Journal of Plant Nutrition

ISSN: 0190-4167 (Print) 1532-4087 (Online) Journal homepage: www.tandfonline.com/journals/lpla20

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JOURNAL OF PLANT NUTRITION, 21(2), 387–403 (1998)

Hydrogen and Aluminum Inhibition of Soybean Root Extension from Limed Soil into Acid Subsurface Solutions

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ABSTRACT

Soybean [*Glycine max* (L.) Merr. cv. 'Ransom'] root elongation under varying concentrations of solution hydrogen (H) and aluminum (Al) was investigated in a vertical split-root system. Roots extending from a limed and fertilized soil compartment grew for 12 days into a subsurface compartment with solutions adjusted to either different pH values from 3.7 to 5.5 or a factorial combination of pH (4.0, 4.6, and 5.2) and Al (0, 7.5, 15, and 30 μM) levels. Ionic forms of Al were estimated with GEOCHEM and solution Al was determined with ferron. Boron (B) (18.5 μM) and zinc (Zn) (0.5 μM) were supplied to all solution treatments, in addition to 2000 μM Ca, after preliminary studies at pH 5.2 without Al indicated that their omission inhibited length of tap roots and their laterals in the subsurface compartment. Both H^+ and Al inhibited the length of lateral roots more than tap roots. Lateral roots failed to develop on tap roots at pH<4.3 or in treatments with 30 μM Al. Relative tap root length (RRL) among treatments receiving Al correlated with Al as measured by reaction with ferron for 30s. Ferron-reactive Al was correlated to GEOCHEM-predicted Al^{3+} activity (r=0.99). A 50% reduction in RRL

occurred with either 2.1 μM Al^{3+} activity or 4.9 μM ferron-reactive Al. The absence of shoot and soil-root biomass differences among solution treatments in the split-root system indicated that differences in root growth in the subsurface compartment were not directly confounded with differences in top growth.

INTRODUCTION

Plants grown in limed surface soils may be restricted from using water and nutrients in the soil profile if root proliferation is limited by acid subsoil conditions (Richey et al., 1980; Goldman et al., 1989). Aluminum and H^+ toxicity are important components of the soil acidity constraint to root growth. Aluminum disrupts regulatory signals in root cap cells (Bennet and Breen, 1991) and interferes with enzyme activities, cell division in root apices, DNA replication and P availability at membranes. Hydrogen-induced root injury involves changes in root membrane permeability, interference in nutrient transport, and loss of organic substrates and absorbed cations (Foy, 1984).

Studies with soybean have shown that toxic levels of Al and H^+ inhibit root elongation (Lund, 1970) and decrease the adsorption and translocation of nutrients to plant tops (Foy, 1984; Noble and Sumner, 1988). The extent to which H^+ inhibits soybean root elongation remains unclear. Lund (1970) evaluated H^+ inhibition of tap root growth for soybean cv. 'Lee' in a split-root system, and reported a reduction in root elongation for all solution treatments with pH<5.6 (i.e., pH values of 4.75, 4.5, and 4.0). Noble et al. (1988a) found no difference in tap root length among solutions pH values ranging from 4.2 to 4.8. Exposure of germinating seeds for 72 hours to solutions with pH ranging from 4.0 to 5.5 had no affect on soybean radicle length, but transplanting sprouted seeds to solutions with adjusted pH in the range of 3.5 to 4.4 significantly reduced root length at 96 hours (Suthipradit and Alva, 1986).

Recent investigations with soybean have focused on the relative toxicities of Al species to root growth using hydroponic systems, which allow control over variables influencing solution Al behavior (pH, ionic strength, other cations and counter ions) (Alva et al., 1986; Noble et al., 1988a; Parker et al., 1989). Emphasis on Al phytotoxicity and the absence of adequate control treatments for solution pH variables often limit the distinction between root growth inhibition by H^+ or by pH-induced changes in the distribution of solution ionic species of Al (Kinraide, 1991).

