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# Determination of appropriate level(s) of aluminium activity for the screening of tropically adapted soybean genotypes in sand culture

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A sand culture experiment was conducted at the University of Agriculture, Makurdi, Nigeria, in 2001 and 2002, with the objective of determining the appropriate levels of aluminium activity to screen tropically adapted genotypes of soybean for aluminium stress tolerance. Fifteen genotypes of soybean constituted the main plot, while eight levels of aluminium activity (0, 50, 100, 200, 300, 400, 450 and 500  $\mu\text{MAI}^{3+}$ ) constituted the subplots in a split-plot design. The experiment was replicated three times. Plants watered with the 500  $\mu\text{MAI}^{3+}$  treatment got burnt after 3 to 5 days and were removed from the treatment. Data were taken on the root dry weight, shoot dry weight and relative root surface area at 25DAP. Aluminium, genotype and genotype  $\times$  aluminium interaction effects were all highly significant. Aluminium activity at 300  $\mu\text{MAI}^{3+}$  level along with the control (0  $\mu\text{MAI}^{3+}$ ) were considered as appropriate for root dry weight, while the 450  $\mu\text{MAI}^{3+}$  level along with the control (0  $\mu\text{MAI}^{3+}$ ) were considered as appropriate for shoot dry weight and relative root surface area and recommended for the rapid screening of tropically adapted genotypes of soybean in sand culture in the tropics. The relative root surface area was the most sensitive in discriminating between levels of aluminium activity and should have preference in any selection programme.

**Key words:** Aluminium activity, sand culture screening, tropically adapted genotypes of soybean.

## INTRODUCTION

Soybean is a crop that can be easily produced without much requirement for fertilizer. Hence, it is extensively produced in temperate, tropical and subtropical regions of the World. Unfortunately, 40% of the World's arable lands are acidic (Kochian, 1995) and this restricts the production of soybeans and other legumes to the remaining 60%. The growth of leguminous crops and development of symbiosis on acid soils are generally affected by deficiencies of Ca, K, P, Mg, S, Zn and Mo and/or toxicities of Al, Mn and Fe (Clark et al., 1988; Foy et al., 1978; Foy, 1984; Sanchez and Salina, 1981). Aluminium is the major phytotoxic element in acid soils (Kochian, 1995) and toxic levels of aluminium retard root growth causing various root deformations, and discolou-

rations that ultimately result in low grain yield (Blum, 1986; Villagarcia et al., 2001).

Liming has been used to ameliorate the problem of aluminium toxicity/low pH in soils. Liming the top soil however, remains a temporary solution due to subsoil acidity. Moreover, the cost of liming particularly in developing countries is prohibitive and does not justify such huge investment given the return on investment from grain yield of soybeans. The development of aluminium tolerant cultivars of soybean therefore remains a viable alternative.

The screening of genotypes is a prerequisite for the selection and development of tolerant varieties (Ojo et al., 2010). Screening of crop plants for aluminium stress tolerance is not a recent phenomenon and various screening methodologies particularly hydroponics, sand and soil cultures have been employed in such screening experiments. However, soil and hydroponics media have

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**Table 1.** Composition of nutrients in the sand culture.

Chemical	Concentration
KH <sub>2</sub> PO <sub>4</sub>	1.5 mM
K <sub>2</sub> SO <sub>4</sub>	1.5 mM
NaH <sub>2</sub> PO <sub>4</sub> ·2H <sub>2</sub> O	1.0 mM
NH <sub>4</sub> NO <sub>3</sub>	2.0 mM
CaSO <sub>4</sub> ·½H <sub>2</sub> O	3.0 mM
MgSO <sub>4</sub>	100 µM
Fe(NO <sub>3</sub> ) <sub>2</sub>	20 µM
H <sub>3</sub> BO <sub>3</sub>	10 µM
ZnSO <sub>4</sub> ·7H <sub>2</sub> O	1.5 µM
MnCl <sub>2</sub> ·4H <sub>2</sub> O	1.5 µM
CuSO <sub>4</sub> ·5H <sub>2</sub> O	0.5 µM
(NH <sub>4</sub> ) <sub>6</sub> Mo <sub>7</sub> O <sub>24</sub> ·4H <sub>2</sub> O	0.8 µM

mM = Millimole µM = Micromole. The various levels of Al<sup>3+</sup> were supplied in the form of Al<sub>2</sub>(SO<sub>4</sub>)<sub>3</sub>.

