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Screening of acid-tolerant hybrid Corn lines and parents using modified acid mineral soil

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Abstract. Screening of hybrid corn parent lines quickly and cheaply compared to hydroponic methods in the laboratory is needed to support the assembly of new high yielding varieties of acid tolerant corn. Addition of CaCO_3 as equivalent to 2 t/ha (normal soil), while 0.5 t/ha AlCl_3 (acidic). A total of 12 elite maize lines were tested based on relative root growth (RRG). Four sets of experiments were made, namely 2 sets normal soil media (pH 6.5) and 2 sets acid soil media (pH 4.3). Experimental design used in each experimental set was a randomized block with three replications. Results showed that rapid screening of hybrid maize parental lines could be carried out using modified soil acidity method by adding CaCO_3 and AlCl_3 to acid soil by measuring relative root growth (RRG) as the main variable. Corn obtained by genotypes No.80, MAL03x192, and Bisi-18 were classified as tolerant, MAL03x28, MAL03x182, MAL03x42, MAL03x44, MAL03x107, MAL03x115 were classified as moderately tolerant, while MAL03x100, MAL03x56, classified as sensitive. Screening of hybrid parent lines on acid soil modified to pH 4.3 (acidic) and normal pH 6.5 at seedling age 7 days after planting (dap) was more accurate than at seedling age of maize 14 dap.

1. Introduction

Acidic soils occupy about 50% of arable land in tropical and sub-tropical regions. Therefore the use of acid soils for the development of food crops is very potential, for example in Ukraine in 2020 buckwheat has been planted on acid land covering an area of 125,000 ha [1]. In Indonesia, acid soil covers more than 55% of the total marginal land. Utilization of acidic land for the development of food crops, especially corn, supports government policy through a program to expand agricultural areas. However, the development of food crops on acid dry land is still lagging behind due to limiting factors including low nutrient content, low pH, and aluminum toxicity. [2].

From the screening of 141 corn germplasm, it was found that 2 cultivars, namely HZ 85 and HZ 178, had the highest relative root growth, meaning that these cultivars were tolerant of high Al solubility [3]. Meanwhile, in Brazil, 17 maize cultivars were obtained from screening of 50 local varieties classified as Aluminum tolerant [4]. There are two cultivars, namely CMS 30 and CMS 36, have been released and used as gene sources for Al-tolerant hybrid maize in tropical regions around the world [5]. Acid-tolerant maize lines absorb less Al at the root tips than sensitive ones. Additive genetic variation and additive x environmental interactions are very important in cereals [6]. For each species, progeny testing replicated in various environments was the most reliable means of evaluation and for maize full-sib selection increased grain yield by 14% per cycle [7]. Some hybrid maize which is the descendents of crosses between inbred lines tolerant of acid soils showed high seed yields on acid



soils [8].

There were 58% of F1 single-crossed maize hybrids were heterotic for Aluminum tolerance, and 14 inbred lines were selected for Al tolerance based on relative root growth (RRG) [9]. Screening of corn plants against soil acidity can be assessed from the presence of phenolic compounds in plant tissues [10]. Aluminum toxicity in maize led to a significant yield reduction of 18.7% for the line and 14.7% for the hybrid during 2 years of cultivation [11]. Thirty three single cross hybrids were evaluated in acid soils and optimum soils in 11 locations for three seasons showed an average decrease in seed yield ranges from 11 to 37% due to low pH [12]. The importance of the bioavailability of phenolic compounds to get the physiological role, and the main factors affecting phenolic metabolism, from absorption to excretion in plants [13]. Chemical analysis of plant tissue to detect the total percent phenolic can be determined by means of Spectrophotometer and Chromatographic to identify and quantify phenolic compounds, and increasing the phenolic content greater than $10^3 \mu\text{g}$ proved to be very tolerant to Al stress [14].