The objective of this study was to quantify soybean root elongation response to varying concentrations of H^+ and Al in the subsurface compartment of a vertically split-root system. This system simulates an acid subsoil underlying a limed surface soil and allows assessment of root elongation responses to H^+ and Al while minimizing confounding effects on shoot growth.

MATERIALS AND METHODS

Experiments were carried out in greenhouse facilities at Raleigh, North Carolina, using a vertical split-root system as shown in Figure 1. White plastic cylinders, with a 10-cm inside diameter and 52-cm total length, were divided into two vertical compartments by a root-permeable membrane. The membrane was formed by dipping a single layer of cheesecloth attached to the surface compartment into a mixture of one part paraffin and two parts petrolatum heated to 80°C for 10 seconds. The 12-cm long surface compartment contained 1.3 kg of loamy sand taken from the Ap horizon of a Wagram soil (loamy, siliceous, thermic, Arenic Kandiudults) near Whiteville, North Carolina. Selected chemical properties of the soil material before liming and fertilization were pH 4.3, 0.81 $cmol_c\ kg^{-1}$ of calcium (Ca), 0.08 $cmol_c\ kg^{-1}$ of magnesium (Mg), 0.05 $cmol_c\ kg^{-1}$ of potassium (K), 0.34 $cmol_c\ kg^{-1}$ of Al, 26.5 % Al saturation, and 20.5, 0.9, 0.7, and 4.1 mg kg^{-1} of Mehlich-3 extractable phosphorus (P), zinc (Zn), copper (Cu), and manganese (Mn), respectively. The soil was limed to pH 5.5 with $CaCO_3$ and received 30 mg K kg^{-1} before transfer to the surface compartment.

Treatments were imposed to the subsurface solution compartment, which contained 3.0 L of continuously aerated solution. The bottom of the solution compartment was sealed with a white plastic cap. Effects of pH, Al concentration and the omission of B, Ca, or Zn on growth of soybean roots were evaluated in the solution compartment (Table 1). In Experiment 1 treatments were a solution containing Ca, B, and Zn and three treatments in which each of the three nutrients was excluded. Subsequent experiments included Ca, B, and Zn in all solution treatments. In Experiment 2, treatments consisted of a range of solution pH values in the absence of Al, and in Experiment 3 treatments consisted of a range of Al concentrations at each of three pH values. A randomized complete block design was used in all experiments, with four replications in Experiment 1 and three replications in Experiments 2 and 3. Solution pH values were adjusted daily by titration with either 0.05 N HCl or NaOH. In all experiments B was supplied as H_3BO_3. Calcium was supplied as $CaSO_4$ in Experiment 2 and as $CaCl_2$ in Experiments 1 and 3. Zinc was supplied as $ZnSO_4$ in Experiment 2 and as ZnO in Experiments 1 and 3. Aluminum was supplied in Experiment 3 as metallic Al diluted in 5% HCl. All solutions were replaced after eight days of root growth.

Soybean seeds were imbibed in 200 µM $CaSO_4$ and pre-germinated in the dark for 2 days at 25°C in petri dishes containing filter paper moistened with the same Ca solution. Six seedlings with a radicle length of 12±2mm were planted in the soil compartment of each container. After two days each container was thinned to five plants in Experiments 1 and 3, and four plants in Experiment 2. All greenhouse experiments were conducted between May and October. Natural light was supplemented with 150 microeinsteins $m^{-2}\ s^{-1}$ of photosynthetically active radiation from metal halide lamps for 16 hours day^{-1}. Soil moisture was maintained

