

Tolerance of Arabica Coffee Cultivars for Aluminum in Nutritive Solution

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ABSTRACT

This work aimed to evaluate the *Coffea arabica* cultivars for aluminum toxicity tolerance, in modified Hoagland solution. A completely randomized design with five repetitions in a factorial 4 x 4 (cultivar x combinations of aluminum) was used. After 44 days of the sowing, were transferred ten seedlings each cultivar germinated in the absence of Al³⁺ to solution without Al³⁺, and ten for solution with Al³⁺; ten seedlings each cultivar germinated in presence of Al³⁺ to solution without Al³⁺, and ten for solution with Al³⁺. In the treatment with aluminum, the element was added to the nutritive solution in the concentration of 0.83 mmol L⁻¹ as Al₂(SO₄)₃.16H₂O. The cultivars Catuaí Amarelo IAC 62 and Iapar 59 were tolerant to the aluminum; cultivar Oeiras presented intermediate tolerance, while cultivar Obatã IAC 1669-20 was sensitive. The tolerance of the coffee cultivars to the aluminum during the initial development of the seedlings did not depend on the presence of aluminum in the germination phase.

Key words: *Coffea arabica*, aluminum toxicity, vigor, breeding

INTRODUCTION

Brazil is the largest coffee-producing country in the world, responsible for the development of many localities in the Espírito Santo state of Brazil (ABIC, 2006). According to CONAB (2008), Brazilian coffee production during the 2007/2008 harvest year was 33.7 million green coffee 60 kg bags, with mean productivity of 16.27 bags per hectare. However, this was low compared to its genetic productivity potential.

When fructification is low, plagiotropic branches and new leaves and branches replace fruit as a

carbohydrate and nutrient sink (Malavolta et al., 2002), despite the fact that the supply, absorption and balanced use of the essential mineral nutrients are related to the pH and presence of exchangeable aluminum (Marschner, 2003). If the pH is not at the adequate range, nutrient deficiency and toxicity may occur and production would suffer, leading to decreased nutritional efficiency (Fageria, 1998). Considering that superficial lime application under the no-till cultivation system does not totally and adequately correct soil acidity deeper than 10 cm (Rheinheimer et al., 2000), a

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viable alternative would be the use of aluminum-tolerant species or cultivars (Foy et al., 1978).

Al^{3+} solubility increases under pH below 5.5 and its toxicity is particularly severe under pH below 5.0 (Fageria, 1998). Justino et al. (2006) found that Al^{3+} has a wide range of action on plant metabolism, this probably being the reason why its mechanism of action is not well known yet. However, it is known that the negative effect of aluminum does not affect calcium absorption directly but rather through root growth inhibition, decreasing Ca^{2+} absorption, regardless of the direct effect of aluminum on the absorption process (Menosso et al., 2000).

Braccini et al. (2000a), based on the evaluation of primary root length of 26 coffee lines submitted to 45 mg L⁻¹ aluminum concentration, using the paper-solution method, found different levels of tolerance among the lines in relation to the presence of toxic aluminum.

Plant tolerance to aluminum is often associated to the plant's capacity to alter the pH in the rhizosphere (Degenhardt et al., 1998). Mendonça et al. (2005) used nutritive solutions in the absence and presence of aluminum at different rates of $\text{NO}_3^-/\text{NH}_4^+$, and showed that the differential tolerance to aluminum in two rice cultivars could be associated to their capacity to modify the pH of the nutritive solution.

The plant have wide ability to adapt for different agricultural ecosystems, deriving from several factors, such as economic, marginal area utilization and production stability (Menosso et al., 2000). Plant species germinating under certain conditions, such as in the presence of exchangeable aluminum, are likely able to acquire a greater capacity of tolerating the adverse effects of this element. The use of tolerant cultivars to soil Al^{3+} toxicity allows the commercial use of many marginal areas for cultivation. The differential tolerance to aluminum is a characteristic easily detected in greenhouse or laboratory tests using nutritive solution (Dornelles et al., 1997).

This work aimed to evaluate the *Coffea arabica* cultivars aluminum toxicity tolerance in nutritive solution.

