

# Determination of appropriate level of aluminum activity in hydroponics for the screening of tropically adapted soybean varieties

Ojo, G. O. S.<sup>1\*</sup> and L. L. Bello<sup>2</sup>

<sup>1</sup> Department of Crop Production, University of Agriculture, P.M.B. 2373, Makurdi, Nigeria. <sup>2</sup> Department of Plant Breeding and Seed Science, University of Agriculture, P.M.B. 2373, Makurdi, Nigeria. Corresponding author email: \*gosojo2001@yahoo.com

Keywords: aluminium activity, 3D, aluminium stress tolerance, hydroponics screening

# 1 SUMMARY

A hydroponics experiment was conducted at the Crop Science Laboratory of the University of Agriculture, Makurdi, Nigeria, in 2002 with the objective of determining the appropriate levels of aluminium for the screening of tropically adapted genotypes of soybean. Fifteen soybean genotypes constituted the main plots while ten levels of aluminium activity constituted the subplots in a split-plot design. The experiment was replicated three times. The soybean seedlings were germinated for 4 days and transferred to nutrient solutions containing the various levels of aluminium. The seedlings grew for 3 days (3D) in the hydroponics, bringing their total age to 7 days. Data were taken for primary root length, shoot length, root dry weight and shoot dry weight of plants grown at 8 levels of aluminium activity(0, 5, 50, 100, 150, 200, 250 and  $300\mu$ MAl<sup>3+</sup>). No data was taken for the 350 and  $400\mu$ MAl<sup>3+</sup> levels of aluminium activity due to death of seedlings grown at these two levels. Highly significant Al, genotype, and first order interaction effects were observed. Four levels of Al activity (0, 5, 50 and  $300\mu$ MAl<sup>3+</sup>) were selected and recommended as appropriate for the screening of tropically adapted genotypes of soybean.

# 2 INTRODUCTION

Aluminium is the third most abundant element in the earth crust (Kochian, 1995) and considered one of the most limiting factors for plant productivity in acidic soils (Houde and Diallo, 2008; Tani et al., 2004). Toxic aluminium levels retard root growth causing various root discolorations, deformations, and that ultimately result in low grain yield (Blum, 1986; Villagarcia, 2001). Soybean and other legumes are sensitive to aluminium and do not grow well in acids soils, except when the soil is limed to raise the pH to a neutral level (pH 7.0). But the cost of liming is very prohibitive for the poor-resource farmers in tropical Africa. Hence, there is need to develop soybean varieties that are tolerant to aluminium stress. The screening of genotypes is a prerequisite for

the selection and development of tolerant varieties. Various screening methodologies ranging from hydroponics, sand culture, to pot/field experiments have been adopted in searching for aluminium tolerant genotypes of soybean (Campbell and Carter Jr., 1990; Bianchi-Hall et al., 1998; Bianchi-Hall et al., 2000; Ermolayev et al., 2003). Hydroponics screening however, has advantages of close observation of the roots as the experiment progresses and it can be regulated and reproduced. It can also be used to rapidly screen a large number of germplasm. Previous studies on hydroponics screening are however concentrated in the temperate/subtropical regions of the world, where the prevailing weather for the most part of the year are



unfavorable for optimum growth of soybean. Hence, hydroponics screening of seedlings for aluminum stress tolerance in the temperate/subtropical regions of the world is often carried out in phytotrons to provide favorable environment for optimum growth of prevailing soybean. The atmospheric temperature and relative humidity for the most part of the year coupled with constant daily sunshine in tropical Africa, particularly Nigeria, favor the growth of soybean. Thus, hydroponics screening in Nigeria could be carried out at room temperatures  $(25 - 33^{\circ}C)$ on laboratory tables at anytime of the year without incurring the energy cost of controlling

## MATERIALS AND METHODS

The experiment was conducted on the laboratory tables in the Crop Science Laboratory of the University of Agriculture, Makurdi, Nigeria, between 15th January and 10th March, 2002. In order to ensure an appropriate level of aluminium activity for the characterization of tropically adapted soybean genotypes, a wide range of aluminium activity comprising ten levels (0, 5, 50, 100, 150, 200, 250, 300, 350 and 400µMAl3+) were selected for the experiment. Fifteen tropically adapted IITA released varieties of soybeans were selected for the experiment on the basis of previous rating for adaptability on acid soils of Nigeria (Okpara and Ibiam, 2000; Yusuf and Idowu, 2001) and the availability of seeds. The varieties that were selected based on previous reports on adaptability studies include: TGX 923-2E, TGX 1740-2E, TGX 1805-31F, TGX 1802-1F, TGX 1485-1D, and TGX 1440-1E. Variety TGX 923-2E had been previously cited as acid stress sensitive (Yusuf and Idowu, 2001). Varieties TGX 1740-2E, TGX 1805-31F and TGX 1802-1F have also been rated as acid stress sensitive by Okpara and Ibiam (2000). TGX 1485-1D and TGX 1440-1E are the only genotypes previously observed to be acid stress tolerant and recommended for production on the acid soils of South - East Nigeria (Okpara and Ibiam, 2000). The variety TGX 1448-2E was included because it is the most popular genotype currently in production in the major soybean producing areas of Nigeria (the Southern Guinea Savanna ecological zone of Nigeria). The following eight varieties, TGX 1830-20E, TGX 1844-18E, TGX 1873-16E, the temperature, relative humidity or light. The difficulty encountered in sourcing for aluminum tolerant genotypes of soybean restricts the screening of soybean for aluminium stress tolerance to the availability of seeds. Villagarcia et al. (2001) had previously alluded to such difficulty.