To meet the increasing demand for corn, both domestically and for export, it is necessary to continue to strive to increase corn productivity. The use of acid dry land in Indonesia to increase maize productivity is a strategic choice in terms of land area potential, however, it is known that in acid land the main production limiting factor is soil acidity. For this reason, rapid screening or selection of hybrid maize parent lines that are tolerant to acid soils is required.

2. Methods

2.1. Soil sample

Screening Using PVC pipe pots of lines and genotypes of acid-tolerant corn was carried out in the Greenhouse of the Cereal Crops Research Institute in Maros from June to August 2021. The acid soil used was taken from the Bontobili Food Crops Research Installation (IP2TP), Gowa Regency, South Sulawesi Province, at a depth of 0-20 cm. Soil samples were weighed (1.4 kg) in a plastic bag and separated into two groups with half (0.7 kg) mixed CaCO_3 at a dose of 2 t/ha to a target pH of 6.5 and in the other half added AlCl_3 at a dose equivalent of 0.5 t/ha to a target pH around of 4.3. Soil on each bag was then put into a PVC pipe pot measuring 19 cm high and 9 cm in diameter, with a bottom cover with 4 little holes perforated to maintain soil drainage in the pipe tube. Soil in PVC was incubated for 4 weeks in a greenhouse with wet and dry cycles. All media in the PVC was watered to 100% field capacity (FC) by initial weight, dried to about 70% FC and then watered again to about 80% FC during incubation. The aim of the 4-week incubation is to allow lime and soil to react and reach the target pH, and to reduce the risk of Mn poisoning in plants.

2.2. Corn genotype

The hybrid parent corn genotype used in the screening experiment is a collection of selected acid-tolerant genetic material research in the 2021 with Bisi-18 (Al-tolerant) hybrids as a control. A total of 7 to 10 seeds of each genotype of corn were grown on moist tissue paper for 1 x 24 hours, then three uniform seeds were selected to be planted in each PVC media with 1 plant with a hole depth of 3 cm. The PVC pots were then placed in a controlled environment greenhouse and watered with 80% FC regularly. The experiment was designed according to a randomized block design with three replications. The experiment was made in 4 sets, two sets using AlCl_3 with a target pH of 4.3 and two sets using lime (CaCO_3) with a target pH of 6.5. Each experimental medium was observed at the age of corn seedlings 7 and 14 days after planting (dap).

2.3. Observation

Observations made were relative root growth, root fresh weight, root dry weight, number of leaves, leaf width, plant length, plant fresh weight, and plant dry weight at 7 and 14 days after planting. To determine the relative root length (RRG) by formula = the relative root length of lime treatment minus

the relative acid root length divided by the relative root length of the lime multiplied by 100% [15]. Analysis of variance was performed using the Statistical Tool of Agricultural Research (STAR) procedure.

Basic fertilization using Grow more 1 g/plant dissolved in 250 ml of distilled water before application at 5 days after planting (dap). The nutrient content (%) of Grow more fertilizer used every 100 g is: nitrogen(N) 32.0, Phosphorus (P) 10.0, Calcium (Ca) 0.05, Sulfur (S) 0.20, Boron (B) 0.20, Copper (Cu) 0.05, Iron (Fe) 0.10, Manganese (Mn) 0.05. Zinc (Zn) 0.05, Molybdenum (Mo) 0.005. The lime used was agricultural lime from a limestone kiln containing about 21% CaO. Lime applied 1 month before planting at a dose equivalent of 2 t/ha CaO per pot as recommended by Hakim and Upadhyaya [15] that soil with a pH < 5.5 requires lime 2.7 t/ha. Meanwhile, according to Baquy, et al. [16] that low soil pH and Al toxicity significantly reduced plant growth, chlorophyll content, and plant biomass.

2.4. Acidic soil used in the experiment

Acid soil was taken of the Food Crops Agricultural Research Installation (IP2TP) Bontobili Village, Gowa Rgency, South Sulawesi Province. Soil textured clay loam, reacted acid, very low organic matter content, low nutrient content of N, very low P, and medium K, Medium base and cation exchange capacity can be ranged from low to high, low Al saturation and low Al₂O₃ (Table 1).