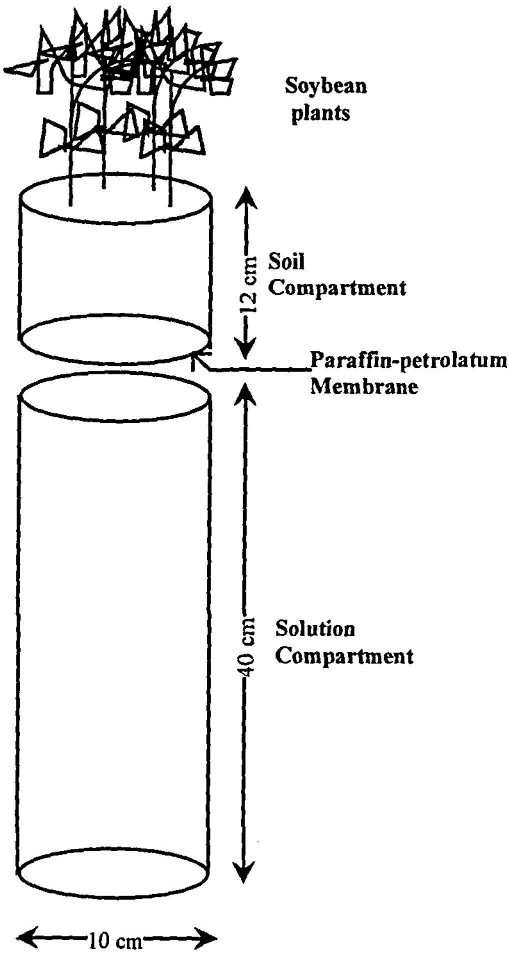

FIGURE 1. Diagram of the split-root system used to evaluate soybean root growth in a subsurface solution compartment under varying acidity conditions.

TABLE 1. Description of subsurface solution compartment treatments for each experiment.

	Treatment Variables				
Experiment	pH	Al	Ca	B	Zn
		---------μM---------			
1	5.2	0	0, 2000	0, 18.5	0, 0.5
2	3.7, 4.0, 4.3, 4.6, 4.9, 5.2, 5.5	0	2000	18.5	0.5
3	4.0	0, 7.5, 15, 30	2000	18.5	0.5
	4.6	0, 7.5, 15, 30	2000	18.5	0.5
	5.2	0, 7.5, 15, 30	2000	18.5	0.5

at 80% of container capacity (Cassel and Nielsen, 1982) by weighing the containers daily and correcting for projected plant weight. This moisture level was chosen to avoid water movement from the soil to the solution compartment during the experiment.

Tap roots began to cross the membrane on the third day after planting and their length in the solution compartment was measured daily for 12 days thereafter. Plants were harvested 15 days after planting when the first trifoliate was in the expansion phase and cotyledons had yellowed but not yet abscised. Dry weight of plant shoots was determined after drying for 24 hours at 70°C in a forced-draft oven. Roots in the soil compartment were washed free of soil, length was measured by the line-intercept method (Tennant, 1975), and dry weight was determined after drying for 24 hours at 70°C. Measurements at harvest in the solution compartment included tap root length, number and length of laterals on the tap roots, and number and length of any other roots extending through the membrane from the soil compartment. Root classes are identified according to the nomenclature of Zobel et al. (1992) whenever possible. For roots extending through the membrane from the soil compartment only tap roots were clearly identified; thus any additional root types (basal, lateral, or adventitious) will be identified as "other" roots. Relative root length for a root class within each experiment was calculated as the ratio between a given treatment and the treatment with the greatest length.

Aluminum reacting in 30 seconds with buffered ferron was determined in aliquots of solutions taken at harvest from all treatments containing Al in Experiment 3 using a procedure adapted from Bersillion et al. (1980). The reagent

TABLE 2. Mean values and standard deviations across treatments in each experiment for soybean shoot dry weight, and dry weight and total length of roots in the soil compartment at harvest.

	Shoot		Root			
Experiment	Dry Weight	SD	Dry Weight	SD	Length	SD
	---------------g/plant---------------				-----m/plant-----	
1	0.33	0.04	0.24	0.07	6.88	1.58
2	0.23	0.07	0.21	0.08	5.77	1.68
3	0.31	0.03	0.24	0.07	9.12	1.35

mixture of ferron, 1-10 phenanthroline and acidified hydroxylamine hydrochloride was aged for 5 to 7 days, as recommended by Jardine and Zelazny (1986), and stored in the dark at room temperature. Volume ratio between sample aliquots and reagent mixture in the final solution used for spectrophotometric readings was 1:0.8.