short comings that make them less suitable in the screening of soybean for aluminium stress tolerance compared to the sand culture. While hydroponics screening is restricted to the seedling stage (Villagarcia et al., 2001), field evaluations for aluminium stress tolerance is frequently affected by large coefficients of variation due to spatial variability (Urrea-Gomez et al., 1996), such that researchers exercise caution and describe acid tolerance instead of aluminium tolerance (Villagarcia et al., 2001). As a solid substrate that is almost inert and physically similar to the soil, experiments in the sand culture can be regulated with consistency (Villagarcia et al., 2001). Seeds could also be directly planted and roots completely excavated at harvest. Furthermore, aluminium stress tolerance screening studies in most crops, particularly soybean, are concentrated in the temperate/subtropical regions of the world where experiments are carried out in controlled environments. There is dearth of such information for the tropical environment. However, prior to any screening, there is the need to minimize cost, by determining the appropriate levels of aluminium activity that could best separate genotypes into sensitive and tolerant status. The objective of the research therefore, was to determine the appropriate levels of aluminium activity that could best separate soybean genotypes into sensitive and tolerant status with a view of recommending such levels for the rapid screening of tropically adapted soybeans for aluminium stress tolerance in sand culture.

## MATERIALS AND METHODS

### Experimental layout and procedure

In order to determine the level of aluminium activity that will appropriately characterize soybean genotypes for aluminium stress tolerance in sand culture, fifteen varieties of soybeans were

selected and grown in sand culture in October/November of 2001 and repeated in 2002 within the same period. The fifteen varieties have been previously described (Ojo, 2010; Ojo and Bello, 2010). A split-plot design comprising the 15 genotypes as the main plots and eight (8) levels of aluminium activity (0, 50, 100, 200, 300, 400, 450, and 500 µMAl<sup>3+</sup>) as the subplots was employed in the experiment. The experiment was replicated three times and was carried out at the College of Agronomy Teaching and Research Farm, University of Agriculture, Makurdi, Nigeria. Makurdi is situated on Latitude 7° 41'N and Longitude 8°37'E.

The experimental procedure was according to Villagarcia et al. (2001), with some modification. Imposition of aluminium treatment commenced at seven days after planting (7 DAP) as against 3 DAPS used by Villagarcia et al. (2001). Hence, data were taken at 25 DAP as against 21 DAP in the previous work (Villagarcia et al., 2001). Polyethylene pots measuring about 20cm in diameter were each filled with 10 kg builders' grade sharp sand. The sand was flushed with deionised water adjusted to pH 4.05 ± 0.05. The sand was flushed again with deionised water adjusted to pH 7.0 to remove the acidity and allowed to drain for 24 h. The next day, the sand was heavily watered with deionised water and six seeds were planted in each bag and lightly covered with the sharp sand. The pots were then watered daily with deionised water (pH 7.0). After five days, the emerged seedlings were thinned to three/pot while the watering with deionised water continued till the seventh day. Thereafter, nutrient solution with the various levels of aluminium activity were used to water the pots for the next eighteen days, with each pot receiving one litre of solution per day. To avoid a build up of nutrients, each pot was flushed daily with deionised water (pH 4.05 ± 0.05) prior to watering with the nutrient solution. Every morning, the pots were flushed and a time lag of two hours was allowed for the pots to drain before applying the nutrient solution. Nutrient stock solution concentration was developed following the procedure of Howell and Bernard (1961) and Villagarcia et al. (2001) (Table 1). At 1 L/pot, the volume of solution required per replicate was 120 litres. Thus, 120 L of deionized water was divided into eight equal parts of 15 L each and used to prepare nutrient solution with a particular level of aluminium activity. In each replicate, each of the eight batches of 15 L corresponds to each of the eight levels of aluminium activity (subplots). To prepare nutrient solution containing a particular level of aluminium, each of the 15 L of deionised water was divided into three equal parts of 5 L each. The first part was used to prepare the nutrient stock solution required for 15 L. This nutrient stock solution was subsequently adjusted to pH 4.05±0.05. The second part was adjusted to pH 4.05 ± 0.05 and used to prepare aluminium stock solution for a particular level of aluminium activity that was required for 15 L. Aluminium in the form of Al<sub>2</sub>(SO<sub>4</sub>)<sub>3</sub> was used to achieve the desired aluminium activity. The third part was adjusted to pH 4.05 ± 0.05 and kept to be used for making up the solution to 15 L. Thereafter, the aluminium stock solution (second part) was poured into the nutrient stock solution (first part) and the solution was made up to 15 L by the third part (deionised water).

