AGRICULTURE ARCHIVES

OPEN ACCESS

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Original Research Article
ISSN: 3041-5322
Journal homepage: agriculture.researchfloor.org

Effects of Aluminium Application and Rhizobium Inoculation on Aluminium Accumulation, Nodulation and Yields of Soy beans Grown in Kenya

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ABSTRACT

Soybean yields output remains low due to the acidity of the soils in western Kenya. Aluminium toxicity is common in acidic soils and detrimentally affects the nodulation and yield of crops. It is important to investigate the effect of rhizobium inoculation of Soybeans on growth and yield. This study aims to examine the effects of inoculating Rhizobium in soy beans under aluminium application, to determine if it is possible to increase the output of GAZZELLE, NAMSOI, and TGX genotypes. The experiment was conducted at Maseno University under greenhouse conditions. In RCBD, eight treatments, three genotypes, and three replicates were employed. Aluminium accumulation, assessment of nodules, fresh weights, dry weights, number of seeds and seed weights at harvest data was collected. Tukey`s HSD tests at 5% was used to separate means. The genotype GAZZELLE was significantly lower ($p<.05$) compared to TGX at T1, T3, T5, T6 and T7 when Al accumulation was determined. This indicate the possibility of faster cell division in TGX plant roots that eventually limited nodulation and yield. Dry weights of nodules for TGX were significantly different at ($p < 0.01$) when compared to those of GAZZELLE and NAMSOI. Therefore, under Rhizobium inoculation and Aluminium application, Al may have interfered with Fe^{2+} capturing of bacteria in the latter two genotypes. Similarly, Rhizobium inoculated genotypes had significantly higher ($p<.05$) number of pink nodules under the Al application. Mean of NAMSOI was significantly higher ($p<.05$) compared to TGX for both total dry weights and dry weights of 100 seeds when treated with Rhizobium and Al. GAZZELLE and NAMSOI had a generally better performance when yield was considered. These results demonstrate that Rhizobium inoculation improves soy bean yield and mitigates the effects of Al. It, therefore, followed that Rhizobium-inoculated genotypes GAZELLE and NAMSOI are recommended for cultivation in acidic soils prone to Al.

Keywords: *Soy bean, Aluminium, Inoculation, Nodulation, Yield Response.*

1.Introduction

Soybean (*Glycine max* L.) grains are mainly utilised as food, medicine, and bioenergy [39]. Compared to Brazil and the United States of America, Kenya produces very less soy beans; this is partly due to the country's acidic soils [36]. In Western Kenya, a lack of genotypes resistant to acidic soils, which are often high in exchangeable Al has been associated with reduced levels of soy bean yield [22].

Acidity in the soil disrupts and restricts the nitrogen-fixing symbiosis [1]. Rhizobium inoculation can replenish nitrogen in acidic soils to provide competitive crop yields because Rhizobium species have the metabolic capacity to reduce Al stress [30]. An estimated 70 million tonnes of fixed nitrogen are added to agricultural soils each year by biological nitrogen fixations (BNF) [17]. However, due to variations in soil characteristics, soybean genotypes, Rhizobium strains, and plant species, the amount of N fixed can differ [11].

Citation: Mmayi M. P and Musyimi D. M (2025). Effects of Aluminium Application and Rhizobium Inoculation on Aluminium Accumulation, Nodulation and Yields of Soy beans Grown in Kenya.
Agriculture Archives: an International Journal.
DOI: https://doi.org/10.51470/AGRI.2025.4.2.07

Received on: April 08, 2025
Revised on: May 12, 2025
Accepted on: June 10, 2025

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Incase soybean seeds are planted in soils without being inoculated with the right symbiotic bacteria, or when nodules begin to senescesce during blooming, soybeans may experience a N shortage in the field [25]. In particular areas where soy bean has not been grown before [43], there is no success in nodulation therefore low seed yields are experienced. Low soil fertility effects can be reduced by inoculating soy bean seeds with the proper strain of Rhizobium before to planting. However, *Rhizobium* is known for its ability to fix up to 300kg/ha atmospheric nitrogen that can lead to increased grain and biomass yield [3]. It can alleviate low biomass and low grain production of soy bean plants in acidic soils caused by Al [3]. Consequently, the problem of soil pollution, which arises from excessive use of nitrogen fertilizers may be solved [6] as well as food insecurity.

Studies by [49] and [28] have revealed that *Rhizobium* inoculation increases nodule number, dry weight, pod number and yield of soy beans. [10] and [28] found that inoculating soy bean with *Rhizobium* may not offer agronomic benefits in some soils. [34] and [37] argued that inoculated *Rhizobium* is discharged by indigenous strains which are normally very competitive.

Due to the limited availability of significant amounts of organic fertilisers, such as farm yard manure, small-scale farmers typically utilise little to no mineral fertilisers, resulting in low crop yields [14. Farmers typically turn to low-cost sustainable alternatives to deal with the issue of poor biomass and grain output [47]. Beneficial microbes like Rhizobium, which can fix atmospheric nitrogen or aid in the uptake of very little amounts of nutrients, can be used to do this. Therefore, it is crucial to fully understand how these organisms affect plant development performance, particularly nutrient uptake and nitrogen fixation. Additionally, because soy beans fix nitrogen at high rates, as discovered in common beans [40], inoculation can increase quality production. A lengthy duration for pod filling may be possible in faba bean legumes due to leaf activity and ongoing N supply from fixation [15], which may forestall leaf senescence.

The objectives of the study were to determine the effect of aluminium application and Rhizobium inoculation on aluminium accumulation, nodulation and yield of three soy bean genotypes grown in Western Kenya. This is because food security is seriously compromised by low soy bean yields caused by Al toxicity. The best alternative method for increasing crop output in acidic soils may be the cultivation of Al-tolerant soy bean cultivars [28]. However, Rhizobium are soil bacteria that are distinguished by their special capacity to engage with legume root hairs and produce nitrogen-fixing nodules [40]. Consequently, Rhizobium inoculation of soy beans can lessen the effects of aluminium stress. Rhizobium raises the concentration of nitrogen in plants under Al, which may enhance crop yield and soybean tolerance under active nodulation for better growth [42]. Through improved nodulation quality and quantity as well grain production, this will assist fulfil the growing population's food needs.

2. Materials and Methods

2.1. Study site

In Vihiga County, the study was conducted from August 2021 to December 2022 in the greenhouse at Maseno University Research Farm which is found at joint operation graphic reference SA36-04 and a UTM position XE79 (Fig 1).