The cost of screening over a wide range of aluminum activity is expensive for the developing countries of the world. Hence the need to first determine the appropriate level of aluminium activity for the screening of tropically adapted soybean genotypes, which was the main objective of this research.

TGX 1876-4E, TGX 1878-7E, TGX 1891-3F, TGX 1895-35F and TGX 1896-3F were randomly selected and included among the fifteen varieties based on availability of seeds.

Twenty seeds of each of the fifteen genotypes were surface sterilized with ethanol (1%v/v) for 1 minute and then with 0.85% sodium hypochlorite for 3 minutes and rinsed 6 times with deionised water (Ramirez et al. 1997), prior to germination. Petridishes were similarly sterilized. Thereafter, cotton wools were soaked with deionised water in the petridishes and the seeds were placed on them for germination. After four days, seedlings with poor vigour and twisted radicles were discarded, while ten vigorous healthy seedlings of each genotype were transferred to each of the continuously bubbling hydroponics tanks, fitted with aerators. Each tank was a 5-litre capacity plastic tank of 20cm diameter covered with a removable plastic lid. Each lid had holes of 0.6cm diameter in which single seedlings were fitted and held in place with cotton wool. The hydroponics nutrient concentrations solution culture was constituted following the procedures of Howell and Bernard (1961) with some modification (Table 1).

Each tank was filled with 3 litres of deionised water, and nutrients required for the tank were weighed and dissolved in the de-ionized water. The pH of the nutrient solution was then adjusted to  $4.05\pm0.05$ by adding a few drops of 0.1M concentrated sulphuric acid. Aluminum treatments were in the form of Al<sub>2</sub>(SO<sub>4</sub>)<sub>3</sub>. The desired aluminum activity for each tank was prepared separately by weighing



out the quantity of  $Al_2(SO_4)$  required and dissolving it in one litre of deionised water (pH of  $4.05\pm0.05$ ).

Table 1: Composition of Nutrients in Hydroponics

| Chemical  | Concentration           |
|---|-------------------------|
| KH <sub>2</sub> PO <sub>4</sub>                     | $0.5 \text{mML}^{-1}$   |
| KCl   | 0.5mM L <sup>-1</sup>   |
| NaH <sub>2</sub> PO <sub>4</sub> .2H <sub>2</sub> O | 1.0mM L <sup>-1</sup>   |
| NH4NO3  | 0.8mM L <sup>-1</sup>   |
| $Ca(NO_3)_2.4H_2O$                                  | 1.5mM L <sup>-1</sup>   |
| MgSO <sub>4</sub>                                   | $1.00 \text{mM L}^{-1}$ |
| $Fe(NO_3)_2$  | 80µM L-1                |
| $H_3BO_3$   | 20µM L <sup>-1</sup>    |
| $ZnSO_4.7H_2O$                                      | 3µM L-1                 |
| MnCl <sub>2</sub> .4H <sub>2</sub> O                | 3µM L-1                 |
| $CuSO_{4.}5H_{2}O$                                  | 3μM L-1                 |
| (NH4)6M07O24.4H2O                                   | 0.8µM L <sup>-1</sup>   |

mM  $L^{-1}$  =Millimole per litre ,  $\mu$ M  $L^{-1}$ =Micromole per litre The various levels of Al<sup>3+</sup> were supplied in the form of Al<sub>2</sub> (SO4)<sub>3.</sub> Adapted from Howell and Bernard (1961)

Thereafter, the one litre aluminum solution was poured into the nutrient solution, and the solution made up to 5-litre mark with deionised water (pH

#### RESULTS

Plants grown at 350 and 400µMAl<sup>3+</sup> levels of aluminium toxicity were characterized by shoot stunting, yellowing and browning of leaves, abnormal root development and eventual death.