### Data recording and analysis

Plants in each pot were harvested at 25 days old that is, 25 days after planting (25 DAP) and data were collected on root dry weight (RDW), shoot dry weight (SDW) and relative root surface area (RRSA). Plants were carefully removed from the sand after tearing the bags to loosen the sand. Roots were then immersed in water and the adhering sand washed off the roots with deionised water. Thereafter, the roots were separated from the shoot and subsequently spread on net trays and air dried for 30 min to drain off water prior to data collection. Data on the RRSA was then taken using the gravimetric method of Carley and Watson (1966). To prepare the solution for the estimation of RRSA, Ca(NO<sub>3</sub>)<sub>2</sub> was

**Table 2.** Mean squares for root and shoot characteristics of 15 tropically adapted soybean genotypes screened at seven levels of aluminium activity (0, 50, 100, 200,300, 400 and 450  $\mu\text{MAI}^{3+}$ ) in acid sand culture for 2 years.

Source of variation	Df	Root dry weight (g)	Shoot dry weight	Relative root surface area
Years(Y)	1	0.000125	0.000115	0.000237
Reps /year	4	0.000060	0.000035	0.002158
Aluminium(Al)	6	0.085359**	0.702708**	48.791847**
Genotype(G)	14	0.666376**	1.337467**	58.891269**
Y $\times$ Al	6	0.000041	0.000006	0.000543
G $\times$ Y	14	0.000056	0.000022	0.006858
G $\times$ Al	84	0.003312**	0.028973**	1.540466**
G $\times$ Al $\times$ Y	84	0.000036	0.000031	0.002863
Error	416	0.000040	0.002450	0.004839

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

**Table 3.** Mean root dry weight, shoot dry weight and relative root surface area of soybean genotypes in response to aluminium treatment in acid sand culture.

Aluminium level ( $\mu\text{MAI}^{3+}$ )	Root dry weight (g)	Shoot dry weight (g)	Relative root surface area (g)
0	0.383	0.771	6.434
50	0.353	0.662	5.628
100	0.339	0.632	5.338
200	0.327	0.596	5.030
300	0.315	0.567	4.790
400	0.302	0.539	4.570
450	0.290	0.507	4.222
LSD <sub>0.05</sub>	0.058	0.054	0.212

dissolved in a beaker of deionised warm water (60°C) in the ratio of 1: 6 by weight of  $\text{Ca}(\text{NO}_3)_2$  to water and allowed to cool down to room temperature. Thereafter, the beaker of the viscous  $\text{Ca}(\text{NO}_3)_2$  solution was placed on a digital analytic balance and roots of each plant immersed in it for 10 s. The roots were then lifted above the beaker and allowed to drain into the beaker for 20 s. The weight of the solution after immersion of roots was subtracted from the weight of solution before immersion of roots and was taken as the RRSA. The RRSA thereafter represents the weight of  $\text{Ca}(\text{NO}_3)_2$  solution adhering to the root surfaces. The roots were immediately rinsed in deionised water to remove the  $\text{Ca}(\text{NO}_3)_2$  and air dried again before oven drying. Roots were oven dried at 70°C for 48 h to a constant weight and their weights were taken as the root dry weight (RDW). Shoots were similarly oven dried to a constant weight and their respective weights taken as the shoot dry weight (SDW).

Data collected were subjected to analysis of variance and mean separation using the General Linear Model (GLM) and the Analysis of Variance (ANOVA) procedures SAS (1990). Mean values were used to rank genotypes within each level of aluminium activity for a particular trait. The highest mean was ranked 1<sup>st</sup> while the lowest mean was ranked 15<sup>th</sup>.

## RESULTS

Plants watered with nutrient solution containing 500  $\mu\text{MAI}^{3+}$  activity got burnt after 3 to 5 days of watering and

were deleted from the experiment. Consequently, no result was available for the 500  $\mu\text{MAI}^{3+}$  level of aluminium activity. Mean squares for root dry weight, shoot dry weight and relative root surface area of 15 soybean genotypes watered with nutrient solution adjusted to seven levels of aluminium activity (0, 50, 100, 200, 300, 400 and 450  $\mu\text{MAI}^{3+}$ ) are summarized in Table 2. Highly significant aluminium, genotype and genotype  $\times$  aluminium interaction effects were observed for the root dry weight, shoot dry weight and relative root surface area. No significant effects of years, rep/years, aluminium  $\times$  year, genotype  $\times$  year and genotype  $\times$  aluminium  $\times$  year were observed for all the traits (root dry weight, shoot dry weight and the relative root surface area).