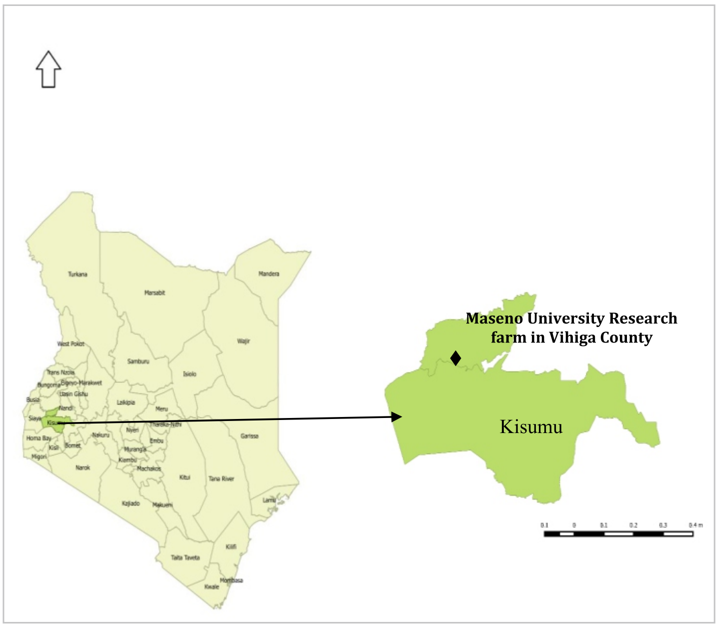

Fig. 1: Map showing study site; Maseno University Research farm. Source, Google maps

The soils of Maseno are nitisol and deep red. High exchangeable Al ions and a pH range of 4.9 characterise the well-drained soils. At Maseno, the average yearly rainfall is 1750 mm, and the average temperature is 27.8 °C. The temperature of the greenhouse fluctuated between 26±3 C (day/night) and 27-99% relative humidity.

2.2 Soil characterization

For laboratory analysis, soil samples were combined to create a composite sample, which was then pulverised, air-dried, and run through a 2 mm screen. The elements were identified and discovered as shown in table 1 below.

Table 1: Soil characteristics of the study site

Soil characters	Units	Result
pH	pH	4.9
Organic carbon	$g.kg^{-1}$	19.0
Total nitrogen	$g.kg^{-1}$	1.9
Total phosphorous	$g.kg^{-1}$	1.1
Total sulfur	$g.kg^{-1}$	0.3
Zinc	$mg.kg^{-1}$	3.4
Copper	$mg.kg^{-1}$	4.5
Cation exchange capacity	$mmol.kg^{-1}$	91.9
Clay	%	53.8
Sand	%	20.4
Total Aluminium	$g.kg^{-1}$	100.0
Total iron	$g.kg^{-1}$	84.0
Phosphorous	$mg.kg^{-1}$	24.6
Total manganese	$g.kg^{-1}$	4541.0

The table shows units of pH, organic carbon, total nitrogen, total phosphorous, total sulfur, Zinc, Copper, CEC, Clay, sand, total Al, total iron. Phosphorous and total manganese measured.

2.3. Inoculation of seeds, Planting and experimental design

The seeds were incubated for germination and considered successful as in [20]. A sterile disposable pipette tip was used to inoculate 1ml of *Bradyrhizobium japonicum* bacterial suspension as in the procedure of [48].

According to [35], soil from Maseno University Research Farm was placed in 20-litre PVC pots, in which seedlings were planted, and then triple superphosphate and potassium sulphate fertilizers were applied. A Randomized Complete Block Design with three replicates was employed. Aluminium chloride ($AlCl_3.6H_2O$) was prepared into varying concentrations to arise at eight treatments according to [35].

2.4. Determination of plant aluminium content

Dry ashing and hot oxidation was done to 0.5 g of the plant leaf sample. The chemical addition procedure of [32] was further followed. A simultaneous multi-element atomic absorption spectrophotometer (model 969; UNICAM, Cambridge, UK) was used to measure the amount of aluminium. According [41], an aluminium stock solution was prepared then diluted to standard series. The absorbance were then read using flame photometer. The absorbance reading was utilised to calculate the Al concentration from the standard curves in reference to [41] procedure;

Al content (g) in a 1 g sample equals C × df …………………….. (1)

where:

C = Al concentration (g/ml), as determined by the standard curve;

Df = dilution factor, which is 1000.

2.5. Determination and assessment of nodules

The nodules were counted and their fresh weights determined using Denver instrument XL-3100D instrument. The nodules were sliced and their colour determined as pink, brown, or green [26]. Nodules were oven dried to constant dry weights and their weights determined.

2.6. Determination of yield

2.6.1. Determination of fresh and dry weights

Fresh and dry weights were determined by the procedure of [12] using a Denver instrument XL-3100D weighing balance. A plant was randomly chosen for dry weight assessment. The above-ground plant system was cut off for dry weight assessment. The root system was gently removed down. The nodules were detached from the roots to be used for nodule assessment.

The above ground and below ground plant systems were air-dried then oven dried at 65^0 C overnight to a constant dry weight.

2.6.2. Determination of number of seeds

Pods from three randomly selected soybean plants at harvest maturity in a pot were opened [21], and number of seeds were counted.

2.6.3. Determination of seed weight at harvest

At harvest maturity all pods were harvested from the plants. The pods were oven-dried at 65^0 C overnight and separated into grains and husks. The weight in grams of clean 100 seeds selected randomly were used to give 100 seed weight [50]. The weight in grams of husks and seeds were recorded as the plant yield.

2.7. Statistical data analysis

The effects of genotypes, aluminium application and *Rhizobium* treatments were tested using the general linear model [46] in a 3 x 4 x 2 factorial way. Statistical differences among aluminium concentrations, between *Rhizobium* inoculations and among soybean genotypes were determined for the parameters measured using Tukey`s HSD test at 5% level.

3. Results

3.1. Aluminium concentrations in plants

Fig. 2 shows Al concentration in soy bean genotypes. Increase in Al application levels generally increased Al concentration in soy bean leaves while inoculation reduced Al concentration in genotypes. The mean of aluminium concentration in TGX was significantly higher than the mean in genotype NAMSOI and GAZZELLE for treatments T2, T4, T6 and T8, respectively.

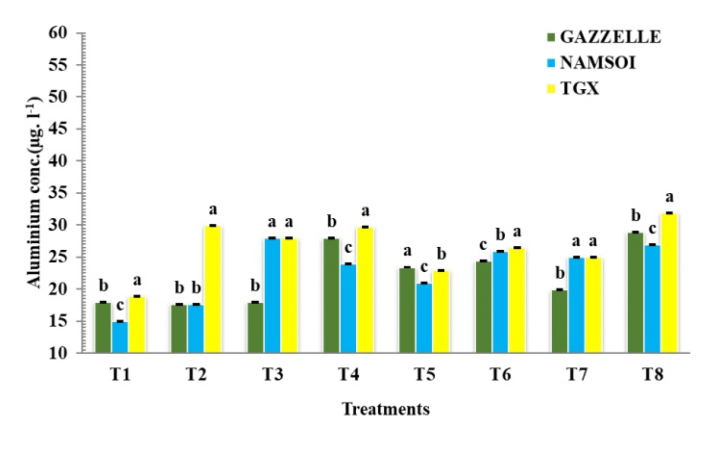

Fig.2. Response of Aluminium concentrations in three soy bean genotypes at maturity to Al application and *Rhizobium* inoculation. Values are means of three replicates±SEs. Means with the same latter are not significantly different. Treatments comprised of Control (T1) {Water*Inoculated}, T2 {480µM Al*Inoculated}, T3 {750µM Al*Inoculated}, T4 {960µM Al*Inoculated}, T5 {Control (Water)}, T6 {480µM Al}, T7 {750µM Al} and T8 {960µM Al}.