Table 1. Acid soil analysis at the experimental site Agricultural Food Crops Research Installation of Bontobili.

Soil Properties	IP2TP Bontobili	
	Value	Criteria
Texture		Clay loam
Sand (%)	46	
Ash (%)	26	
Silt (%)	28	
OM (%)	0.12	Very low
C (%)	1.70	Low
N (%)	0.12	Low
P (%)	14	High
C/N	14	Medium
P ₂ O ₅ HCl 25% (me/100g)	114	Very high
P ₂ O ₅ Bray (ppm)	7	Very low
K ₂ O HCl 25% (mg/g)	34	Medium
CEC (me/100g)	32.77	Medium
Cation		
K(me/100g)	0.15	Low
Na(me/100g)	0.20	Low
Mg(me/100g)	2.00	Medium
Ca(me/100g)	8.97	Medium
Base saturation(%)	38.0	Medium
Al Saturation (%)	23.02	Low
pH (H ₂ O)	5.09	Acid
pH (KCl)	4.30	
Al ₂ O ₃	22.56	Low

Analyzed at Laboratory of Assesment Institute For Agriculture Technology South Sulawesi 2021

2.5. Modified acidic soil to pH 6.5 at the seed age of 7 days after planting

Modification of the original soil that reacts from acid to neutral at a pH of about 6.5 is done by adding lime (CaCO_3) equivalent to 2 t/ha. The experimental results showed that the response of several parental lines of maize hybrid to liming at the age of 7 days after planting (dap) significantly affected plant height, root length, root fresh weight, root dry weight, plant weight root ratio (S/R), plant fresh weight and relative root growth (RRG), but had no significant effect on plant dry weight (Table 2). According to Kar et al., [17] that high levels of Al^{3+} in the soil interfere with the function of transporting potassium (K^+), calcium (Ca^{2+}), magnesium (Mg^{2+}), nitrate (NO_3^-), and ammonium (NH_4^+) in plants. On the other hand, that using Arbuscular Mycorrhiza Fungi (AMF) inoculum mitigated Al phytotoxicity with the chemical inhibitor glomalin being able to absorb Al beyond the root surface, which is an important property of AMF for acid soil management strategies. [18].

Table 2. Responses to several genotypes of maize grown at 7 days seedling age in normal soil pH 6.5

Lines, Genotype	PH (cm)	RL (cm)	FWR (g)	DWR (g)	S/R	PFW (g)	PDW (g)
No.80	30.83 d	32.57 ab	1.30 ab	0.12 b	0.97 a	1.10 abc	0.11 ^{ns}
MAL03x28	21.17 ab	21.20 defg	0.67 bc	0.13 ab	0.36 b	0.46 ef	0.05
MAL03x182	21.25 ab	22.93 cdef	1.30 ab	0.20 a	0.32 b	0.64 cdef	0.06
MAL03x42	18.67 bc	23.40 cdef	1.27 ab	0.12 ab	0.61 ab	0.56 def	0.07
MAL03x44	22.77 ab	26.01 bcd	0.67 bc	0.14 ab	0.74 ab	0.57 def	0.07
MAL03x56	24.83 ab	15.03 fg	1.07 abc	0.18 ab	0.65 ab	1.02 bcd	0.13
MAL03x100	20.90 ab	15.97 efg	0.87 abc	0.15 ab	0.57 ab	0.45 ef	0.07
MAL03x107	29.90 a	30.17 abc	1.43 a	0.15 ab	0.67 ab	1.40 ab	0.08
MAL03x115	19.01 bc	24.16 cde	1.37 a	0.14 ab	0.56 ab	1.61 a	0.07
MAL03x124	9.67 c	13.33 g	0.47 c	0.10 b	0.53 ab	0.33 f	0.05
MAL03x192	19.27 ab	33.17 a	1.43 a	0.13 ab	0.75 ab	1.01 bcd	0.10
Bisi-18	29.17 ab	31.20 abc	1.10 abc	0.11 b	0.90 a	0.94 bcde	0.10
CV	15.71	12.01	20.07	19.02	19.40	20.71	31.13