Mean separation for the 7 levels of aluminium activity are presented in Table 3. A consistent decrease in means was observed for each of the traits as aluminium activity increased from 0 to 450  $\mu\text{MAI}^{3+}$ . Significant difference in aluminium activity for the root dry weight was only observed between the control and the highest levels of aluminium activity (300, 400 and 450  $\mu\text{MAI}^{3+}$ ). Shoot dry weight was more sensitive to changes in the levels of aluminium activity than the root dry weight. Shoot dry weight mean for each of the level of aluminium

**Table 4.** Root dry weight (g plant<sup>-1</sup>) and genotypic ranking (superscript) of 15 soybean genotypes grown at seven levels of aluminium activity (0, 50, 100, 200, 300, 400 and 450 µMAl<sup>3+</sup>) in acid sand culture.

Genotype	0	50	100	200	300	400	450
TGX 1740-2E	0.310 <sup>10</sup>	0.244 <sup>14</sup>	0.236 <sup>14</sup>	0.230 <sup>14</sup>	0.224 <sup>14</sup>	0.214 <sup>14</sup>	0.210 <sup>14</sup>
TGX 1485-1D	0.500 <sup>3</sup>	0.446 <sup>3</sup>	0.406 <sup>3</sup>	0.364 <sup>3</sup>	0.324 <sup>4</sup>	0.296 <sup>4</sup>	0.274 <sup>4</sup>
TGX 1830-20E	0.300 <sup>13</sup>	0.296 <sup>10</sup>	0.264 <sup>12</sup>	0.254 <sup>13</sup>	0.234 <sup>13</sup>	0.220 <sup>13</sup>	0.214 <sup>12</sup>
TGX 1876-4E	0.310 <sup>10</sup>	0.300 <sup>8</sup>	0.280 <sup>10</sup>	0.264 <sup>11</sup>	0.236 <sup>12</sup>	0.226 <sup>12</sup>	0.216 <sup>11</sup>
TGX 1805-31F	0.350 <sup>6</sup>	0.334 <sup>5</sup>	0.314 <sup>6</sup>	0.296 <sup>6</sup>	0.280 <sup>6</sup>	0.270 <sup>7</sup>	0.264 <sup>6</sup>
TGX 1873-16E	0.300 <sup>13</sup>	0.290 <sup>12</sup>	0.284 <sup>9</sup>	0.280 <sup>7</sup>	0.276 <sup>7</sup>	0.274 <sup>6</sup>	0.274 <sup>4</sup>
TGX 1878-7E	0.366 <sup>5</sup>	0.360 <sup>4</sup>	0.360 <sup>4</sup>	0.354 <sup>4</sup>	0.354 <sup>3</sup>	0.346 <sup>3</sup>	0.344 <sup>3</sup>
TGX 1802-1F	0.330 <sup>8</sup>	0.314 <sup>7</sup>	0.296 <sup>7</sup>	0.274 <sup>9</sup>	0.257 <sup>10</sup>	0.246 <sup>9</sup>	0.236 <sup>9</sup>
TGX 1891-3F	0.410 <sup>4</sup>	0.300 <sup>8</sup>	0.276 <sup>11</sup>	0.276 <sup>8</sup>	0.254 <sup>11</sup>	0.236 <sup>11</sup>	0.214 <sup>12</sup>
TGX 1896-3F	0.700 <sup>1</sup>	0.686 <sup>1</sup>	0.686 <sup>1</sup>	0.674 <sup>1</sup>	0.674 <sup>1</sup>	0.664 <sup>1</sup>	0.654 <sup>1</sup>
TGX 1844-18E	0.600 <sup>2</sup>	0.576 <sup>2</sup>	0.574 <sup>2</sup>	0.564 <sup>2</sup>	0.556 <sup>2</sup>	0.546 <sup>2</sup>	0.546 <sup>2</sup>
TGX 1440-1E	0.274 <sup>15</sup>	0.270 <sup>13</sup>	0.264 <sup>12</sup>	0.264 <sup>11</sup>	0.260 <sup>9</sup>	0.246 <sup>9</sup>	0.246 <sup>8</sup>
TGX 1448-2E	0.310 <sup>10</sup>	0.296 <sup>10</sup>	0.286 <sup>8</sup>	0.274 <sup>9</sup>	0.274 <sup>8</sup>	0.270 <sup>7</sup>	0.256 <sup>7</sup>
TGX 1895-35F	0.350 <sup>6</sup>	0.334 <sup>5</sup>	0.330 <sup>5</sup>	0.324 <sup>5</sup>	0.316 <sup>5</sup>	0.296 <sup>4</sup>	0.224 <sup>10</sup>
TGX 923-2E	0.330 <sup>8</sup>	0.244 <sup>14</sup>	0.230 <sup>15</sup>	0.210 <sup>15</sup>	0.204 <sup>15</sup>	0.186 <sup>15</sup>	0.184 <sup>15</sup>
Mean	0.383	0.353	0.339	0.327	0.315	0.302	0.290
LSD <sub>0.05</sub>	0.016	0.008	0.011	0.006	0.011	0.009	0.008