There was a significant interaction ($p < .01$) among aluminium application, *Rhizobium* inoculation and genotypes on aluminium concentrations. The mean plant aluminium concentration for each of aluminium application {960 µM Al (28.21 µg.l^{-1}), 750 µM Al (23.91 µg.l^{-1}), 480 µM Al (23.65 µg.l^{-1}) and control (19.83 µg.l^{-1})} were significantly different. The mean plant Al concentration of TGX (26.58 µg.l^{-1}) and NAMSOI (22.88 µg.l^{-1}) soy bean genotypes inoculated with *Rhizobium* and treated with aluminium were significantly higher than that of genotype GAZZELE (22.25 µg.l^{-1}).

3.2. Assessment of plant nodules

3.2.1. Fresh weights of nodules

Table 2 shows fresh weights of nodules of the three soy bean genotypes. Mean of soy bean genotype TGX was significantly higher than the means of GAZZELLE and NAMSOI, respectively at control*inoculated treatment (Table 2). Generally, mean at treatment T5 was significantly higher than the other seven treatment means.

Aluminium application ($p = .0046$) and soy bean genotypes ($p = .0082$) on fresh weights of nodules showed that there were significant differences. Mean fresh weights of nodules at control (0.45g) was significantly higher than those of applications of 480 µM Al (0.30g), 960 µM Al (0.26g) and 750 µM Al (0.22g). Mean of fresh weights of nodules at application 480 µM Al was significantly higher than mean at 750 µM Al and at 960 µM Al, respectively. Considerably, means of GAZZELE genotype (0.36) and TGX (0.36) were significantly higher than that of NAMSOI (0.20).

3.2.2. Dry weights of nodules

Table 2 shows dry weights of nodules of the three soy bean genotypes. There were no significant differences in nodule dry weights between GAZZELLE, NAMSOI and TGX at the eight treatments (Table 2). Generally, mean at treatment 5 (T5) was significantly different than the other seven treatments means.

The interaction between Rhizobium inoculation and aluminium applications showed that there was a significant difference ($p = .0118$) on dry weights of nodules. The mean dry weight of nodules of the control (0.13g) was significantly higher than means at applications 480 µM Al (0.05g), 750 µM Al (0.03g) and 960 µM Al (0.03g). Meanwhile, mean of dry weight of TGX (0.08) was significantly higher than GAZZELE (0.05) and NAMSOI (0.04) genotypes, respectively.

3.2.3 Number of Pink nodules

Table 3 shows number of pink nodules of the three soy bean genotypes. The mean number of pink nodules of TGX was significantly higher compared to those of NAMSOI and GAZZELLE at treatments T1, T2, T5 and T7 (Table 3). Generally, mean of pink nodules at treatment T1 was significantly higher than the means of the other seven treatments.

Number of pink nodules showed that there was a significant interaction between and soy bean genotypes ($p = .0113$). Mean of numbers of pink nodules at control (11.28) was significantly higher than means at 750 µM Al (6.78), 480 µM Al (6.33) and 960 µM Al (5.67), respectively. Similarly, mean pink nodules of *Rhizobium*-inoculated (9.19) was significantly higher than that of non-inoculated (5.83) genotypes.

3.2.4. Number of Brown nodules

Table 3 shows number of brown nodules of the three soy bean genotypes. There was no statistical difference in mean number of brown nodules of NAMSOI, GAZZELLE and TGX for the eight treatments (Table 3). Generally, mean at treatment 6 was significantly lower than the seven treatment means.

There was a significant interaction ($p = .0115$) between the effects of aluminium application and Rhizobium. The mean number of brown nodules at control (9.17) was significantly higher than that at 960 µM Al (8.11), 750 µM Al (5.94), and 480 µM Al (5.56) Al applications, respectively. Similarly, mean at 960 µM Al application was significantly higher than applications at 750 µM Al and 480 µM Al. The mean of number of brown nodules of Rhizobium-inoculated (8.11g) plants was significantly higher than that of non-inoculated (6.28g).

3.2.5. Number of green nodules

Table 4 shows number of green nodules of soy bean genotypes. The mean number of green nodules for genotype NAMSOI was significantly higher than those of GAZZELLE and TGX (Table 4) at treatment 4 (T4).

Table 2. Response of Fresh weights and dry weights of nodules of three soybean genotypes to to Al application and *Rhizobium* inoculation. Values are means of three replicates ±SEs. Means with the same latter in the row are not significantly different.

Treatments of Al application and *Rhizobium* inoculation	Fresh weights (g) of nodules				Dry weights (g) of nodules			
	GZZL	NMSI	TGX	Tukey`s grouping for Treatments	GZZL	NMSI	TGX	Tukey`s grouping for Treatments
T1	0.43±00.07ab	0.13±0.05b	0.68±0.26a	**0.68±0.6ab**	0.04±0.03a	0.09±0.05a	0.14±0.05a	**0.09±0.03ab**
T2	0.28±0.05a	0.18±0.07a	0.43±0.29a	**0.43±0.29ab**	0.04±0.02a	0.06±0.03a	0.1±0.06a	**0.06±0.02b**
T3	0.29±0.01a	0.08±0.03a	0.13±0.04a	**0.13±0.0.04b**	0.01±0.00a	0.05±0.02a	0.06±0.01a	**0.04±0.01b**
T4	0.64±0.14a	0.08±0.00b	0.27±0.05b	**0.33±0.09ab**	0.01±0.00b	0.03±0.01b	1.03±0.01a	**0.02±0.0b**
T5	0.52±0.04a	0.41±0.07a	0.57±0.2a	**0.5±0.07a**	0.15±0.02a	0.09±0.0.04a	0.24±0.07a	**0.16±0.03a**
T6	0.33±0.001a	0.25±0.02a	0.33±0.23a	**0.31±0.07ab**	0.05±0.04a	0.01±0.00a	0.02±0.01a	**0.03±0.01b**
T7	0.24±0.05a	0.29±0.003a	0.277±0.05a	**0.27±0.02ab**	0.03±0.02a	0.01±0.003a	0.06±0.01a	**0.02±0,01b**
T8	0.2±0.00a	0.19±0.01a	0.21±0.06a	**0.2±0.02ab**	0.03±0.02a	0.01±0.00a	0.06±.0.02a	**0.03±0.01b**
Tukey`s grouping for genotypes	**0.36±0.04a**	**0.2±0.03b**	**0.36±0.06a**		**0.05±0.01b**	**0.04±0.01b**	**0.08±0.02a**	

Treatments comprised of Control (T1) {Water*Inoculated}, T2 {480μM Al*Inoculated}, T3 {750μM Al*Inoculated}, T4 {960μM Al*Inoculated}, T5 {Control (Water)}, T6 {480μM Al}, T7 {750μM Al} and T8 {960μM Al}.