Note. PH= Plant Height; RL= Root Length; FWR= Fresh Weight of Roots; DWR= Dry Weight of Root; S/R= Ratio of plants to roots; PFW= Plant Fresh Weight; PDW= Plant Dry Weight, RRG= Relative Root Growth

The correlation of several plant growth parameter under normal soil conditions (pH 6.5) showed that the correlation between relative root growth (RRG) with root length gave the highest value ($r^2=0.9388$, $P<0.05$), followed sequentially by RRG and plant height ($r^2=0.7481$, $P<0.05$) and plant dry weight with shoot and root ratio (S/R) ($r^2=0.7267$, $P<0.05$). This phenomenon shows that root length contributes the most to plant growth compared to other parameters (Table 3). The effect of liming on acid soils gave a 100% increase in root length density of wheat at a depth of 0–10 and 10–20 cm, and increased grain yield of wheat by more than 210% [19].

Table 3. Correlation of parameter on the growth of several varieties of acid tolerant corn at 7 days seedling age in normal soil pH 6.5

Parameters	PH	RL	FWR	RDW	S/R	PFW	DWP	RRG
PH	1							
RL	0.6847	1						
FWR	0.5711	0.5710	1					
RDW	0.0235	0.1809	0.3566	1				
S/R	0.6056	0.3991	0.2129	0.3235	1			
PFW	0.4872	0.4851	0.6677	0.0107	0.3966	1		
DWP	0.5316	0.2517	0.4544	0.2011	0.7267	0.4180	1	
RRG	0.7481	0.9388	0.5692	-0.0247	0.3829	0.4689	0.3396	1

Note: PH= Plant Height; RL= Root Length; FWR= Fresh Weight of Roots; RDW= Root Dry Weight; S/R=Shoot and Root Ratio; PFW= Plant Fresh Weight; DWP= Dry Weight of Plants; RRG= Root Relative Growth.

2.6 Modified acidic soil to pH 4.3 at the seed age of 7 days after planting

While the modification of the original soil which reacted acid from pH 5.1 to become more acidic at pH around 4.3 was carried out by adding aluminum chloride (AlCl_3) which was equivalent to 0.5 t/ha. The experimental results showed that the response of several parental maize hybrid lines at the age of 7 days after planting (dap) had a significant effect on plant height, root length, plant to root ratio (S/R), fresh plant weight, and relative root growth (RRG). Had no significant effect on root fresh weight, plant fresh weight, and plant dry weight (Table 4). When viewed from the RRG value, the genotypes N0.80, MAL03x192 and Bisi-18 were classified as acid tolerant by ($\text{RRG} > 30\%$), while the genotypes MAL03x28, MAL03x182, MAL03x42, MAL03x44, MAL03x107 were classified as medium tolerant ($\text{RRG} = 15\text{--}30\%$), whereas the genotypes MAL03x56, MAL03x115 and MAL03x124 were classified as sensitive ($\text{RRG} < 15\%$). This shows that a decrease in pH value or an increase in soil acidity greatly affects plant growth and root length as the highest contributor to plant growth (shoot). Aluminum exposure to maize seeds triggers changes in the transcript level for several genes, especially those related to cell wall structure and metabolism, oxidative stress response, membrane transporters, organic acid metabolism, signaling and hormones, all of which affect plant growth [20].