 $4.05\pm0.05$ ). Aerators were then connected to the tank and the solution allowed to bubble continuously for two hours before seedlings were transferred to it. The seedlings grew for three days (3D) in the hydroponics and were harvested at seven days old i.e. germinated for four days and grown for three days in the hydroponics. The experimental design was a split-plot with the 15 genotypes as the main plot and ten levels of aluminum activity as the sub-plots. Each tank represented an aluminum activity level and three replications were employed. Diurnal temperatures ranged from 30°C – 33°C while night temperatures ranged from 27°C - 29°C. On harvesting, five seedlings were randomly selected from each tank, and data taken on primary root length, shoot length, root dry weight and shoot dry weight. Primary root length was defined as the distance from the root tip to the junction region between the root and the hypocotyls (Bianchi-Hall, 1998). Shoot length was simply measured as the length of the primary shoot. Roots and shoots were separated, air dried for three hours, and then oven dried at 70°C for 48 hours before taking their respective weights.

Consequently, no data were available for these concentrations. Mean squares for root and shoot traits of 15 soybean genotypes grown at eight levels of aluminium activity are summarized in Table 2.

**Table 2:** Mean squares for root and shoot traits of 15 soybean genotypes grown at 8 levels of aluminium activity  $(0, 5, 50, 100, 150, 200, 250 \text{ and } 300 \mu \text{mal}^{3+})$  in hydroponics for 3 days (3d).

| Source of Variation  | Df  | Primary      | Shoot Length  | Root       | Shoot      |  |  |  |
|----------------------|-----|--------------|---------------|------------|------------|--|--|--|
|                      |     | Root Length  |               | Dry Weight | Dry Weight |  |  |  |
| Reps.                | 2   | 0.076800     | 0.860000      | 0.000004   | 0.000003   |  |  |  |
| Aluminium            | 7   | 176.714900** | 1791.742400** | 0.000686** | 0.002654** |  |  |  |
| Genotype             | 14  | 89.843000**  | 106.563500**  | 0.000843** | 0.000961** |  |  |  |
| Genotype × Aluminium | 98  | 20.063200**  | 39.194300**   | 0.000467** | 0.001520** |  |  |  |
| Error                | 238 | 0.095000     | 0.496300      | 0.000003   | 0.000002   |  |  |  |

\*, \*\*: Significant at P<0.05 and P<0.01 respectively.

Highly significant differences in aluminium activities, genotypic differences, as well as genotype X aluminium interaction were observed for primary root length, shoot length, root dry weight and shoot dry weight. Primary root length decreased from 8.6000cm to 4.0411cm as aluminium toxicity increased from 0 to 300µMAl<sup>3+</sup> (Fig. 1). The highest decrease in the rate of growth in primary root length was observed between 0µMAl<sup>3+</sup> and

50μMAl<sup>3+</sup>. Further increase in aluminium activity beyond 50μMAl<sup>3+</sup> did not result in any substantial decrease in the rate of growth of primary root length. Shoot length decreased from 25.3655cm to 5.4107cm as aluminium toxicity increased from 0 to 300μMAl<sup>3+</sup> activity (Fig. 2). The highest decrease in the rate of growth in shoot length was observed between 0 and 5μMAl<sup>3+</sup>. Shoot length inhibition due to aluminium treatment at low level (5μMAl<sup>3+</sup>)



of activity, was very severe as it reduced shoot length to 31% of its length in the absence  $(0\mu MAl^{3+})$  of aluminium. The figure also shows that there was a decrease in the rate of growth of shoot length with increasing aluminium activity between  $5\mu MAl^{3+}$  and  $50\mu MAl^{3+}$ . Subsequent increase in aluminium activity resulted in a very little decrease in the rate of growth of the shoot length. Root dry weight decreased from 0.0200 to 0.0102g plant<sup>1</sup> as aluminium toxicity increased from 0 to  $300\mu$ MAl<sup>3+</sup> (Fig. 3).



Figure 1: Mean primary root length (cm) of soybeans in response to aluminium treatment in hydroponics



Figure 2: Mean Shoot Length (cm) of Soybeans in Response to Aluminium Treatment in Hydroponics





Figure 3: Mean root dry weight (g plant<sup>-1</sup>) of soybeans in response to aluminium treatment in hydroponics

The rate of decrease in root dry matter accumulation was however highest between 0 and  $50\mu$ MAl<sup>3+</sup>. Subsequently, the rate of root dry matter accumulation gradually decreased as aluminium activity increased up to  $300\mu$ MAl<sup>3+</sup> except between

 $100\mu$ MAl<sup>3+</sup> and  $150\mu$ MAl<sup>3+</sup>. The pattern of decrease in the rate of shoot dry matter accumulation with increase in aluminium activity (Fig. 4) was similar to the pattern of decrease in the rate of growth of shoot length.



Figure 4: Mean Shoot Dry Weight (g plant<sup>1</sup>) of Soybeans in Response to Aluminium Treatment in Hydroponics



Shoot dry weight decreased from 0.1231 to 0.0621g plant<sup>-1</sup> as aluminium activity increased from 0 to  $300\mu$ MAl<sup>3+</sup>. The rate of decrease in shoot dry matter accumulation was highest between 0 and  $5\mu$ MAl<sup>3+</sup> aluminium activity. Aluminium treatment at low level ( $5\mu$ MAl<sup>3+</sup>) resulted in 56% decrease (Fig. 4) in shoot weight relative to the control ( $0\mu$ MAl<sup>3+</sup>).