Table 3. Response of Number of pink and brown nodules of three soybean genotypes to Al application and *Rhizobium* inoculation Values are means of three replicates ±SEs. Means with the same latter in row are not significantly different.

Treatments of Al application and *Rhizobium* inoculation	Number of pink nodules				Number of brown nodules			
	GZZL	NMSI	TGX	Tukey`s grouping for	GZZL	NMSI	TGX	Tukey`s grouping for
T1	13±.2.52ab	7.67±1.76b	17±1.73a	**12.56±1.73a**	10.33±1.33a	9.0.±0.00a	11.00±2.00a	**10.11±0.75a**
T2	8.3±0.33ab	6.67±1.21b	11.67±1.45a	**8.89±0.92ab**	8.33±0.33a	8.67±0.88a	7.33±0.33a	**8.11±0.351ab**
T3	7.65±0.67a	6.33±1.2a	8.33±0.33a	**7.44±0.5bc**	6±0.58a	7.67±20.88a	7.67±0.88a	**7.11±0.48abc**
T4	8.33±0.55a	6.67±1.2a	8.67±0.33a	**7.89±0.48abc**	7±1.15a	7.67±40.67a	6.67±0.33a	**7.11±0.42abc**
T5	11.67±1.3ab	5.33±1.76b	13±2.52a	**10±1.51ab**	11.33±3.38a	6.33±2.33a	7.00±23.51a	**8.22±1.75ab**
T6	6±1.53a	2±0.58ab	3.33±0.88b	**3.78±0.8c**	5.33±2.91a	2±0.58a	1.67±0.88a	**3.00±1.07c**
T7	3.67±1.45b	3.33±0.33b	11.33±2.96a	**6.11±1.62bc**	4±1.33a	6.33±2.33a	4.00±1.53a	**4.78±0.99bc**
T8	3.33±0.33a	3.3.±0.9a	3.67±2.96a	**3.44±0.5c**	7.67±0.58a	12±3.79a	7.67±0.88a	**9.11±1.35a**
Tukey`s grouping for genotypes	**7.75±0.78b**	**5.17±0.53c**	**9.63±1.04a**		**7.5±0.73a**	**7.46±0.78a**	**6.63±0.72a**	

Treatments comprised of Control (T1) {Water*Inoculated}, T2 {480μM Al*Inoculated}, T3 {750μM Al*Inoculated}, T4 {960μM Al*Inoculated}, T5 {Control (Water)}, T6 {480μM Al}, T7 {750μM Al} and T8 {960μM Al}.

Table 4. Response of Number of green nodules of three soybean genotypes to Al application and *Rhizobium* inoculation. Values are means of three replicates ±SEs. Means with the same latter in row are not significantly different.

Treatments of Al application and *Rhizobium* inoculation	Number green nodules			Tukey`s grouping for Treatments
	GZZL	NMSOI	TGX	
T1	1.67±0.33b	4.67±0.88a	2.00±0.58a	**2.78±0.57b**
T2	4.67±0.00a	5.00±1.73a	2.67±1.20a	**4.11±0.75ab**
T3	6.00±2.08a	6.33±3.53a	6.67±2.03a	**6.33±1.32ab**
T4	1.00±0.58b	6.67±1.45a	1.00±0.58b	**2.89±1.06b**
T5	4.67±1.45a	5.00±1.53a	3.33±1.76a	**4.33±0.83ab**
T6	5.00±0.58a	5.67±0.33a	5.00±2.00a	**5.22±0.62ab**
T7	6.33±1.76a	4.33±0.33a	5.00±1.00a	**5.33±0.66ab**
T8	8.33±0.6a	6.33±1.45a	7.66±2.33a	**7.44±0.91a**
Tukey`s grouping for genotypes	**4.71±0.6a**	**5.5±0.53a**	**4.17±0.65a**	

Treatments comprised of Control (T1) {Water*Inoculated}, T2 {480μM Al*Inoculated}, T3 {750μM Al*Inoculated}, T4 {960μM Al*Inoculated}, T5 {Control (Water)}, T6 {480μM Al}, T7 {750μM Al} and T8 {960μM Al}.

There was a significant interaction ($p = .0215$) between the effects of aluminium application and Rhizobium for green nodules. Tukey's HSD test for mean of green nodules at *Rhizobium*-inoculated (4.03) was significantly lower than non-inoculated (5.56) plants.

3.3. Plant yield

3.3.1. Plant fresh weights above ground

Table 5 shows plant fresh weight above ground of soybean genotypes. Inoculation increased fresh weights above ground while increased Al generally decreased fresh weights above ground. The mean yield of NAMSOI was significantly higher than GAZZELLE and TGX at treatments T2, T3, T5, T6 and T8 (Table 5), respectively.

The mean of fresh weights above ground of the control (17.99g) was significantly higher than the means at both 480 μM Al (14.24g), 750 μM Al (13.98g) and 960 μM Al (11.54g). However, means of fresh weight above ground of Rhizobium-inoculated genotypes (15.31g) was significantly higher than the mean of the non-inoculated (13.57g) plants. In consideration to genotypes, Mean number of fresh weight above ground for NAMSOI (19.78g) soybean genotypes inoculated with Rhizobium and treated with aluminium was significantly higher than those of GAZZELE (13.32g) and TGX (10.33g), respectively.

3.3.2. Plant dry weights above ground

Table 5 shows plant dry weights above ground of soybean genotypes. There was a general increase in dry weights above ground on inoculation while Al increase generally decreased dry weights above ground. The mean of above ground dry weight of NAMSOI was significantly higher than those of GAZZELLE and TGX at treatment 5 (Table 5).

A significant interaction ($p = .0237$) was observed between the effects of aluminium application and Rhizobium on plant dry weights above ground. The mean dry weights of control (5.78g) was significantly higher than those of applications 480 μM Al (4.16g), 750 μM Al (3.78g), and 960 μM Al (3.77g). However, mean above ground weight of Rhizobium-inoculated plants (4.67g) was significantly higher than the mean of non-inoculated (4.07g). Mean above ground dry weight of NAMSOI (5.85) was significantly higher than GAZZELE (3.76) and TGX (3.51), respectively.