Table 4. Responses to several maize lines seedling aged 7 days after planting on acid soil pH 4.3

Lines, Genotype	PH(cm)	RL (cm)	FWR (g)	DWR (g)	S/R	PFW (g)	PDW (g)	RRG (%)
No.80	11.53 abcde	1.43 d	0.45 ^{ns}	0.11 ab	0.46 ab	0.33 ^{ns}	0.05 ^{ns}	33.6 ab
MAL03x28	16.27 ab	4.11 abc	0.47	0.12 ab	0.52 ab	0.36	0.05	21.2 bcd
MAL03x182	18.27 a	2.50 cd	0.58	0.14 a	0.70 a	0.62	0.10	22.9 abcd
MAL03x42	15.30 abc	4.97 ab	0.48	0.13 a	0.27 b	0.31	0.04	22.3 abcd
MAL03x44	10.73 abcde	2.96 cd	0.47	0.10 ab	0.53 b	0.28	0.04	29.2 ab
MAL03x56	5.50 de	1.90 d	0.48	0.11 ab	0.39 ab	0.15	0.03	15.0 de
MAL03x100	8.01 cde	1.27 d	0.40	0.07 b	0.38 ab	0.21	0.04	11.1 cde
MAL03x107	11.01 abcde	3.10 bcd	0.47	0.12 ab	0.34 b	0.37	0.04	28.4 abc
MAL03x115	11.20 abcde	5.63 a	0.49	0.16 a	0.31 b	0.45	0.04	24.2 abcd
MAL03x124	4.03 e	1.47 d	0.70	0.10 ab	0.38 ab	0.57	0.04	4.3 e
MAL03x192	12.50 abcd	2.23 cd	0.53	0.14 ab	0.40 ab	0.36	0.05	35.4 a
Bisi-18	9.17 bcde	3.13 bcd	0.51	0.10 ab	0.60 ab	0.22	0.08	34.5 a
CV.	15.11	21.81	22.8	17.91	21.36	28.53	29.55	17.9

Note: PH= Plant Height; RL= Root Length; FWR= Fresh Weight of Roots; DWR= Dry Weight of Root; S/R= Ratio of Plants to Roots; PFW= Plant Fresh Weight; PDW= Plant Dry Weight, RRG= Relative Root Growth

Meanwhile the correlation of several variables of plant growth under acidic conditions showed that the ratio of dry weight of shoot and roots (S/R) to dry weight of plants gave the highest value ($r^2 = 0.8242$, $P < 0.05$), followed by sequential correlation of root fresh weight and plants fresh weight ($r^2 = 0.7544$, $P < 0.05$) and root length with root dry weight ($r^2 = 0.5947$, $P < 0.05$).

The phenomenon indicates that root length contributes more significantly to plant growth compared to other parameters. This is means by the largest contributor to plant growth switching from RRG with root length in normal soil (pH 6.5) to S:R variable with plant dry weight of acidic soil conditions in pH 4.3 (Table 5).

Table 5. Correlation of parameters on the growth of several genotypes of acid tolerant corn seedling age 7 days after planting on soil pH 4.3

Parameter	PH	RL	FWR	RDW	S/R	PFW	DWP	RRG
PH	1							
RL	0.4843	1						
FWR	-0.4030	0.1080	1					
RDW	0.3865	0.5943	0.1255	1				
S/R	0.3757	-0.1373	0.1435	-0.099	1			
PFW	0.3163	0.1071	0.7544	0.2012	0.3451	1		
DWP	0.5302	0.1328	0.2863	0.1924	0.8242	0.5038	1	
RRG	0.3859	0.2098	-0.2496	0.1631	0.2143	0.1108	0.3400	1

Note: PH= Plant Height; RL=Root Length; FWR= Fresh Weight of Roots; RDW = Root Dry Weight; S/R= Shoot and Root Ratio; PFW= Plant Fresh Weight; DWP= Dry Weight of Plants; RRG= Root Relative Growth