MATERIAL AND METHODS

This work was carried out from December 2005 to March 2006. The seeds were obtained from Incaper experimental station in Venda Nova do

Imigrante. The experiment was conducted in a completely randomized design with five repetitions, each one constituted by two seedlings. Arabica coffee cultivars were distributed in the 4 x 4 factorial scheme in four aluminum treatments: pre-treatment without aluminum during the germination and transplanted to a nutritive solution without aluminum (-Al / -Al); pre-treatment without aluminum during the germination and transplanted to the nutritive solution with aluminum (-Al / +Al); pre-treatment with aluminum during the germination and transplanted to the nutritive solution without aluminum (+Al / -Al); pre-treatment with aluminum during the germination and transplanted to nutritive solution with aluminum (+Al / +Al).

The seedlings of the cultivars Catuaí Amarelo IAC 62, Iapar 59, Obatã IAC-1669/20 and Oeiras (MG 6851), derived from the seeds without parchment were removed manually and germinated in three Germitest[®] type tissues. The seeds were moistened (at the proportion of 2.5 times the tissue weight) with a nutritive solution which contained (mmol L⁻¹) MgSO_4 0.1, KNO_3 0.1, NH_4NO_3 0.15 and $\text{KHC}_8\text{H}_4\text{O}_8$ 8.0 (potassium biphthalate to maintain the pH around 4.0) with or without 0.83 mmol L⁻¹ of Al^{3+} in the form of $\text{Al}_2(\text{SO}_4)_3 \cdot 16\text{H}_2\text{O}$. In order to prevent the fungus development, the seedlings were treated with the fungicide Captan (Orthocide[®]) at the concentration of 0.1%. The seedlings were placed at the vertical position inside the plastic vases (1 liter volume), containing 300 ml of nutritive solution at different levels of Al^{3+} so as to keep the tissue always moistened. The recipients were maintained in the dark inside the germinator at 30±1°C.

After 44 days from the start of sowing, the uniform seedlings were selected, at the "match stick" stage to transplant to a hydroponic system, which at the primary root presented approximately six centimeter of length. Later, 10 seedlings of each cultivar, germinated in the absence of Al^{3+} , were transferred to the nutritive solution in the absence and presence of Al^{3+} , as well as 10 seedlings of each cultivar, germinated in the presence of Al^{3+} , were transferred to the nutritive solution in the absence and presence of Al^{3+} . The nutritive solution used was Hoagland and Arnon (1950), modified, containing macronutrients (mmol L⁻¹): N = 7.5; P = 0.5; K = 3.0; Ca = 2.5; Mg = 1.0; S = 1.0; and micronutrients (μmol L⁻¹): Mn = 4.6; Cu = 0.2; Zn = 0.4; Mo = 0.06; B = 23.1; Fe, in the form of Fe-EDTA = 0.05; Cl = 4.6.

In the aluminum treatment, the element was added to the nutritive solution at the concentration of 0.83 mmol L^{-1} , in the form of $\text{Al}_2(\text{SO}_4)_3 \cdot 16\text{H}_2\text{O}$. The nutritive solutions were renewed after 21 days from the day the seedlings were transferred to the hydroponic system and the pH of the nutritive solutions was adjusted to 4.0 ± 0.2 . The P concentration (0.5 mmol L^{-1}) was low to minimize aluminum precipitation.

The hydroponic system was installed in a growth chamber under ambient temperature of $25 \pm 2^\circ\text{C}$, relative humidity of 60% and 8 h photoperiod, using four 40 Watt fluorescent lights. Plastic vases of 1 liter were used, wrapped in aluminum paper. As supports for the seedlings, styrofoam plates with two holes were used, with each plantlet being supported by a styrofoam cylinder, longitudinally sectioned, with the same diameter of the plate's hole. The solution was aired through air bubbling in a continuous way supplied by a motor-compressor.