3.3.3. Plant fresh weights below ground

Table 6 shows plant fresh weight below ground of soybean genotypes. Al decrease generally decreased fresh weights below ground which were also increased on Rhizobium inoculation. The below ground mean fresh weight of NAMSOI was significantly higher than GAZZELLE and TGX, respectively at treatment 8 (Table 6). The below ground mean fresh weight of NAMSOI was significantly higher than TGX and GAZZELLE at treatment 6, respectively.

Plant fresh weights below ground showed that there was a significant difference ($p = .0044$) in soybean genotypes. Tukey's HSD test for below ground fresh weight showed that genotypes means of NAMSOI (1.58) was significant higher than that of TGX (1.23) and GAZZELE (0.63), respectively.

3.3.4. Plant dry weights below ground

Table 6 shows plant dry weight below ground of soybean genotypes. The table shows that there was a general decrease in dry weights below ground on Al application. Dry weights were also increased on Rhizobium inoculation. The mean below ground dry weight for NAMSOI was significantly higher than that of GAZZELLE and TGX at treatments 4 and 7, respectively (Table 6).

Table 5. Response of Fresh weights above ground and Dry weights above ground of three soybean genotypes to Al application and *Rhizobium* inoculation. Values are means of three replicates ±SEs. Means with the same latter in row are not significantly different.

Treatments of Al application and *Rhizobium* inoculation	Fresh weights (g) above ground				Dry weights (g) above ground			
	GZZZ	NMSI	TGX	Tukey`s grouping for Treatments	GZZL	NMSI	TGX	Tukey`s grouping for Treatments
T1	20.4±0.4a	23.61±3.99a	13.27±3.45a	**19.09±2.31a**	4.37±0.1ab	6.74±1.35a	4.7±0.22a	**5.27±0.54ab**
T2	9.87±0.92b	19.73±2.28a	13.8±1.43b	**14.47±1.65a**	2.83±0.8a	6.11±0.92a	4.73±0.11a	**5.08±0.38ab**
T3	15.72±1.16b	20.14±1.17a	9.44±1.85c	**15.15±1.62a**	4.51±0.46a	5.62±0.31a	3.03±0.25b	**4.39±0.41ab**
T4	13.78±0.38a	14.39±1.8a	9.57±0.66a	**12.54±1.08a**	3.58±0.28a	4.58±0.72a	3.68±0.61a	**3.95±0.33ab**
T5	14.76±1.57b	22.61±1.64a	13.61±1.99b	**16.89±2.1.62a**	4.98±0.3ab	9.17±2.11a	4.72±0.28b	**6.29±0.95a**
T6	9.72±1.72b	27.87±7.74a	4.46±0.69b	**14.01±4.23a**	4.4±0.7a	5.35±0.18a	1.52±0.27a	**3.23±0.6b**
T7	10.37±1.64a	17.5±5.08a	11.13±2.06a	**12.83±1.96a**	2.61±0.05a	4.22±1.97a	2.72±0.38a	**4.18±0.63b**
T8	11.12±2.56b	13.19.±3.6a	7.33±1.93b	**10.55±1.62a**	2.81±0.38a	5.01±1.4a	2.98±0.68a	**3.59±0.58b**
Tukey`s grouping for genotypes	**13.22±0.93b**	**19.78±1.51a**	**10.33±0.81b**		**3.76±0.21b**	**5.85±10.49a**	**3.51±10.26b**	

Treatments comprised of Control (T1) {Water*Inoculated}, T2 {480μM Al*Inoculated}, T3 {750μM Al*Inoculated}, T4 {960μM Al*Inoculated}, T5 {Control (Water)}, T6 {480μM Al}, T7 {750μM Al} and T8 {960μM Al}.

Table 6. Response of Fresh weights below ground and Dry weights below ground of three soybean genotypes to Al application and *Rhizobium* inoculation. Values are means of three replicates ±SEs. Means with the same latter in row are not significantly different.

Treatments of Al application and *Rhizobium* inoculation	Fresh weights (g) below ground				Dry weights (g) below ground			
	GZZL	NMSI	TGX	Tukey`s grouping for Treatments	GZZL	NMSI	TGX	Tukey`s grouping for Treatments
T1	0.54±0.11a	1.42±0.29a	2.67±1.62a	**1.55±0.57a**	0.28±0.8a	1.21±0.72a	0.54±0.13a	**0.68±0.25a**
T2	0.52±0.09a	1.74±0.48a	1.49±0.12ab	**1.25±0.24a**	0.22±0.02a	0.81±0.31a	0.78±0.61a	**0.6±0.23a**
T3	0.51±0.06b	1.3±0.12a	0.84±1.0.26ab	**0.89±0.14a**	0.27±0.01a	0.62±0.26a	0.30±0.07a	**0.4±0.1a**
T4	1.25±0.34a	1.61±1.15a	1.29±0.21a	**1.38±0.26a**	0.22±0.03b	0.92±0.31a	0.35±0.08ab	**0.5±0.14a**
T5	0.54±0.17a	1.39±0.64a	1.05±0.43a	**0.99±0.26a**	0.64±0.19a	0.71±0.11a	0.76±0.25a	**0.7±0.1a**
T6	0.4±0.05b	2.77±1.37a	0.29±0.06b	**1.15±0.57a**	0.16±0.02b	0.33±0.08ab	0.46±0.05a	**0.32±0.06a**
T7	0.61±0.09a	1.01±0.19a	1.29±0.53a	**0.97±0.19a**	0.21±0.02c	0.46±0.02a	0.32±0.04b	**0.33±0.04a**
T8	0.49±30.08b	1.4.±0.02a	0.88±0.53b	**0.92±0.38a**	0.18±0.03a	0.45±0.11a	0.31±0.14a	**0.31±0.07a**
Tukey`s grouping for genotypes	**0.61±0.07b**	**1.58±0.23ab**	**1.23±0.24a**		**0.27±0.04b**	**0.69±10.11a**	**0.48±0.08ab**	

Treatments comprised of Control (T1) {Water*Inoculated}, T2 {480μM Al*Inoculated}, T3 {750μM Al*Inoculated}, T4 {960μM Al*Inoculated}, T5 {Control (Water)}, T6 {480μM Al}, T7 {750μM Al} and T8 {960μM Al}.

There was a significant difference (p = .0044) in the effects of genotypes on plant dry weights below ground (Table 6). Mean dry weight below ground of Rhizobium-inoculated (0.54g) was significantly higher than that of non-inoculated (0.42g) plants. However, the means dry weight below ground for NAMSOI (0.69) was significantly higher than those of TGX (0.48) and GAZZELE (0.27), respectively.