2.7. Modify acidic soil to pH 6.5 at the seedling age of 14 days after planting

Modification of the original soil that reacts from acid to neutral at a pH of about 6.5 is done by adding lime (CaCO_3) equivalent to 2 t/ha. The experimental results showed that the response of several parental maize hybrid lines to liming at the age of 14 days after planting (dap) had a significant effect on plant height, root length, dry root weight, plant fresh weight, plant dry weight, and relative root growth (RRG). Have no significant effect on the fresh weight of roots, and the ratio of plant weight to plant roots (S/R). When viewed from the RRG value, there are two genotypes that are acid tolerant, namely MAL03x56 and Bisi-18, the others are classified as medium and sensitive (Table 6). Exposure to aluminum (90 M AlCl_3) can reduce root length, number of seminal roots, seminal root length, number of lateral roots, and root dry weight by 28, 8, 19, 14 and 30%, respectively [21]. While type of lime did not significantly affect the yield of corn and corn does not respond to lime without fertilizer [22].

Table 6. Responses to several maize lines in normal soil pH 6.5 aged seedling 14 days after planting.

Lines, Genotype	PH(cm)	RL(cm)	FWR (g)	DWR (g)	S/R	PFW (g)	PDW (g)
No.80	35.53 ab	40.93 a	1.40 ns	0.173 ab	0.69 ns	1.30 ab	0.20 ab
MAL03x28	24.30 bc	32.03 a	1.67	0.163 ab	1.04	1.32 ab	1.53 ab
MAL03x182	24.01 abc	41.47 a	2.53	0.100 b	0.90	0.54 b	0.06 b
MAL03x42	30.17 abc	40.87 a	1.77	0.120 b	1.24	1.42 ab	0.10 b
MAL03x44	30.13 abc	36.01 a	0.76	0.147 ab	0.60	0.93 ab	0.15 ab
MAL03x56	43.07 a	30.67 ab	1.80	0.103 b	1.70	2.34 a	0.18 ab
MAL03x100	33.43 bc	21.70 ab	1.27	0.127 ab	0.49	0.58 b	0.12 ab
MAL03x107	35.53 ab	37.50 a	1.83	0.153 ab	0.76	1.44 ab	0.18 ab
MAL03x115	20.52 cd	35.67 a	1.17	0.137 ab	0.54	0.24 b	0.05 b
MAL03x124	7.90 d	2.43 b	0.37	0.050 b	1.02	0.22 b	0.05 b
MAL03x192	34.13 abc	45.37 a	2.23	0.153 b	1.04	0.16 ab	0.15 ab
Bisi-18	42.37 a	46.23 a	6.83	0.316 a	1.43	2.18 a	0.33 a
CV.	17.12	23.80	22.31	18.45	25.47	25.46	19.49

Note: PH= Plant Height; RL= Root Length; FWR= Fresh Weight of Roots; DWR= Dry Weight of Root; S/R= Ratio of Plants to Roots; PFW= Plant Fresh Weight; PDW= Plant Dry Weight, RRG= Relative Root Growth Rate

The correlation of several plant growth variables at the age of 14 days after planting in normal soil conditions (pH 6.5) showed that the correlation between leaf length and plant height gave the highest value ($r^2 = 0.9255$, $P < 0.05$), followed by a sequential correlation between plant fresh weight and plant height ($r^2 = 0.7983$, $P < 0.05$) and plant dry weight with plant height ($r^2 = 0.7250$, $P < 0.05$). This phenomenon showed that root length and plant height contributed the most to plant growth compared to other parameters in normal soil at age seedling 14 dap (Table 7).

Table 7. Correlation of Parameters on the growth of several genotypes of acid-tolerant maize aged 14 days after planting in normal soil pH 6.5.