Subsequent increase in aluminium activity resulted in a gradual decrease in the rate of decrease in shoot dry matter accumulation. Mean separation for the primary root length, shoot length, root dry weight and shoot dry weight of the 15 soybean genotypes treated with eight(8) levels of aluminium activity (0, 5, 50, 100, 150, 200, 250, and  $300\mu$ MAl<sup>3+</sup>) are presented in Tables 3, 4, 5 and 6.

**Table 3:** Primary Root Length (cm) Means for 15 Soybean Genotypes Grown at 8 Levels of Aluminium Activity (0, 5, 50, 100, 150, 200, 250 and 300µMAl<sup>3+</sup>) in Hydroponics for 3 Days (3D)

| GENOTYPE     | 0       | 5       | 50     | 100    | 150    | 200    | 250    | 300    |
|--------------|---------|---------|--------|--------|--------|--------|--------|--------|
| TGX 1740-2E  | 6.5000  | 3.2000  | 2.8000 | 2.5000 | 2.3000 | 2.0800 | 2.0300 | 2.0167 |
| TGX 1485-1D  | 10.0000 | 4.3000  | 2.6000 | 2.6000 | 2.6000 | 2.5500 | 2.5200 | 2.5000 |
| TGX 1830-20E | 2.8000  | 2.8000  | 2.4000 | 2.3800 | 2.3500 | 2.3500 | 2.2500 | 2.2083 |
| TGX 1876-4E  | 8.5000  | 5.5000  | 2.2000 | 2.2000 | 2.1800 | 2.1500 | 2.1200 | 2.1100 |
| TGX 1805-31F | 9.0000  | 5.0000  | 4.7000 | 4.6000 | 4.6000 | 4.0000 | 3.6500 | 3.0333 |
| TGX 1873-16E | 6.5000  | 5.8000  | 5.6000 | 5.3000 | 5.2000 | 5.1000 | 5.1000 | 5.0833 |
| TGX 1878-7E  | 9.5000  | 11.2000 | 8.6000 | 8.4000 | 8.2000 | 8.2000 | 8.1500 | 8.0000 |
| TGX 1802-1F  | 13.2000 | 5.1000  | 3.0000 | 3.0000 | 3.0000 | 2.9000 | 2.9000 | 2.5030 |
| TGX 1891-3F  | 10.6000 | 5.0000  | 3.6333 | 3.5500 | 3.5100 | 3.5000 | 3.3000 | 3.1000 |
| TGX 1896-3F  | 9.5000  | 14.0000 | 9.6167 | 9.5000 | 9.4000 | 9.3000 | 9.2000 | 9.1800 |
| TGX 1844-18E | 10.9000 | 15.0000 | 9.5167 | 9.2000 | 8.5000 | 8.2000 | 8.2000 | 7.8000 |
| TGX 1440-1E  | 8.0000  | 4.1000  | 3.5167 | 3.5100 | 3.5000 | 3.3100 | 3.3000 | 3.1000 |
| TGX 1448-2E  | 9.0000  | 3.5000  | 3.2100 | 3.2000 | 3.2000 | 3.1000 | 2.9000 | 2.6000 |
| TGX 1895-35F | 4.7000  | 4.3000  | 4.0167 | 4.0000 | 3.9000 | 3.8500 | 3.8500 | 3.3000 |
| TGX 923-2E   | 10.3000 | 4.5450  | 4.5167 | 4.3000 | 4.2000 | 4.1133 | 4.1000 | 4.0830 |
| Mean         | 8.6000  | 6.2230  | 4.6418 | 4.4442 | 4.4427 | 4.3136 | 4.1379 | 4.0411 |
| $LSD_{0.05}$ | 1.0735  | 1.0158  | .8274  | .5531  | 1.1087 | .9138  | .8028  | .9961  |

| Table 4: Shoot Length (cm)    | Means for 15 Soybean | Genotypes Grown at     | 8 Levels of Aluminium / | Activity (0, |
|-------------------------------|----------------------|------------------------|-------------------------|--------------|
| 5, 50, 100, 150, 200, 250 and | 300µMAl3+) in Hydrop | onics for 3 Days (3D). |                         |              |