Solution pH and elemental concentrations for experimental treatments were used to compute the distribution and activities of Al species by the GEOCHEM-PC program (Parker et al., 1995). Statistical analysis of experiment data was performed with analysis of variance and regression procedures (SAS, 1985).

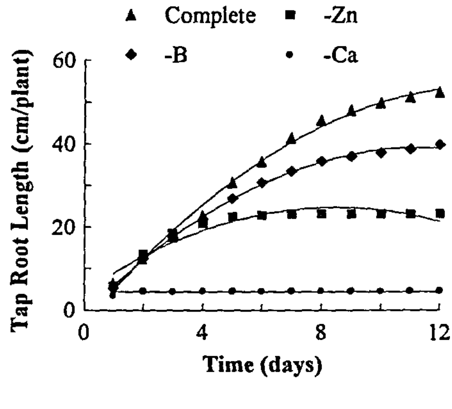

FIGURE 2. Observed (symbols) and predicted (lines) primary soybean root length as a function of time of exposure to solutions with and without B, Ca, and Zn. Polynomial equations for predicted root elongation are shown in Table 3.

TABLE 3. Regression equations for primary root elongation (y in cm plant^{-1}) as a function of time (x in days) in the subsurface solution compartment treatments for Experiment 1.

Treatment	Equation§	R^2
Complete	$y=-3.32+8.37x-0.31x^2$	0.99
B omission	$y=-1.07+7.10x-0.31x^2$	0.99
Zn omission	$y=4.31+4.79x-0.28x^2$	0.99
Ca omission	$y=4.52$	--

§Linear and quadratic coefficients are significant at $p<0.01$.

RESULTS AND DISCUSSION

Treatment means for soybean shoot dry weight, and total length and dry weight of roots in the limed and fertilized soil compartment at harvest were not significantly different ($p<0.05$) in any of the experiments. Lower mean values for these parameters in Experiment 2, relative to Experiments 1 and 3, were attributed to fewer plants per container and less natural photosynthetically active radiation during the growth period (Table 2). Similarities in shoot and soil-root biomass within experiments suggested an equal supply of assimilates for roots growing into different growth-limiting treatments in the subsurface solution compartments. Differences in root growth among subsurface compartment treatments, therefore, were not confounded directly with differences in top growth.

Root Growth Without Calcium, Boron, or Zinc

Omission of either Ca, B, or Zn from solution reduced tap root elongation in the subsurface compartment (Figure 2). Polynomial equations for predicted root elongation are shown in Table 3. The magnitude of root length reductions were greatest in the absence of Ca, followed by Zn, and then B, and increased with the time roots were exposed to these solutions. Root growth stopped after the first day in -Ca solutions, continued to day 5 in the -Zn treatment, and essentially stopped after day 10 in the -B solution.

The relative contributions of each root class to total root length/plant in the subsurface compartment at harvest were significantly ($p<0.05$) influenced by omissions of Ca, B, or Zn from solutions (Figure 3). No lateral roots were detected on tap roots in solutions without Ca (Table 4). The average length and total number of laterals decreased with the omission of B and Zn from solution, but the number of laterals per unit length of tap root (lateral root density) remained

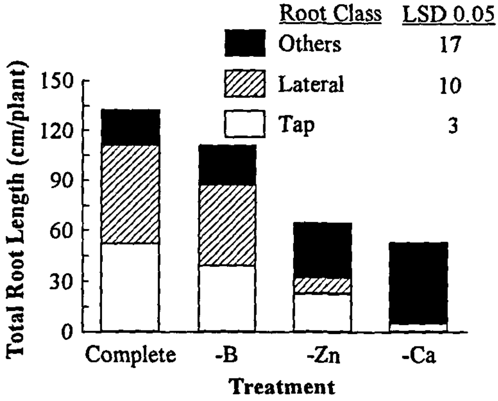

FIGURE 3. Total length as a function of soybean root classes in the subsurface compartment after 12 days of exposure to solutions with and without B, Ca, and Zn.

relatively constant. Reductions in total length of lateral roots/plant in solutions without B and Zn, therefore, resulted primarily from reduced elongation of both tap roots and their lateral roots, rather than changes in lateral root density.