Parameter	PH	RL	FWR	RDW	PFW	DWP	S/R	LL	LW
PH	1								
RL	0.6201	1							
FWR	0.3362	0.3478	1						
RDW	0.5263	0.4892	0.3391	1					
PFW	0.7983	0.3659	0.3854	0.4593	1				
DWP	0.7250	0.3629	0.3461	0.6387	0.7496	1			
S/R	0.2660	0.0357	0.1066	0.0296	0.3520	0.3520	1		
LL	0.9255	0.6759	0.3997	0.4564	0.6614	0.6614	0.2603	1	
LW	0.5531	0.5745	0.1434	0.4622	0.4309	0.4309	-0.1208	0.4603	1

Note: PH= Plant Height; RL= Root Length; FWR= Fresh Weight of Roots; RDW = Root Dry Weight; S/R= Shoot and Root Ratio; PFW= Plant Fresh Weight; DWP= Dry Weight of Plants; RRG= Root Relative Growth; LL= Leaf Length; LW= Leaf Width

2.8. Modified acidic soil to pH 4.3 at the seed age of 14 days after planting

Meanwhile, modification of the original soil which reacted with acid to become more acidic at a pH of around 4.3 was carried out by adding aluminum chloride (AlCl_3) at a dose equivalent to 0.5 t/ha. The experimental results showed that the response of several parental lines of maize hybrid at the age of 14 (dap) had a significant effect on plant height, root length, and root relative growth (RRG), but had no significant effect on plant shoot-to-root ratio (S/R) and plant fresh weight (Table 8). If seen from the RRG value, the genotypes N0.80, MAL03x182, MAL03x42 and MAL03x192, were classified as tolerant, while MAL03x28, MAL03x42, MAL03x44 and MAL03x107 were classified as medium tolerant, while MAL03x56, MAL03x100, MAL03x115, MAL03/x124 are sensitive. This shows that a decrease in pH or an increase in soil acidity greatly affects plant growth and root length as the highest contributor to plant growth (shoot).

When Al^+ in soil solution touches the surface of corn roots, the roots immediately produce and release oxalic acid which chelates Al^+ so that it is deactivated by organic acids (Oxalic) and is not readily absorbed by corn roots [23]. According to Bano, et al. [24] that the active organic acids released by corn roots were malic acid and citric acid, and ZmALMT1 at root tips, critical for Al resistance. Expression of height ZmAT6 in maize increased root growth and reduced Al accumulation, and contributed to Al tolerance. [25]. Meanwhile Aluminum toxicity stress on purple corn seedlings can be detected using biochemical indicators (chlorophyll a, chlorophyll b and total chlorophyll, total carotenoids, and lipid [26]. Likewise reported by Caroline, et al. [27] that the greater expression difference in the tolerant landrace V 18 variety of the ZmMATE1 gene, suggests that citrate exudation may be the main mechanism of Al tolerance in this genotype.

Table 8. Responses of several maize lines seedling aged 14 days after planting planted on acid soil pH 4.3.

Lines, Genotype	PH (cm)	RL (cm)	FWR (g)	DWR (g)	S:R	PFW (g)	PDW (g)	RRG (%)
No.80	12.83 b	1.67 b	0.37 ab	0.11 b	0.55 ns	0.34 b	0.04 b	36.47 a
MAL03x28	13.42 b	2.77 ab	0.32 b	0.08 b	0.77	0.33 b	0.05 b	23.36 a
MAL03x182	19.57 ab	2.67 ab	0.35 ab	0.06 b	1.31	0.57 ab	0.08 ab	34.77 a
MAL03x42	15.90 b	2.73 ab	0.34 ab	0.13 b	0.63	0.37 b	0.07 b	33.10 a
MAL03x44	14.70 b	3.33 ab	0.63 ab	0.07 b	0.84	0.38 b	0.06 b	26.13 a
MAL03x56	29.71 a	17.43 ab	1.74 a	0.64 ab	1.25	2.06 a	0.83 a	-28.53 a
MAL03x100	19.83 ab	4.07 ab	0.76 ab	0.32 ab	0.29	0.74 ab	0.08 ab	2.23 a
MAL03x107	16.10 b	7.03 ab	0.38 ab	0.07 b	1.08	0.57 ab	0.08 ab	20.03 a
MAL03x115	22.10 ab	10.07 ab	0.61 ab	0.20 ab	0.52	0.81 ab	0.10 ab	9.10 a
MAL03x124	10.83 b	3.01 ab	0.56 ab	0.12 b	0.46	0.45 b	0.05 b	-121.63 b
MAL03x192	11.01 b	2.70 ab	0.49 ab	0.10 b	0.53	0.36 b	0.05 b	39.17 a
Bisi-18	28.01 a	20.02 a	1.74 a	0.77 a	0.65	1.60 ab	0.65 ab	9.33 a
CV.	21.58	26.11	18.53	20.30	15.71	17.75	17.47	27.5