| GENOTYPE     | 0       | 5       | 50      | 100     | 150     | 200     | 250     | 300     |
|--------------|---------|---------|---------|---------|---------|---------|---------|---------|
| TGX 1740-2E  | 20.5000 | 3.0170  | 2.2170  | 2.2100  | 2.2000  | 2.2000  | 2.0000  | 1.9800  |
| TGX 1485-1D  | 10.1330 | 4.0330  | 3.2670  | 3.1000  | 3.0800  | 3.0500  | 3.0500  | 2.7670  |
| TGX 1830-20E | 12.0000 | 4.2000  | 3.4920  | 3.4000  | 3.2000  | 3.1000  | 3.1000  | 3.0500  |
| TGX 1876-4E  | 15.1000 | 4.9170  | 3.5080  | 3.5000  | 3.4100  | 3.2000  | 3.1100  | 3.0000  |
| TGX 1805-31F | 35.5000 | 13.8420 | 10.7830 | 10.5000 | 10.3000 | 10.1000 | 10.1000 | 8.0830  |
| TGX 1873-16E | 15.1170 | 6.0500  | 4.8000  | 4.8000  | 4.4000  | 4.3000  | 4.1000  | 4.1000  |
| TGX 1878-7E  | 32.5000 | 11.8330 | 6.5170  | 6.5000  | 6.5000  | 6.2000  | 6.2000  | 6.0500  |
| TGX 1802-1F  | 23.0830 | 5.5580  | 4.0830  | 4.0500  | 4.0500  | 4.0400  | 4.0400  | 4.0000  |
| TGX 1891-3F  | 44.8330 | 4.6580  | 3.2080  | 3.2000  | 3.2000  | 3.1500  | 3.1200  | 3.0170  |
| TGX 1896-3F  | 25.9170 | 13.4170 | 10.5330 | 10.5000 | 10.2000 | 10.1000 | 10.1000 | 9.9170  |
| TGX 1844-18E | 43.2330 | 23.5830 | 20.5000 | 19.9000 | 19.8000 | 19.6000 | 19.5000 | 18.0800 |
| TGX 1440-1E  | 25.0000 | 4.1330  | 3.2170  | 3.2000  | 3.1000  | 3.1000  | 3.0500  | 3.0170  |
| TGX 1448-2E  | 32.6000 | 6.6500  | 6.5670  | 5.4000  | 5.2000  | 5.2000  | 5.0000  | 4.4830  |



| TGX 1895-35F | 29.8000 | 4.0930 | 3.0800 | 3.0200 | 3.0100 | 3.0100 | 3.0000 | 3.0000 |
|--------------|---------|--------|--------|--------|--------|--------|--------|--------|
| TGX 923-2E   | 15.1670 | 7.6500 | 6.5333 | 6.4000 | 6.2000 | 6.1000 | 6.0500 | 6.0167 |
| Mean         | 25.3655 | 7.7756 | 6.0870 | 5.9787 | 5.8567 | 5.7633 | 5.7013 | 5.4107 |
| $LSD_{0.05}$ | 3.0462  | 2.4443 | 1.1538 | 1.8599 | 2.0503 | 1.6925 | 1.7671 | 1.5343 |

**Table 5:** Root Dry Weight (g plant<sup>-1</sup>) Means for 15 Soybean Genotypes Grown at 8 Levels of Aluminium Activity (0, 5, 50, 100, 150, 200, 250 and 300µMAl<sup>3+</sup>) in Hydroponics for 3 Days (3D).

| GENOTYPE            | 0     | 5     | 50    | 100   | 150   | 200   | 250   | 300   |
|---------------------|-------|-------|-------|-------|-------|-------|-------|-------|
| TGX 1740-2E         | .0190 | .0110 | .0102 | .0100 | .0098 | .0095 | .0095 | .0093 |
| TGX 1485-1D         | .0178 | .0122 | .0115 | .0111 | .0110 | .0100 | .0100 | .0092 |
| TGX 1830-20E        | .0132 | .0130 | .0090 | .0088 | .0085 | .0085 | .0082 | .0082 |
| TGX 1876-4E         | .0180 | .0162 | .0098 | .0090 | .0090 | .0088 | .0085 | .0085 |
| TGX 1805-31F        | .0170 | .0130 | .0130 | .0130 | .0128 | .0128 | .0122 | .0122 |
| TGX 1873-16E        | .0182 | .0170 | .0150 | .0150 | .0140 | .0140 | .0130 | .0130 |
| TGX 1878-7E         | .0220 | .0258 | .0180 | .0140 | .0120 | .0120 | .0110 | .0110 |
| TGX 1802-1F         | .0230 | .0120 | .0093 | .0090 | .0085 | .0080 | .0080 | .0080 |
| TGX 1891-3F         | .0258 | .0150 | .0083 | .0070 | .0065 | .0065 | .0062 | .0062 |
| TGX 1896-3F         | .0198 | .0260 | .0200 | .0123 | .0122 | .0122 | .0122 | .0122 |
| TGX 1844-18E        | .0202 | .0250 | .0200 | .0140 | .0138 | .0135 | .0122 | .0122 |
| TGX 1440-1E         | .0182 | .0130 | .0120 | .0110 | .0108 | .0105 | .0103 | .0103 |
| TGX 1448-2E         | .0252 | .0140 | .0113 | .0112 | .0111 | .0110 | .0108 | .0103 |
| TGX 1895-35F        | .0230 | .0183 | .0142 | .0120 | .0110 | .0110 | .0108 | .0102 |
| TGX 923-2E          | .0202 | .0183 | .0123 | .0120 | .0120 | .0110 | .0110 | .0110 |
| Mean                | .0200 | .0167 | .0130 | .0129 | .0109 | .0106 | .0103 | .0101 |
| LSD <sub>0.05</sub> | .0027 | .0042 | .0031 | .0031 | .0017 | .0017 | .0016 | .0016 |