3.3.5. Total plant fresh weights

Table 7 shows total plant fresh of soybean genotypes. Al decreased total plant fresh weights which were also found to be lower under non-inoculation. The mean of total plant fresh weight for NAMSOI was significantly higher than those of GAZZELLE and TGX at treatments T2, T3, T5 and T6 (Table 7), respectively.

Mean of total plant fresh weights at control (19.71g) was significantly higher than those at 480 µM Al (15.74g), 750 µM Al (15.14g) or 960 µM Al (12.96g) applications respectively. Mean at treatment 960 µM Al was significantly lower than those either at 750 µM Al and 480 µM Al. Mean total fresh weights of NAMSOI (21.56) was significantly higher than those of GAZZELE (14.19) and TGX (11.91), respectively.

3.3.6. Total plant dry weights

Table 7 shows total plant dry weights of soybean genotypes. Total plant dry weights decreased on Al applications which were also found to be higher on inoculation. The mean total plant dry weight of NAMSOI was significantly higher compared to those of GAZZELLE and TGX at treatments T2, T3, and T6 (Table 7), respectively. Generally mean plant dry weight at treatment T1 was significantly higher than seven treatments.

Total plant dry weights showed that there was a significant interaction (p = .0232) between the effects of aluminium application and *Rhizobium* inoculation. The mean of total dry weight of control treatment (14.69g) was significantly higher than those of applications 480 µM Al (10.28g), 750 µM Al (9.61g), and 960 µM Al (9.27g), respectively. The mean total dry weight of NAMSOI (13.36g) soybean genotypes inoculated with *Rhizobium* and treated with aluminium was significantly higher than those of TGX (10.15g) and GAZZELE (9.37g), respectively.

3.3.7. Number of seeds per plant

Table 8 shows number of seeds per plant of soybean genotypes. In general, Al application and *Rhizobium* inoculation decreased the number of seeds per plant. It shows that NAMSOI had a significantly higher number of seeds than those for GAZZELLE and TGX at treatments T2, T4 and T6, respectively. Generally, mean number of seeds at treatment T1 was significantly higher than those of seven treatments.

Table 7. Response of Total plant fresh weights and Total plant dry weights of three soybean genotypes to Al application and *Rhizobium* inoculation. Values are means of three replicates ±SEs. Means with the same latter in row are not significantly different.

Treatments of Al application and *Rhizobium* inoculation	Total plant fresh weights (g)				Total plant dry weights (g)			
	GZZL	NMSI	TGX	Tukey`s grouping for Treatments	GZZL	NMSI	TGX	Tukey`s grouping for Treatments
T1	21.37±4.17a	25.16±4.21a	16.62.5±3.48a	**21.05±2.81a**	12.84±1.45b	18.62±1.57b	33.33±6.98a	**15.02±1.24a**
T2	10.67±0.97b	21.65±1.81a	15.72±1.76b	**16.01±1.77a**	10.43±1.17b	15.57±0.64a	17.67±1.45ab	**12.97±0.84abc**
T3	16.54±1.68b	21.53±1.19a	10.54±0.85c	**16.2±1.68a**	8.89±1.25b	12.44±0.99ab	20.33±3.53a	**9.88±0.85cd**
T4	15.67±0.6a	16.08±2.58a	11.00±2.28a	**14.25±1.72a**	9.89±1.16b	10.79±1.19ab	19.67±3.28a	**10.36±0.54bcd**
T5	15.6±1.73b	24.1±2.18a	15.23±1.8b	**18.38±1.29a**	11.99±1.32a	18.05±3.02ab	30.33±2.89a	**14.36±1.39ab**
T6	10.45±1.76b	30.89±9.094a	5.08±0.89b	**15.47±4.76a**	6.65±1.42b	10.29±0.84ab	19±3.21a	**7.59±0.89d**
T7	11.21±1.26a	18.3±5.28a	12.69±2.59a	**14.07±2.07a**	7.54±0.54b	10.95±2.87ab	21.33±3.38a	**9.34±0.99cd**
T8	11.81±2.4a	14.78.±2.18a	8.42±1.98a	**11.07±1.66a**	9.37±0.58b	10.25±2.74ab	17.33±0.88a	**8.17±1.05d**
Tukey`s groupin for genotypes	**14.19±0.97b**	**21.56±1.66a**	**11.91±0.99b**		**9.37±0.58a**	**13.36±0.9b**	**22.38±1.58a**	

Treatments comprised of Control (T1) {Water*Inoculated}, T2 {480µM Al*Inoculated}, T3 {750µM Al*Inoculated}, T4 {960µM Al*Inoculated}, T5 {Control (Water)}, T6 {480µM Al}, T7 {750µM Al} and T8 {960µM Al}.

Table 8. Response of Number of seeds per plant and Dry weights (g) of 100 seeds of three soybean genotypes to Al application and *Rhizobium* inoculation. Values are means of three replicates ±SEs. Means with the same latter are not significantly different.

Treatments of Al application and *Rhizobium* inoculation	Number of seeds per plant				Dry weights (g) of 100 seeds			
	GZZL	NMSI	TGX	Tukey`s grouping for Treatments	GZZL	NMSI	TGX	Tukey`s grouping for Treatments
T1	31.33±5.61a	48.68±5.7a	33.33±6.98a	**37.78±4.11a**	5.42±1.72a	6.67±1.96a	4.77±1.132a	**5.62±0.89a**
T2	22.33±2.08ab	29.33±2.67a	17.67±1.45b	**23.11±2.01bc**	2.53±0.54a	2.93±0.11a	1.61±0.20a	**2.51±0.28b**
T3	21.33±2.4a	28.67±2.19a	20.33±3.53a	**23.44±1.89bc**	2.48±0.96a	2.57±0.15a	1.22±0.64a	**2.3±0.36b**
T4	21.67±0.88ab	33±4.73a	19.67±3.28b	**24.78±2.67bc**	3.88±1.72a	1.45±0.08b	2.15±0.0.22b	**2.48±0.4b**
T5	30.67±2.73a	35±0.58a	30.33±2.89a	**3±1.27ab**	3.67±0.5a	3.3±0.17a	2.59±10.15a	**3.15±0.22b**
T6	23±2.08ab	22.67±3.53a	19±3.21**b**	**21.56±1.63c**	3.07±0.57a	2.85±00.3a	1.06±0.6a	**2.15±0.38b**
T7	21.33±2.4a	27.33±1.86a	21.33±3.38a	**24±1.61bc**	1.68±0.44b	2.67±0.13a	1.83±0.06b	**1.86±0.0.25b**
T8	20±0.88a	19.67±1.2a	17.33±0.88a	**23.11±1.49c**	1.9±0.5a	2.11.±0.69a	0.99±0.22a	**1.67±0.33b**
Tukey`s grouping for genotypes	**24.21±1.27b**	**30.54±1.98a**	**22.38±1.58b**		**3.07±0.34a**	**3.05±0.38a**	**19.15±0.57b**	

Treatments comprised of Control (T1) {Water*Inoculated}, T2 {480µM Al*Inoculated}, T3 {750µM Al*Inoculated}, T4 {960µM Al*Inoculated}, T5 {Control (Water)}, T6 {480µM Al}, T7 {750µM Al} and T8 {960µM Al}.