Note. PH= Plant Height; RL= Root Length; FWR= Fresh Weight of Roots; DWR= Dry Weight of Root; S/R= Ratio of Plants to Roots; PFW= Plant Fresh Weight; PDW= Plant Dry Weight, RRG= Relative root growth.

While the correlation of several variables of plant growth under soil conditions pH 4,3 seedling age 14 dap showed that plant height and leaf length gave the highest value ($r^2 = 0.9179$ $P < 0.05$), followed sequentially by a correlation between plant fresh weight and plant height ($r^2 = 0.8343$, $P < 0.05$) and root length with plant height ($r^2 = 0.8100$, $P < 0.05$). This phenomenon showed that root length contributed greatly to plant growth compared to other variables in acid soils, this was proven to be the correlation between plant height and leaf length as the largest contributor to plant growth in acidic soil conditions pH 4.3, seedling age 14 days after planting (Table 9). This means that screening of maize lines on acid soil pH 4.3 at 7 days after planting is more accurate than in acid soil pH 4.3 at 14 days after planting, indicated by the number of maize genotypes tolerant screened at pH 4.3 which was lower in seed age of 7 dap compared to 14 dap.

Table 9. Correlation of parameters on the growth of several genotypes of acid tolerant maize seedling age 14 days after planting in soil pH 4.3.

Parameter	PH	RL	FWR	RDW	PFW	DWP	S/R	RRG	LL	LW
PH	1									
RL	0.8100	1								
FWR	0.7522	0.7783	1							
RDW	0.7861	0.8300	0.9394	1						
PFW	0.8543	0.8259	0.8853	0.8819	1					
DWP	0.7733	0.7869	0.9364	0.9092	0.9350	1				
S:R	0.3514	0.2512	0.1998	0.1185	0.4261	0.3778	1			
RRG	-0.2076	0.3668	0.3830	0.3430	-0.3953	0.3553	-0.0244	1		
LL	0.9179	0.8453	0.7618	0.8262	0.8354	0.7802	0.2921	-0.2069	1	
LW	0.2922	0.2593	0.3034	0.2533	0.3854	0.3711	0.4017	-0.0587	-0.3336	1

Note: PH= Plant Height; RL= Root Length; FWR= Fresh Weight of Roots; RDW = Root Dry Weight; S/R= Shoot and Root Ratio; PFW= Plant Fresh Weight; DWP= Dry Weight of Plants; RRG= Root Relative Growth; LL= leaf Length; LW= Leaf Width

3. Conclusion

Rapid screening of tolerant hybrid maize parental lines can be done by using a modified method of adding CaCO_3 and AlCl_3 to acid soils by measuring relative root growth (RRG) as the main parameter; screening of hybrid corn parent lines on acid soil pH 5.1 modified to more acidic (pH 4.3) and normal soil (pH 6.5) at seedling age 7 days after planting was more accurate than at seedling age 14 days after planting; genotypes N0.80, MAL03x192 and Bisi-18 were classified as acid tolerant ($\text{RRG} > 30\%$), while genotypes MAL03x28, AL03x182, MAL03x42, MAL03x44, MAL03x107 were classified as medium ($\text{RRG} 15\text{-}30\%$), while the genotypes MAL03x56, MAL03x115 and MAL03x124 were classified as sensitive ($\text{RRG} < 15\%$).

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