| Table 6: Shoot dry weight (g plant <sup>-1</sup> ) mean   | s for 15 soybean genotypes grow | n at 8 levels of aluminium activity |
|---|---------------------------------|-------------------------------------|
| (0, 5, 50, 100, 150, 200, 250 and 300µmal <sup>3+</sup> ) | in hydroponics for 3 days (3d). |                                     |

| (0, 5, 50, 100, 150, 200, 250 and 500µmar ) in hydropolites for 5 days (50). |       |       |       |       |       |       |       |       |  |
|--|-------|-------|-------|-------|-------|-------|-------|-------|--|
| GENOTYPE   | 0     | 5     | 50    | 100   | 150   | 200   | 250   | 300   |  |
| TGX 1740-2E  | .1533 | .0578 | .0483 | .0470 | .0470 | .0465 | .0460 | .0460 |  |
| TGX 1485-1D  | .1300 | .0607 | .0507 | .0500 | .0490 | .0490 | .0490 | .0482 |  |
| TGX 1830-  | .1228 | .0588 | .0478 | .0475 | .0472 | .0471 | .0470 | .0470 |  |
| 20E  |       |       |       |       |       |       |       |       |  |
| TGX 1876-4E  | .1312 | .0613 | .0507 | .0500 | .0490 | .0490 | .0490 | .0488 |  |
| TGX 1805-31F   | .1105 | .0630 | .0588 | .0580 | .0580 | .0570 | .0570 | .0565 |  |
| TGX 1873-  | .0978 | .1062 | .0975 | .0950 | .0920 | .0920 | .0891 | .0891 |  |
| 16E  |       |       |       |       |       |       |       |       |  |
| TGX 1878-7E  | .1133 | .0840 | .0797 | .0790 | .0770 | .0760 | .0760 | .0755 |  |
| TGX 1802-1F  | .1010 | .0512 | .0443 | .0440 | .0420 | .0400 | .0400 | .0400 |  |
| TGX 1891-3F  | .1617 | .0942 | .0875 | .0820 | .0810 | .0810 | .0808 | .0806 |  |
| TGX 1896-3F  | .1238 | .0773 | .0700 | .0700 | .0680 | .0670 | .0650 | .0645 |  |
| TGX 1844-  | .1248 | .0962 | .0940 | .0910 | .0910 | .0900 | .0900 | .0885 |  |
| 18E  |       |       |       |       |       |       |       |       |  |
| TGX 1440-1E  | .0917 | .0742 | .0650 | .0630 | .0630 | .0630 | .0630 | .0628 |  |
| TGX 1448-2E  | .1548 | .0862 | .0752 | .0720 | .0720 | .0720 | .0720 | .0715 |  |
| TGX 1895-35F   | .1102 | .0738 | .0673 | .0610 | .0550 | .0530 | .0510 | .0500 |  |
| TGX 923-2E   | .1203 | .0458 | .0397 | .0390 | .0380 | .0370 | .0360 | .0353 |  |
| Mean   | .1231 | .0689 | .0651 | .0632 | .0619 | .0613 | .0607 | .0603 |  |
| LSD <sub>0.05</sub>  | .0168 | .0158 | .0165 | .0159 | .0160 | .0161 | .0160 | .0159 |  |



Primary root length ranged from 2.8cm for TGX1830-20E to 13.2000cm for TGX 1802-1F in the absence (0µMAl3+) of aluminium, with most genotypic means greater than the population mean (Table 3). Aluminium treatment at low level (5µMAl3+) produced a wider range in genotypic means (2.8000cm to 15cm) compared to the control, discriminating the genotypes into sensitive and tolerant groups. Three genotypes, namely, TGX 1896-3F, TGX 1844-18E, and TGX 1878-7E produced longer primary roots than the population mean. There was a significant increase in the growth of the primary root length between 0 and 5µMAl<sup>3+</sup> for TGX 1896-3F and TGX 1844-18E. No significant decrease in the primary root length was observed for all genotypes as aluminium activity increased from 50µMAl3+ to 300µMAl3+ with most genotypes producing shorter primary roots compared to the population means at the respective levels of aluminium activity. Conspicuous changes were observed in the ranking of genotypes with progressive increases in the levels of aluminium activity up to 50µMAl3+. For example, TGX 1802-1F produced the longest primary roots in the control, but moved back to the 6th position at 5µMAl<sup>3+</sup> level of activity and further declined to the 11th position at 50µMAl3+ level of aluminium activity. Only a few genotypes changed rank between 50 and 300 µMAl<sup>3+</sup>.