\*Superscript indicates genotypic ranking from the highest (1<sup>st</sup>) to the lowest (15<sup>th</sup>).

**Table 5.** Shoot dry weight (g plant<sup>-1</sup>) and genotypic ranking (superscript) of 15 soybean genotypes grown at seven levels of aluminium activity (0, 50, 100, 200, 300, 400 and 450 µMAl<sup>3+</sup>) in acid sand culture.

Genotype	0	50	100	200	300	400	450
TGX 1740-2E	0.643 <sup>10</sup>	0.587 <sup>12</sup>	0.553 <sup>9</sup>	0.512 <sup>8</sup>	0.480 <sup>7</sup>	0.450 <sup>7</sup>	0.353 <sup>11</sup>
TGX 1485-1D	0.562 <sup>14</sup>	0.500 <sup>14</sup>	0.453 <sup>15</sup>	0.422 <sup>15</sup>	0.380 <sup>15</sup>	0.380 <sup>14</sup>	0.360 <sup>10</sup>
TGX 1830-20E	0.632 <sup>11</sup>	0.587 <sup>12</sup>	0.553 <sup>9</sup>	0.500 <sup>12</sup>	0.450 <sup>11</sup>	0.390 <sup>13</sup>	0.328 <sup>14</sup>
TGX 1876-4E	0.672 <sup>8</sup>	0.622 <sup>5</sup>	0.563 <sup>8</sup>	0.508 <sup>11</sup>	0.472 <sup>9</sup>	0.420 <sup>10</sup>	0.370 <sup>9</sup>
TGX 1805-31F	1.017 <sup>3</sup>	0.818 <sup>3</sup>	0.752 <sup>3</sup>	0.683 <sup>4</sup>	0.620 <sup>4</sup>	0.560 <sup>5</sup>	0.532 <sup>6</sup>
TGX 1873-16E	0.602 <sup>13</sup>	0.600 <sup>6</sup>	0.600 <sup>5</sup>	0.600 <sup>5</sup>	0.590 <sup>5</sup>	0.590 <sup>4</sup>	0.567 <sup>4</sup>
TGX 1878-7E	0.608 <sup>12</sup>	0.600 <sup>6</sup>	0.600 <sup>5</sup>	0.600 <sup>5</sup>	0.560 <sup>6</sup>	0.560 <sup>5</sup>	0.560 <sup>5</sup>
TGX 1802-1F	0.670 <sup>9</sup>	0.600 <sup>6</sup>	0.552 <sup>11</sup>	0.510 <sup>9</sup>	0.450 <sup>11</sup>	0.402 <sup>11</sup>	0.350 <sup>12</sup>
TGX 1891-3F	1.040 <sup>2</sup>	0.600 <sup>6</sup>	0.542 <sup>13</sup>	0.500 <sup>12</sup>	0.452 <sup>10</sup>	0.452 <sup>7</sup>	0.440 <sup>7</sup>
TGX 1896-3F	1.120 <sup>1</sup>	1.097 <sup>1</sup>	1.097 <sup>1</sup>	1.090 <sup>1</sup>	1.090 <sup>1</sup>	1.080 <sup>1</sup>	1.078 <sup>1</sup>
TGX 1844-18E	0.945 <sup>4</sup>	0.877 <sup>2</sup>	0.877 <sup>2</sup>	0.875 <sup>2</sup>	0.872 <sup>2</sup>	0.872 <sup>2</sup>	0.872 <sup>2</sup>
TGX 1440-1E	0.522 <sup>15</sup>	0.498 <sup>15</sup>	0.483 <sup>14</sup>	0.450 <sup>14</sup>	0.420 <sup>14</sup>	0.380 <sup>14</sup>	0.312 <sup>15</sup>
TGX 1448-2E	0.732 <sup>7</sup>	0.600 <sup>6</sup>	0.583 <sup>7</sup>	0.550 <sup>7</sup>	0.480 <sup>7</sup>	0.402 <sup>11</sup>	0.332 <sup>13</sup>
TGX 1895-35F	0.903 <sup>5</sup>	0.748 <sup>4</sup>	0.720 <sup>4</sup>	0.712 <sup>3</sup>	0.700 <sup>3</sup>	0.700 <sup>3</sup>	0.690 <sup>3</sup>
TGX 923-2E	0.903 <sup>5</sup>	0.598 <sup>11</sup>	0.552 <sup>11</sup>	0.510 <sup>9</sup>	0.450 <sup>11</sup>	0.422 <sup>9</sup>	0.408 <sup>8</sup>
Mean	0.770	0.662	0.632	0.601	0.564	0.537	0.353
LSD <sub>0.05</sub>	0.012	0.011	0.011	0.010	0.011	0.013	0.009