As the length of tap roots and their laterals in the solution compartment decreased for treatments with nutrient omissions the length of other roots crossing the membrane from the soil compartment increased (Figure 3). The number of other roots increased with the severity of the constraint in growth of tap roots and their laterals, but average length of other roots decreased from 2.0 cm for the complete treatment to 1.4, 1.3, and 1.2 cm for the -B, -Zn and -Ca treatments, respectively (Table 4). Reduced elongation of tap roots and their laterals in the absence Zn and Ca apparently stimulated root branching or elongation of existing roots in the surface soil compartment, which led to more roots crossing the membrane into the solution compartment.

Observed reductions in elongation of tap roots and their laterals with the omission of B or Zn from the solution compartment suggest little downward translocation of these nutrients following uptake from the soil compartment. The essentiality of an external Ca supply for normal root development has been documented (Clarkson, 1984; Ferguson and Clarkson, 1976; Rios and Pearson, 1964). Studies with species other than soybean have also indicated that basipetal transport of B and Zn, in the absence of an external supply to roots, is inadequate for normal root functions (Lukaszewski and Blevins, 1996; Webb and Loneragan, 1990). Boron and Zn were added to all treatment solutions in subsequent experiments to minimize growth limiting factors other than H and Al.

TABLE 4. Measured and calculated characteristics of roots at harvest time in the subsurface solution compartment influencing distribution of length among root classes in treatments for Experiment 1.

Treatment	Number of Roots		Average Root Length[§]		Lateral Root[†]
	Lateral	Other	Lateral	Other	Density
	number/plant		----------cm----------		number/cm
Complete	128	11	0.46	2.00	4
B omission	121	17	0.39	1.44	4
Zn omission	85	25	0.11	1.34	5
Ca omission	0	42	--	1.16	0
LSD 0.05	9	12			

[§]Derived from mean treatment values for root length and number.
[†]Number of lateral roots/length of tap root.

Root Growth with Variable pH

Solution pH values and maximum variation within any 24-h period for each treatment in Experiment 2 were 3.7±0.03, 4.0±0.05, 4.3±0.07, 4.6±0.05, 4.9±0.07, 5.2±0.06, and 5.5±0.09. Tap roots in solutions with pH<4.6 had visual symptoms of H^+ injury, namely stunted growth, brownish color and little lateral root development. Islam et al. (1980) reported similar symptoms for wheat, corn and tomato roots when grown in complete nutrient solutions at pH<4.8. In our experimental system there were significant ($p<0.05$) differences in lengths of tap roots and lateral roots in the subsurface compartment among pH treatments at harvest (Figure 4). Length of tap roots increased at a linear rate of 26 cm plant^{-1} (r^2=0.94) with each unit increase in solution pH. No lateral roots formed on tap roots at pH<4.3. Between pH 4.3 and 5.5 both the length and number (Table 5) of lateral roots increased exponentially. Lateral root density on tap roots was greatest at pH 4.3, but remained relatively constant between pH 4.6 and 5.5. The average length of a lateral root doubled between pH 4.3 and pH 5.2.

For each unit increase in solution pH the number of other roots crossing the membrane from the soil compartment decreased by 3.4 plant^{-1} (r^2=0.88) and their average length increased by 1.0 cm (r^2=0.87) (Table 5). These opposing trends in characteristics of other roots led to a relatively constant contribution of their length (mean=15.8 cm plant^{-1}) to the total root length among pH treatments in the solution compartment.

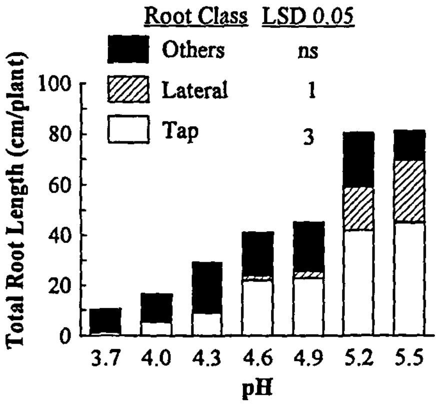

FIGURE 4. Total length as a function of soybean root classes in the subsurface compartment after 12 days of exposure to solutions adjusted to varying pH values. A nonsignificant effect at $p<0.05$ for a root class is denoted by 'ns'.