Shoot length ranged from 10.1330cm to 44.8330cm in the absence of aluminium (0µMAl3+) with seven of the genotypes recording means greater than population mean (Table 4). Genotypic means at 5µMAl<sup>3+</sup> level of activity ranged from 3.0170cm to 23.5830cm. A decrease in shoot length relative to the control was observed at low level aluminium (5µMAl<sup>3+</sup>) for all genotypes. However the rate of decrease between the control and 5µMAl3+ varied among the genotypes leading to conspicuous changes in genotypic ranking. The shoot length of TGX 1891-3F decreased to as low as 10% of the control and moved from the 1st position in the control to the 10th position at 5µMAl3+ level. The shoot length of TGX 1896-3F on the other hand decreased to only 55% of the control and moved

## DISCUSSION

The inability of the germplasm to survive beyond 300µMAl<sup>3+</sup> is an indication that screening of tropically adapted population should not exceed 300µMAl<sup>3+</sup> level of aluminium activity. The reduction of means in response to aluminium

from the 7<sup>th</sup> position in the control to the 3<sup>rd</sup> position at 5 $\mu$ MAl<sup>3+</sup> level. The shoot length of TGX 1844-18E was almost constant from 50 to 300 $\mu$ MAl<sup>3+</sup> levels and significantly longer than that of any other variety at 300 $\mu$ MAl<sup>3+</sup> (18.0800cm). The changes in genotypic ranking relative to the control, was sustained until 50 $\mu$ MAl<sup>3+</sup> level of activity. A good agreement in genotypic ranking was observed between 50 $\mu$ MAl<sup>3+</sup> and higher levels of activity.

The pattern of variation in genotypic means for the root dry weight is similar to that observed for the primary root length. Genotypic means for the root dry weight ranged from 0.0132g plant-1 for TGX 1830-20E to 0.0258g plant-1 for TGX 1891-3F in the control (Table 5). A progressive decrease in root dry matter accumulation was observed with increasing level of aluminium toxicity. Genotypic response also varied with increasing aluminium level. Aluminium treatment at low level of 5µMAl3+ enhanced root dry matter accumulation in TGX 1878-7E, TGX 1896-3F and TGX 1844-18E, leading to conspicuous changes in genotypic ranking relative to the control. The change in genotypic ranking was also observed at 50µMAl3+ relative to the control. A good agreement in genotypic ranking was observed between 50µMAl3+ and higher levels of activity.

Shoot dry weight ranged from 0.0917g plant<sup>-1</sup> for TGX 1440-1E to 0.1617g plant-1 for TGX 1891-3F at 0µMAl3+ level (Table 6). A gradual decrease in shoot dry matter accumulation was observed with increasing severity in aluminium stress for all genotypes. However, varying degrees of response to aluminium stress were observed in the genotypes, leading to changes in genotypic ranking between 0µMAl<sup>3+</sup> and 5µMAl<sup>3+</sup> levels of activity. While the shoot dry weight of TGX 1802-1F at 5µMAl3+ decreased to 50% of its weight in the control, that of TGX 1873-16E was enhanced by aluminium treatment at 5µMAl<sup>3+</sup> level, increasing the shoot dry weight to 109% of its weight in the control. The greatest change in genotypic ranking was observed between 0µMAl3+ and 5µMAl3+. Only very little changes in ranking were observed with increasing aluminium activity beyond 5µMAl3+.

treatment in all the traits studied and the change in genotypic ranking observed between the control  $(0\mu MAl^{3+})$  and other levels of aluminium activity are indications that the growth inhibition observed was not as a result of acid stress, but the presence of



phytotoxic elements in the media. Such phenomenon had been previously observed (Foy et al., 1969; Ermolayev, 2003) and attributed to aluminium toxicity. Foy et al., (1969) had observed that with no added aluminium to solution culture, Perry and Chief Varieties of soybean produced the same absolute root yield. But under aluminium stress, a marked difference in yield was observed. Similarly, Ermolayev et al., (2003) in their studies on gene expression in Tambora (Al-tolerant) and Malabar (Al-sensitive) cultivars of soybean observed visible morphological differences between the two cultivars as a result of aluminium treatment. The higher percentage of reduction in shoot traits compared to root traits between the control (0µMAl<sup>3+</sup>) and 5µMAl<sup>3+</sup> level of aluminium activity indicates that shoot traits are more sensitive to the phytotoxic effect of aluminium than root traits. The inconsistency of this observation with previous findings of (Villagarcia et al., 2001), could be attributed to differences in the environment in which the experiments were carried out. The current work was carried out at room temperature in the tropics without control on the natural temperature, relative humidity and light while the