There was a significant interaction ($p = .0232$) between the effects of aluminium application and soybean genotypes on the number of seeds per plants (Table 8). Tukey`s HSD test showed that mean number of seeds per plant at control (34.89) was significantly higher than mean of 750 μM Al (23.72), 480 μM Al (22.33) and 960 μM Al (21.89) applications, respectively. Similarly, mean number of seeds per plant at *Rhizobium*-inoculated (27.28) was significantly higher than that of non-inoculated (24.14) plants. The mean of number of seeds per plant of NAMSOI (30.54) soybean genotypes inoculated with *Rhizobium* and treated aluminium was significantly higher than those of GAZZELE (24.21) and TGX (22.38), respectively.

3.3.8. Dry weights of 100 seeds

Table 8 shows weights of 100 seeds of soybean genotypes. Al application and *Rhizobium* inoculation had a general decrease in weights of 100 seeds. The mean weight of 100 seeds of soybean genotype GAZZELLE was significantly higher than the means for NAMSOI and TGX genotypes at treatment T4.

The mean of genotype for NAMSOI was also significantly higher than those of GAZZELLE and TGX at treatment T7 (Table 8). Generally, mean of weights of 100 seeds at treatment T1 was significantly higher than the other seven treatment means.
The mean of dry weights of 100 seeds at control (4.39g) was significantly higher than those at 480 μM Al (2.33g), 750 μM Al (2.08g) and 960 μM Al (2.07g) applications, respectively. The mean for USDA-inoculated (3.23g) plants was also significantly higher than mean of non-inoculated (2.21g). The mean dry weights of 100 seeds of GAZZELE (3.07) and NAMSOI (3.05) soybean genotypes inoculated with *Rhizobium* and aluminium were significantly higher than that of genotype TGX (2.03), respectively.

3.3.9. Total weights of husks and seeds

Table 9 shows total weights of husks and seeds of soybean genotypes. Total weights of husks and seeds decreased on Al application and increased in *Rhizobium* inoculation. The mean weight of husks and seeds at treatment T1 was significantly higher than that of other seven treatments (Table 9).
There was a significant interaction ($p = .0109$) between the effects of aluminium application and *Rhizobium* inoculation on total weights of husks and seeds. The mean of husks and seeds of control (8.09g) was significantly higher than that mean applications 480 μM Al (5.62g), 750 μM Al (5.43g) and 960 μM Al (4.06g), respectively. Similarly, the mean for *Rhizobium*-inoculated (6.79g) soybean plants was significantly higher (Appendix 3; Table 26) than the non-inoculated (5.31g). The mean of NAMSOI (6.78) genotype was also significantly higher than those of TGX (6.08) and GAZZELE (5.29), respectively.

Table 9. Response of Total weights of husks and seeds of three soybean genotypes to Al application and *Rhizobium* inoculation. Values are means of three replicates ±SEs. Means with the same latter in row are not significantly different.

Treatments of Al application and *Rhizobium* inoculation	Total weights of husks and seeds			Tukey`s grouping for Treatments
	GZZL	NMSI	TGX	
T1	8.15±1.33a	10.58±0.59a	8.21±2.08a	**8.98±0.83a**
T2	3.61±1.11a	8.54±0.48a	7.37±0.95a	**7.23±0.6ab**
T3	4.09±0.81a	6.15±0.97a	4.92±3.53a	**5.05±10.57bc**
T4	6.08±0.95a	5.26±0.98a	6.35±0.37a	**5.9±20.44bc**
T5	6.21±0.87a	8.1±1.01a	7.3±20.98a	**7.2±0.55ab**
T6	5.29±0.75a	4.61±0.76a	3.83±0.76a	**4.02±0.41c**
T7	4.64±0.6a	6.26±1.11a	6.47±0.75a	**5.8±0.48bc**
T8	3.73±1.08a	4.76±1.39a	4.18±0.69a	**4.22±0.57c**
Tukey`s grouping for genotypes	**5.29±0.41b**	**6.78±0.58a**	**6.08±0.44ab**	

Treatments comprised of Control (T1) {Water*Inoculated}, T2 {480μM Al*Inoculated}, T3 {750μM Al*Inoculated}, T4 {960μM Al*Inoculated}, T5 {Control (Water)}, T6 {480μM Al}, T7 {750μM Al} and T8 {960μM Al}.

4. Discussion

4.1. Effects of aluminium application and *Rhizobium* inoculation on Al concentrations

Differences in accumulation of Al were noted in the three genotypes where TGX accumulated more than NAMSOI and GAZZELLE in many treatments. Highest aluminium concentrations (Fig. 8) in all the soy bean genotypes were observed at treatment 8 (T8). It was also observed that, lowest treatment (T1) resulted to lowest mean aluminium concentration among all the genotypes. Soy bean sensitivity to aluminium stress is not a unique occurrence. It was found in other plants, for instance in blueberry by [32], in *Thinopyrum bessarabicum* [2], in cowpea by [8] and in common bean by [40]. Overally, Al increase in concentration in substrate resulted in higher Al content in soy bean plants.

Aluminium is often transported from root cells into other plant cells [35]. In this regard, the plant develops aluminium stress causing low yields in soy bean and, therefore, massive losses [16]. This study established that, high aluminium concentrations in plant tissues inhibited soy bean`s morphological and physiological development which is in agreement with previous studies by [35]. However, it is known that soy bean genotype just like any other legume accumulate aluminium at different rates [40]. This was observed in this study where TGX accumulated significantly more Al than GAZZELLE and NAMSOI. The availability of other nutrients in the soil play a crucial role in determining the amount of Al absorption [42]. For instance, there might have been high calcium (Table 1) concentration in the soil that stimulated the soybean cells to accumulate more Al [45]. Some researches [31]; [8]; [40] have demonstrated that Al stimulates uptake of ions like iron, manganese and zinc, a phenomenon that led to limited uptake of Al in control plants.

A difference that was not significant was found when inoculated soy bean plants were compared to non-inoculated plants under this study with non-inoculated plants accumulating more Al in leaves. A symbiotic co-existence between legumes and Rhizobium increases Al resistance in leguminous plants [27]. Therefore, Rhizobium treated soy bean plants accumulate high Al content, which might be less toxic to them. Similar results were noted when Alfalfa (*Medicago sativa*) grew robustly under inoculation with *Sinorhizobium melitoti* regardless of the high Al stress [44].