\*Superscript indicates genotypic ranking from the highest (1<sup>st</sup>) to the lowest (15<sup>th</sup>).

activity was at least significantly different from five other levels. The relative root surface area was very sensitive to changes in the level of aluminium activity. Relative root surface area for each of the level of aluminium activity (including the control) was significantly different from all the other levels.

Mean separation for the 15 soybean genotypes at each of the seven levels of aluminium activity are summarized in Tables 4, 5 and 6. Root dry matter accumulation was lowest in TGX 923 – 2E and highest in TGX 1896 – 3F for all levels of aluminium activity except in the control (0 µMAl<sup>3+</sup>) where the lowest root dry weight was observed

**Table 6.** Relative root surface area ( $\text{g plant}^{-1}$ ) and genotypic ranking (superscript) of 15 soybean genotypes grown at seven levels of aluminium activity (0, 50, 100, 200, 300, 400 and 450  $\mu\text{MAI}^{3+}$ ) in acid sand culture.

Genotype	0	50	100	200	300	400	450
TGX 1740-2E	5.540 <sup>10</sup>	4.500 <sup>14</sup>	4.183 <sup>14</sup>	3.600 <sup>15</sup>	3.308 <sup>15</sup>	3.198 <sup>15</sup>	3.058 <sup>14</sup>
TGX 1485-1D	4.547 <sup>15</sup>	4.007 <sup>15</sup>	3.855 <sup>15</sup>	3.608 <sup>14</sup>	3.408 <sup>14</sup>	3.302 <sup>13</sup>	3.100 <sup>13</sup>
TGX 1830-20E	5.370 <sup>12</sup>	5.000 <sup>11</sup>	4.643 <sup>12</sup>	4.250 <sup>12</sup>	3.808 <sup>12</sup>	3.402 <sup>12</sup>	3.018 <sup>15</sup>
TGX 1876-4E	5.985 <sup>9</sup>	5.480 <sup>7</sup>	4.995 <sup>7</sup>	4.503 <sup>9</sup>	4.215 <sup>9</sup>	3.598 <sup>11</sup>	3.282 <sup>10</sup>
TGX 1805-31F	7.540 <sup>4</sup>	6.015 <sup>6</sup>	5.435 <sup>6</sup>	4.900 <sup>5</sup>	4.337 <sup>7</sup>	3.812 <sup>9</sup>	3.568 <sup>8</sup>
TGX 1873-16E	5.347 <sup>13</sup>	5.010 <sup>9</sup>	4.880 <sup>8</sup>	4.880 <sup>6</sup>	4.870 <sup>5</sup>	4.870 <sup>5</sup>	4.870 <sup>5</sup>
TGX 1878-7E	7.580 <sup>3</sup>	7.580 <sup>1</sup>	7.580 <sup>1</sup>	7.570 <sup>1</sup>	7.570 <sup>1</sup>	7.560 <sup>1</sup>	5.930 <sup>3</sup>
TGX 1802-1F	5.482 <sup>11</sup>	5.100 <sup>8</sup>	4.822 <sup>9</sup>	4.360 <sup>11</sup>	4.125 <sup>11</sup>	3.632 <sup>10</sup>	3.170 <sup>11</sup>
TGX 1891-3F	5.090 <sup>14</sup>	4.870 <sup>13</sup>	4.648 <sup>11</sup>	4.403 <sup>10</sup>	4.192 <sup>10</sup>	4.168 <sup>6</sup>	3.970 <sup>6</sup>
TGX 1896-3F	7.260 <sup>5</sup>	7.000 <sup>3</sup>	7.000 <sup>2</sup>	6.950 <sup>2</sup>	6.950 <sup>2</sup>	6.940 <sup>2</sup>	6.940 <sup>1</sup>
