	0.5	NS	NS	0.81	6.21
$LSD_{0.05}$ variety vs sticker	0.1	0.6	NS	NS	NS	NS