4.2. Effects Al application and *Rhizobium* inoculation on nodulation and yield of soy bean

Genotypes TGX and GAZZELLE were found to respond by reducing a number of days to harvest maturity for Al treatments at T4 and T6, respectively. However, GAZZELLE is early maturing genotype [19] and it indicated better results. TGX is known to be late maturing genotype and may be affected on the other hand by Al in acid soils under inoculation, a phenomenon that may have caused premature browning of pods in the genotype TGX. [4] found that promiscuous non nodulated TGX soy beans genotype in Ghana's farming systems had this effect of premature browning of pods.

Nodule dry weights (Table 2) and total plant dry weights (Table 7) were both generally significantly higher in TGX genotype under Al and Rhizobium inoculation compared to GAZZELE genotype, respectively. [33], found similar differences for nodule number and nodule weights within non-promiscuous soy beans. According to [7] this is influenced by genotypic differences and environmental interplay. Large size of nodules and increased number of nodules through the infection threads of inoculated TGX genotype may have caused this significantly higher value in total dry weights [33]. Plant growth promoting rhizobacteria (PGPR) traits may have also increased TGX nodule induction and function under Al stress of acid soils [5]; [24]. Infection thread elongation, calcium spiking and proliferation of Rhizobium may have been inhibited in GAZZELLE leading to root hair deformation and therefore reduced dry weight of nodules [15].

There was a decrease in dry weight of root nodules which could have been as a result of Al application. Similarly, in common beans, [34] found there was a decrease in root length, just after a short period of Al exposure. Dry weights of nodules at non-inoculated were not statistically different from the *Bradyrhizobium japonicum* inoculated genotypes. This outcome could be because Al may have interfered with Fe transport systems therefore limiting bacterial activity to Fe^{2+} capturing [42]. Owing to those reasons, dry weights of nodules are reduced due to lack of Fe^{2+} that is required for nitrogenase activity within plants. *Bradyrhizobium* ssp. is known to be very sensitive to Al [34]. Therefore Al affected enzymatic activities for nitrate and nitrite reduction.

TGX had more pink nodules at most of eight treatments compared to GAZZELLE and NAMSOI, respectively. According to [23], colour as a nodulation character is controlled by bacterial specific genes. In this regard, Rhizobium may have occurred naturally in the soils used in this study which were not specific to soy bean but highly competitive relative to introduced Bradyrhizobium japonicum [38]. As a consequence, these infected soy bean, but nodules formed may not be able to fix nitrogen. Such nodules are often green and said to be inactive as compared to active pink nodules. This trend might have been pronounced in genotype GAZZELLE that possessed the green nodules compared to NAMSOI and TGX. This effect was more pronounced in NAMSOI at Al treatment T4. [52] and [13] who studied legume *Acmispon strigosus* and soy bean respectively also found that Anz11 and Cla10 genotypes of *Acmispon strigosus* and PI 438133B genotypes of soy bean had more of inactive nodules that could not fix nitrogen. Nod genes are specific to different stages of nodule formation [29]. Thus first, legume plant interacting with Rhizobium cause the release of high complex chemical by the root cells into the soil, which then encourage bacterial growth around the root`s rhizosphere and control nodulation [23]. Therefore, considering the two colors of active nodules, high amounts of compounds in cell walls of the bacteria and the root surface might have helped Rhizobium to identify and infect the correct host plant and attach to root hairs causing differences in Bradyrhizobium japonicum inoculated plants [49]. For instance, flavonoids may have been secreted by legume plant roots to activate nod genes in the bacteria cell hence good nodulation [23].

This study revealed that total dry weights above and below ground of soy bean plants increased on inoculation. NAMSOI exhibited significantly higher weights than GAZZELLE and TGX, respectively, which was repeated under inoculations at various Al treatments. According to [18], NAMSOI had a significantly higher anthocyanin concentration at T5 that may have increased water and nutrient uptake, and high photosynthesis hence faster growth under aluminium compared to GAZZELLE and TGX. NAMSOI and GAZZELLE genotypes also performed better for the number of seeds per pod, weight of 100 seeds and total weight of husks plus seeds. Similar results were also found by [34] who had a general review study on legumes and concluded that such plants did not accumulate most of it food reserve for higher biomass formation at the expense of seed formation

Rhizobium inoculated plants also performed better than those that were non-inoculated in yield response parameters. Rhizobium inoculated seeds were found yielding high number of seeds per pod and number of pods per plant hence a high grain yield in comparison to the non-inoculated [50]. The fact that NAMSOI performed better in terms of pod number implies that they may have higher nitrogen absorption capacity that led to a direct dry weight accumulation in the plant parts including the pods and also seed yield when the seeds were inoculated with *Bradyrhizobium japonicum*. Control plants were found to perform better for these yield parameters than aluminium treated ones. This was probably because aluminium suppresses nutrient (phosphates and nitrogen) uptake by forming complexes that limits most of the nutrients to be absorbed [42]. These results are at variance with those of [40] who inoculated soy bean with *Bradyrhizobium japonicum* and found no significant differences in pod number, pod weight and yield. This was attributed to the fact that, the presence of a healthy rhizosphere in inoculated controls meant production of hormones, and phosphate solubilizing microorganisms thus improving nutrient and water uptake as previously established by [50].

5. Conclusion and Recommendations

This study established that aluminium accumulation was toxic to soy bean plants. Nonetheless, the accumulation of Al by soy bean genotypes varied whereby genotypes GAZZELLE and NAMSOI accumulated less Al in leaves in most of the eight treatments. In these genotypes, less Al was transported from roots into other tissues causing reduced Al stress. Future studies should concentrate on Al partitioning in different organs of plants grown under Al. This may help explain the tolerance mechanism of these plants to Al stress. There is need to carry out research to determine different mechanisms of minerals nutrients absorption under Al stress.

The application of Al to soy bean genotypes led to a reduction in their growth. Aluminium inhibited root development as evidenced by reduced root dry weight. To some extent, Rhizobium inoculation ameliorated the negative effects of aluminium on soybean plants. Rhizobium contributed to enhanced atmospheric nitrogen fixation leading to improved plant growth and development**.** By inoculating soy bean genotype with Bradyrhizobium japonicum, plants potentially benefited from the nitrogen fixation capability of the bacteria, which compensated for the overall growth of soybean plants under unfavourable conditions of aluminium stress. Future studies should concentrate on determining the effects of Al on nitrogen fixation and nitrogenase activity of soy beans as these have roles in reducing nodulation and yields under Al. This will help us understand the mechanisms involved in Al stress in legumes. Similar research should be extended to Rhizobium inoculated plants which are treated with Al.

Acknowledgements

We thank the Consortium of International Agricultural Center (CGIAR) station at Maseno for providing the TGx 1871-12E; NAMSOI and GAZELLE genotype seeds.

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