TGX 1844-18E	7.250 <sup>6</sup>	6.890 <sup>4</sup>	6.850 <sup>3</sup>	6.800 <sup>3</sup>	6.800 <sup>3</sup>	6.700 <sup>3</sup>	6.670 <sup>2</sup>
TGX 1440-1E	7.642 <sup>2</sup>	4.918 <sup>12</sup>	4.720 <sup>10</sup>	4.522 <sup>8</sup>	4.357 <sup>6</sup>	4.160 <sup>7</sup>	3.440 <sup>9</sup>
TGX 1448-2E	6.643 <sup>8</sup>	6.040 <sup>5</sup>	5.445 <sup>5</sup>	4.840 <sup>7</sup>	4.283 <sup>8</sup>	3.922 <sup>8</sup>	3.652 <sup>7</sup>
TGX 1895-35F	8.078 <sup>1</sup>	7.003 <sup>2</sup>	6.517 <sup>4</sup>	6.167 <sup>4</sup>	6.125 <sup>4</sup>	6.050 <sup>4</sup>	5.560 <sup>4</sup>
TGX 923-2E	7.157 <sup>7</sup>	5.002 <sup>10</sup>	4.500 <sup>13</sup>	4.000 <sup>13</sup>	3.500 <sup>13</sup>	3.233 <sup>14</sup>	3.112 <sup>12</sup>
Mean	6.434	5.628	5.472	5.024	4.790	4.570	4.222
LSD <sub>0.05</sub>	0.015	0.060	0.048	0.118	0.081	0.032	0.018

\*Superscript indicates genotypic ranking from the highest (1<sup>st</sup>) to the lowest (15<sup>th</sup>).

for TGX 1440 – 2E (Table 4). There was near consistency in genotypic ranking (superscript) among the six levels of aluminium activity (50 to 450  $\mu\text{MAI}^{3+}$ ). Two varieties, namely, TGX 1896 – 3F and TGX 1844 – 18E consistently ranked first and second in root dry matter accumulation across all the levels of aluminium activity including the control. Aluminium stress tolerance increased with increasing aluminium activity in four genotypes (TGX 1873 – 16E, TGX 1878 – 7E, TGX 1440 – 1E and TGX 1448 – 2E), while TGX 1891 – 3F increased in sensitivity to aluminium stress as aluminium activity increased.

The trend in genotypic response to aluminium stress for shoot dry weight is however different from that observed for the root dry weight (Table 5). Shoot dry matter accumulation was lowest in TGX 1485 – 1D (0, 50 and 450  $\mu\text{MAI}^{3+}$ )/TGX 1440 – 1E (100, 200, 300 and 400  $\mu\text{MAI}^{3+}$ ) and highest in TGX 1896 – 3F across all the levels of aluminium activity. While TGX 1896 – 3F consistently ranked highest in shoot dry matter accumulation at all levels of aluminium activity, a noticeable change in rank was observed between the control (0  $\mu\text{MAI}^{3+}$ ) and the six levels of aluminium treatment (50, 100, 200, 300, 400 and 450  $\mu\text{MAI}^{3+}$ ) for most of the genotypes. The variety TGX 1844 – 18E changed rank from 4<sup>th</sup> in the control to 2<sup>nd</sup> across the six levels of aluminium activity (50, 100, 200, 300, 400 and 450  $\mu\text{MAI}^{3+}$ ). All other genotypes were inconsistent in ranking except three (TGX 1873 – 16E, TGX 1878 – 7E and TGX 1895 – 35F). Aluminium stress tolerance in these three genotypes increased with increasing aluminium activity.