TABLE 5. Measured and calculated characteristics of roots at harvest time in the subsurface solution compartment influencing distribution of length among root classes in treatments for Experiment 2.

Solution	Number of Roots		Average Root Length[§]		Lateral Root[†]
pH	Lateral	Other	Lateral	Other	Density
	number/plant		----------cm----------		number/cm
3.7	0	12	--	0.78	0
4.0	0	10	--	1.11	0
4.3	3	11	0.10	1.88	6.5
4.6	11	9	0.16	1.96	2.1
4.9	15	9	0.18	2.25	1.9
5.2	80	8	0.21	2.75	2.2
5.5	119	5	0.21	2.42	3.0
LSD 0.05	3	1			

[§]Derived from mean treatment values for root length and number.
[†]Number of lateral roots/length of tap root.

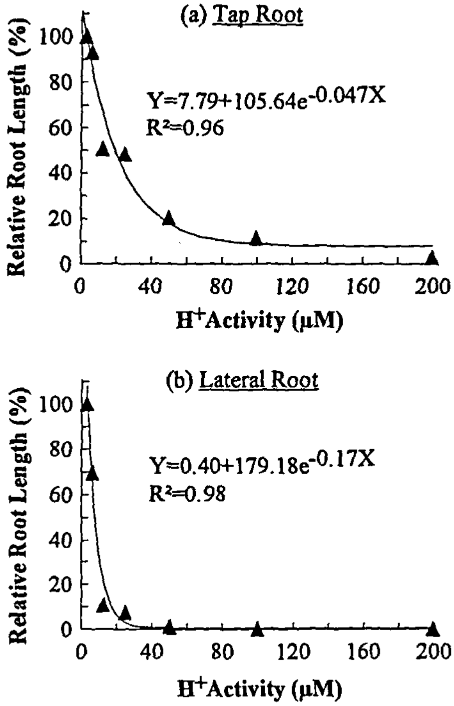

FIGURE 5. Observed (symbols) and predicted (lines) relative length of (a) primary and (b) secondary soybean roots in the subsurface compartment as a function of solution H^+ *activity.*

Relations between H^+ activity (H^+) and relative length of tap and lateral roots in the solution compartment suggested a greater inhibition of lateral roots by H^+ (Figure 5). A 50% reduction in tap root length was obtained with 19.5 µM (H^+), whereas a similar reduction in lateral root length occurred with 7.6 µM (H^+). Greater inhibition of lateral root length by H^+ cannot be solely attributed to a multiplier effect from reduced elongation of tap roots, because lateral root density and the average length of laterals also changed with solution pH (Table 5). These data are in agreement with Lund's (1970) observation of a greater adverse effect of H^+ on lateral roots than on tap roots for soybean cv. 'Lee'.

Effects of Solution pH and Al

Root Growth

There were significant main effects of solution pH and Al treatments on the length of each root class, their sum, and the number of laterals and other roots in

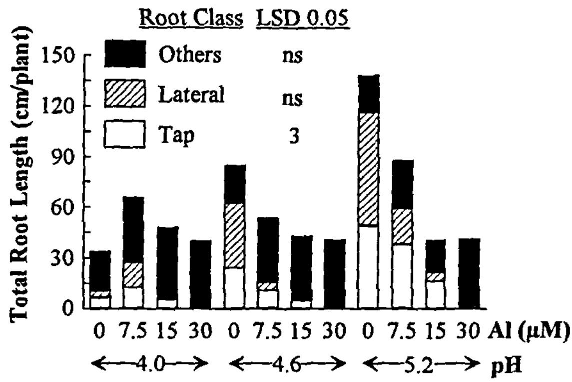

FIGURE 6. Total length as a function of soybean root classes in the subsurface compartment after 12 days of exposure to solution Al and pH treatments. Least significant difference values are for interaction effects between Al and pH treatments; ns stands for non significant at $p<0.05$.