#### REFERENCES

- Bianchi-Hall, C.M., T.E. Carter Jr., T.W. Rufty, C. Arellano, H.R. Boerma, D.A. Ashley and J.W. Burton (1998). Heritability and resource allocation of aluminium tolerance derived from soybean PI416937. *Crop Science* 38: 513 – 522.
- Bianchi-Hall, C.M., T.E. Carter Jr., M.A. Bailey, M.A.R. Mian, T.W. Rufty, D.A. Ashley, H.R. Boerma, C. Arellano, R.S. Hussey, and W.A. Parrott (2000). Aluminium tolerance associated with quantitative traits loci derived from soybean PI416937 in hydroponics. *Crop Science* 40: 538 – 545.
- Blum A. (1986). Plant Breeding for Stress Environments. CRC Press, USA. 223Pp. Campbell, K.A., K.A.G. and T.E. Carter Jr., (1990). Aluminium tolerance in soybean: 1. Genotypic correlation and repeatability of solution culture and green house screening methods. *Crop science* 30: 1049 – 1054.
- Campbell, K.A., K.A.G. and T.E. Carter Jr., (1990). Aluminium tolerance in soybean:1. Genotypic correlation and repeatability of solution culture and green house screening

previous experiment was carried out in a temperate country with control on the environment.

The change in genotypic ranking observed for all the traits studied is consistent with previous findings (Villagarcia et al., 2001; Ermolayev et al., 2003; Ojo, 2010). The change in genotypic ranking observed between  $0\mu MAl^{3+}$  and  $5\mu MAl^{3+}$  and between 5µMAl3+ and 50µMAl3+, indicates that no inference can be drawn on the tolerance status of the tropical soybean germplasm until they have been screened through these three levels of aluminium activity. However, the good agreement in ranking observed between 50µMAl3+ and higher levels of aluminium activity for all the traits studied, indicates that screening beyond the 50µMAl<sup>3+</sup> level of aluminium activity may be unnecessary. Screening at 300µMAl3+ is however necessary, because it reflects the aluminium tolerance status of tropical soybean genotypes at the highest level of aluminium toxicity. Hence, the 0, 5, 50 and 300µMAl3+ levels of aluminium activity could be selected for the screening of tropically adapted soybean genotypes in the tropical environment at room temperature.

methods. Crop science **30**: 1049 – 1054.

- Ermolayev, V., W. Weschke, and R. Manteufel (2003). Comparison of Al-induced gene expression in sensitive and tolerant soybean cultivars. *Journal of Experimental Biology* **54** (393): 2745 – 2756.
- Foy, C.D., A.L. Fleming and W.H. Armiger (1969). Aluminium tolerance of soybean varieties in relation to calcium nutrition. *Agronomy Journal* **61**: 505 – 511.
- Furlani, R. B. and R. B. Clark (1981). Screening sorghum for aluminium tolerance in nutrient solutions. Agronomy Journal 73:587 – 594.
- Houde M. and A.O. Diallo (2008). Identification of genes and pathways associated with aluminum stress and tolerance using transcriptome profiling of wheat near – isogenic lines. BMC Genomics 9:400.
- Howell, R.W., and R.L. Bernard (1961). Phosphorus response of soybean varieties. *Crop Science* **1**: 311 313.
- Kochian, L. V. (1995). Cellular mechanisms of aluminium toxicity and resistance in plants. Annual Rev. Plant Physiol. Plant Mol. Biol.



46:237 - 260.

- Okpara, D.A. and B. Ibiam (2000). Evaluation of soybean varieties for adaptability to a humid tropical environment in South Eastern Nigeria. *Journal of Sustainable Agricultural Environment* **2**(1): 26 – 31.
- Ojo, G. O. S. (2010). Genotypic variation and diallel analysis for aluminium sress tolerance in soybean (*Glycine max* (L.) Merrill). PhD Diss., University of Agriculture, Makurdi, Nigeria.
- Ramirez, M.E., D.W. Isarael and A.G. Wollum (1997). Phenotypic characterization of soybean *Bradyrhizobia* in two soils of North Carolina. *Soil Biology and Biochemistry* 29: 1547 – 1555.

Tani, A., D. Zhang, J. A. Duine and F. Kawai

(2004). Treatment of the yeast *Rhodotorula* glutinis with AlCl<sub>3</sub> leads adaptive acquirement of heritable aluminium resistance. Applied Microbiol. Biotechnol. 65:344 - 348.

- Villagarcia, M., T.E. Carter, Jr., T.W. Rufty, A.S. Niewoehner, M.W. Jennette, and C. Arrellano (2001). Genotypic rankings for aluminium tolerance of soybean roots grown in hydroponics and sand culture. *Crop Science* **41**(5): 1499 – 1507.
- Yusuf, I.A. and A.A. Idowu (2001). Evaluation of four soybean varieties for performance under different lime regimes on the acid soil of Uyo, Nigeria. *Tropical Oilseeds Journal* 6: 65 – 70.