No consistent agreement in genotypic ranking was observed among all the levels of aluminium activity (including the control) for the relative root surface area (Table 6). The most conspicuous change in genotypic ranking was observed between the control (0  $\mu\text{MAI}^{3+}$ ) and the 50  $\mu\text{MAI}^{3+}$  level of aluminium activity. Two varieties, namely, TGX 1896 – 3F and TGX 1844 – 18E changed rank from 5<sup>th</sup> and 6<sup>th</sup> in the control (0  $\mu\text{MAI}^{3+}$ ) to 3<sup>rd</sup> and 4<sup>th</sup> at the 50  $\mu\text{MAI}^{3+}$  and to 1<sup>st</sup> and 2<sup>nd</sup> at the highest level of aluminium activity (450  $\mu\text{MAI}^{3+}$ ) respectively. Aluminium stress tolerance increased with increasing aluminium activity in four genotypes (TGX 1873 – 16E, TGX 1891 – 3F, TGX 1896 – 3F and TGX 1844 – 18E), while TGX 1830 – 20E, TGX 1805 – 31F and TGX 1895 – 35F increased in sensitivity to aluminium stress as aluminium activity increased.

## DISCUSSION

The highly significant aluminium effect observed for all the traits (root dry weight, shoot dry weight and relative root surface area) in the current work is consistent with the previous observation (Villagarcia et al., 2001). Villagarcia et al. (2001) studied the genotypic response of ten varieties of soybean at two levels of aluminium activity and observed a highly significant aluminium effect. The change in genotypic ranking observed between the control (0  $\mu\text{MAI}^{3+}$ ) and other levels of aluminium activity for root dry weight, shoot dry weight and relative root surface area in the current work is not due to acid stress, but the presence of phytotoxic

elements in the media. Such phenomenon had been previously observed (Foy et al., 1969; Ermolayev et al., 2003) and attributed to aluminium toxicity. Foy et al. (1969) investigated aluminium tolerance of 'Chief' and 'Perry' varieties of soybean and observed similar root growth in both varieties prior to the imposition of aluminium treatment. However, significant differences in the growth of the roots of the two varieties (Perry and Chief) in response to aluminium stress were observed, leading to their being classified as tolerant and intolerant. Ermolayev et al. (2003) in their comparison of Al-induced gene expression in sensitive (Malabar) and tolerant (Tambora) soybean cultivars observed visible morphological differences in the lateral roots of the two varieties due to aluminium treatment.

The highly significant genotypic differences observed for all the traits (root dry weight, shoot dry weight and relative root surface area) in the current work is an indication of genetic diversity in the soybean population studied. The highly significant genotype x aluminium interaction observed for all the traits is an indication of genotypic variation in response to the imposition of aluminium stress. The wider range in sensitivity for root dry weight, shoot dry weight and relative root surface area observed in the present study compared to the work of Villagarcia et al. (2001) could be due to differences in the soybean genotypes studied. The soybean genotypes studied in the current work are tropically adapted while the populations studied in the previous work are of temperate origin.

The significant difference between the control and any of the three highest levels of aluminium activity (300, 400 and 450  $\mu\text{MAl}^{3+}$ ) observed for the root dry weight in the current study, is an indication that any of the three levels could be selected with the control (0  $\mu\text{MAl}^{3+}$ ) in the screening of tropically adapted genotypes of soybean for this trait. The significant difference between each level of aluminium activity and five other levels of aluminium activity observed for the shoot dry weight is an indication that only the 450  $\mu\text{MAl}^{3+}$  levels could be selected along with the control in the screening of genotypes for this trait. The significant difference between all the levels of aluminium activity and the change in genotypic ranking observed between all the levels of aluminium activity for relative root surface area indicates that no inference can be drawn on the tolerance status of the tropical soybean germplasm for these traits until they have been screened through all the levels of aluminium activity used in the current work. It also indicates that relative root surface area is the most sensitive trait in genotypic variation for aluminium stress tolerance in tropically adapted soybeans.

Therefore, aluminium activity at 300  $\mu\text{MAl}^{3+}$  could be selected along with the control (0  $\mu\text{MAl}^{3+}$ ) for root dry weight, while 450  $\mu\text{MAl}^{3+}$  along with the control (0  $\mu\text{MAl}^{3+}$ ) should be considered as appropriate for both the shoot dry weight and the relative root surface area in the rapid screening of tropically adapted soybean genotypes for aluminium stress tolerance in sand culture in the tropics. The relative root surface area was the most sensitive in discriminating between levels of aluminium activity and should be preferred in any selection programme.

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