the subsurface compartment at harvest. Interaction effects between pH and Al treatments were only significant for tap root length, lateral root number and total root length. In solutions adjusted to pH 4.0 tap roots in all treatments receiving Al had visual symptoms of both H^+ and Al injury, namely short, swollen and twisted with short lateral roots. In the 30 μM Al treatments elongation of tap roots stopped after one day of exposure to solutions at pH 4.0 and 4.6, but increased from 1.3 to 1.6 cm $plant^{-1}$ during 12 days of growth in the solution at pH 5.2.

In the absence of Al, length of tap roots and their laterals at harvest increased with solution pH (Figure 6) as in Experiment 2. At pH 4.0 length of tap and lateral roots between 0 and 7.5 μM Al increased by 17 cm $plant^{-1}$, whereas length of these root classes at higher pH decreased with increasing Al concentrations. This apparent alleviation of H^+ toxicity by low Al concentrations was observed in previous studies (Llugany et al., 1995; Kinraide and Parker, 1987) and may be associated with H-Al competition for binding sites on cell surfaces (Kinraide and Parker, 1987). Severity of root injury by 30 μM Al treatments was such that no lateral roots were detected on tap roots at all solution pH levels. Inhibition of elongation by Al at pH 4.6 and 5.2 was proportionately greater for laterals than for tap roots. Horst and Klotz (1990) observed that 4-day elongation of both of these soybean root classes were equally inhibited by nutrient solutions with 74 μM Al, but lateral roots were more sensitive at lower (unspecified) Al supply.

Length of tap and lateral roots averaged across Al treatments increased by 17 ($r^2=0.90$) and 15 cm $plant^{-1}$ ($r^2=0.96$), respectively, with each unit increase in

TABLE 6. Measured and calculated root characteristics at harvest in the subsurface solution compartment influencing distribution of length among root classes in treatments for Experiment 3.

Treatment		Number of Roots		Average Root Length[§]		Lateral Root[†]
pH	Al	Lateral	Other	Lateral	Other	Density
	μM	number/plant		----------cm----------		number/cm
4.0	0	26	45	0.14	0.53	5.0
	7.5	19	39	0.78	0.99	2.6
	15	4	50	0.15	0.85	6.0
	30	0	58	--	0.69	--
	Mean	12	48	0.36	0.77	4.5
4.6	0	59	22	0.64	1.04	3.1
	7.5	40	38	0.12	1.02	3.2
	15	12	43	0.14	0.85	4.8
	30	0	56	--	0.71	--
	Mean	28	40	0.30	0.91	3.7
5.2	0	212	12	0.31	1.83	4.9
	7.5	92	17	0.23	1.73	3.2
	15	40	30	0.13	0.63	3.2
	30	0	50	--	0.79	--
	Mean	86	27	0.22	1.24	3.8
	---------- Al Treatment Means ----------					
	0	99	26	0.36	1.13	4.3
	7.5	50	31	0.38	1.25	3.0
	15	19	41	0.14	0.78	4.7
	30	0	55	--	0.73	--
LSD 0.05						
pH		4	2			
Al		5	3			
pHxAl		8	ns			

[§]Derived from mean treatment values for root length and number.
[†]Number of lateral roots/length of tap root.

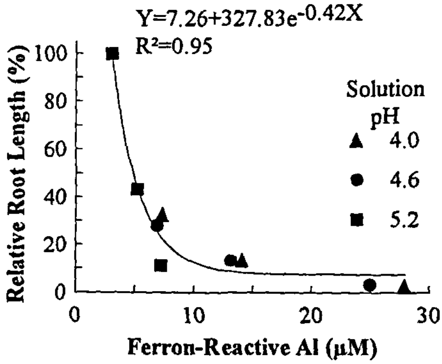

FIGURE 7. Observed (symbols) and predicted (line) relative length of primary soybean roots in the subsurface compartment treatments receiving Al as a function of Al reacting with ferron by 30 seconds.

solution pH. Mean effects of pH across Al treatments increased lateral root number 7-fold between pH 4.0 and 5.2, whereas average length of a lateral root decreased from 0.36 to 0.22 cm (Table 6). For each unit increase in solution pH, total length of other roots crossing the membrane decreased by 9 cm plant^{-1} (r^2=0.90) and their number decreased by 21 plant^{-1} (r^2=0.98). The average length of other roots also increased with solution pH.

Total length and number of lateral roots averaged across pH treatments decreased exponentially with increasing Al levels, and their average length decreased from 0.36cm at 0 μM Al to 0.14 cm at 15 μM Al (Table 6). When averaged across pH treatments the total length of other roots increased by 0.5 cm plant^{-1} (r^2=0.94) and their number increased by 1 plant^{-1} (r^2=0.99) with each unit increase in solution Al. The observed trends for increased number of other roots crossing the membrane and their proportionately greater contribution to total root length in the solution compartment with increasing severity of the H^+ and Al constraints was similar to the pattern observed in Experiment 1 upon the omission of Ca, B, or Zn supply.

Relations Between Solution Aluminum and Tap Root Length

Relative length of tap roots among treatments receiving Al was related to Al in solutions which reacted with ferron by 30 seconds (Figure 7). There was a significant linear correlation (r=0.99) between ferron-reactive Al and Al^{3+} activity (Al^{3+}) as predicted by GEOCHEM. Consequently, relative length of tap roots (y) among solutions with Al was closely related to (Al^{3+}) ($y=6.67+223.09e^{-0.79X}$; R^2=0.97). Several investigators have reported that ionic activities for other forms of monomeric Al in nutrient solutions were more closely related to elongation of soybean tap roots (Alva et al., 1986; Noble et al., 1988a). In our study correlations and visual inspection of plots for relations of either ferron-reactive Al or tap root length with activities of GEOCHEM-predicted $AlOH^{2+}$, $Al(OH)_2^+$, or the sum of monomeric Al were either non-significant or inferior to the relations with (Al^{3+}). Likewise, the relation between length of tap roots and the valence-weighted sum of (Al^{3+}), ($AlOH^{2+}$) and [$Al(OH)_2^+$], as proposed by Noble et al. (1988b), was also inferior (R^2=0.82) to the relationship with (Al^{3+}).

In our experiment a 50% reduction in relative length of soybean tap roots occurred with 2.1 μM (Al^{3+}). This is similar to the value of 2.4 μM (Al^{3+}) which Alva et al. (1986) reported for a 50% reductions in length of tap roots for 6-day-old soybean grown in complete nutrient solutions.

Although collinearity among Al species in our experimental design limits the assessment of their relative phytotoxicities, GEOCHEM-predicted activities of $Al(OH)_3^0$ and $Al(OH)_4^-$ were <0.01 μM in solutions with pH 4.0 and 4.6, and never exceeded 0.3 μM in solutions with pH 5.2. According to the diagnostic criteria proposed by Kinraide (1991), the potential formation of polynuclear Al, such as triskaidekaaluminium (Al_{13}) (Parker et al., 1989), would only be feasible in solution treatments with pH 5.2. Furthermore, the presence of the highly phytotoxic Al_{13} in these solutions would not be consistent with the lower inhibition of root growth by Al at pH 5.2 relative to pH 4.0 and 4.6 (Figure 6).

CONCLUSIONS

Results from these experiments indicate that both Al and H^+ inhibited length of lateral roots to a greater extent than tap roots. Elongation of tap roots at pH<5.2 was inhibited by both H and Al. Within this pH range, root length inhibition of Al-containing solutions cannot be solely attributed to Al. This split-root system was also used to investigate alleviation Al and H^+ rhizotoxicity across a range of solution Ca concentrations, and is reported elsewhere (Sanzonowicz et al., 1997). The observed reductions in root elongation in the subsurface compartment, in the absence of B and Zn supply, merit further investigations in both subsoils and split-root hydroponics systems to determine optimal levels of these nutrients for root growth.

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