The completion of the treatment coincided with seedling collection after 42 days of transplant by sectioning them at the stem height. The height of the aerial part, primary root length, fresh and dry mass of the aerial part and root system of each repetition were evaluated. Dry mass was obtained after the different seedling parts were dried in an forced air circulation oven at $80 \pm 2^\circ\text{C}$, until reaching constant weight. The RCR rate (root length percent reduction ratio) was calculated

according to the equation below, suggested by Baligar et al. (1989):

$$\% \text{RCR} = [1 - (\text{Growth with Al} / \text{Growth without Al})] \times 100.$$

The experimental data were submitted to variance analysis and when significant, the means were compared by the Tukey test at 1% and 5% probability, using the statistical software SAEG (Statistical Analyses Systems of the Universidade Federal de Viçosa - UFV), 9.0 version (Euclides, 2004). The "Lilliefors" and "Cochran and Bartlett" tests were applied at 1% significance level to verify data normality and variance homogeneity, respectively.

RESULTS AND DISCUSSION

The highest pH values were observed in the nutritive solutions 21 days after solution exchange (42 days after of transplant). However, there was no daily adjustment of pH. Without aluminum, regardless of the cultivar, the pH increased, on an average, in 3.0 units (Table 1). However, with aluminum, there was no alteration. These results suggested that pH variation was dependent on the absence or presence of aluminum in the medium, and that the different cultivars did not interfere in this variation, confirming the results of Braccini et al. (2000b).

Table 1 - Mean of the final pH values of the nutritive solutions in the absence (-Al) and presence of aluminum (+Al) of each *Coffea arabica* cultivar.

Cultivars	Final pH of the nutritive solution	
	- Al	+ Al
Catuaí Amarelo IAC-62	7.2	3.9
Iapar 59	7.0	3.8
Obatã IAC-1669/20	7.0	3.9
Oeiras MG-6851	6.8	3.9
Mean	7.0	3.9

The evaluation of the fresh mass of the seedling is related to the capacity of the cultivar to accumulate water under a particular condition. The treatment in the absence of aluminum during the germination and growth (-Al / -Al) represented the control, with the lowest FMAP values being observed in the cultivars Oeiras MG-6851 and Catuaí Amarelo IAC 62, indicating adaptation of cultivars Iapar-59 and Obatã IAC-1669/20 to neutral pH and aluminum zero soil (Table 2). In

the treatment -Al / +Al (pre-treatment without aluminum during the germination and transplanted to nutritive solution with aluminum), none of the cultivars presented significant difference in relation to FMAP. Cultivar Catuaí Amarelo IAC 62 presented a lower level of FMAP in the treatment with aluminum during the germination (+Al / -Al), not differing from the cultivar Oeiras MG 6851. However, a significant difference was observed between these two cultivars in the

treatment +Al / +Al (pre-treatment with aluminum during the germination and transplanted to nutritive solution with aluminum) indicating aluminum sensibility of 'Catuaí Amarelo IAC-62' and tolerance of Iapar-59', not differing from 'Oeiras MG-6851' and 'Obatã IAC-1669/20'.

Comparing the treatments with and without aluminum, for the cultivar Catuaí Amarelo IAC 62 in the both treatments in germination better growth occurred in aluminum nutritive solution, indicating tolerance. In the cultivar Iapar-59, the growth was same and good in both the nutritive solutions with

and without Al+ in germination and with and without after transplanting, better than 'Catuaí Amarelo IAC-62' indicating intermediate tolerance. Cultivar Obatã IAC-1669/20 developed better than 'Catuaí Amarelo IAC-62' but there was reduction in the growth when compared to nutritive solution without aluminum indicating sensitivity. Cultivar Oeiras MG 6851 presented statistically better growth with and without aluminum in the germination phase, indicating better tolerance than 'Iapar-59' (Table 2).

Table 2 - Fresh mass of the aerial part (FMAP) and root length (RL) of seedlings of *Coffea arabica* cultivars derived from seeds germinated in the absence and presence of aluminum and developed in the absence and presence of aluminum ⁽¹⁾.

Cultivars	FMAP (mg)**			
	-Al / -Al	-Al / +Al	+Al / -Al	+Al / +Al
IAC 62	317.01 bcAB	337.07 aA	268.50 cAB	256.62 bB
IAPAR 59	368.04 abAB	337.90 aB	441.98 aA	373.24 aAB
Obatã IAC-1669/20	414.38 aA	323.63 aBC	382.64 abAB	307.29 abC
Oeiras MG-6851	281.65 cA	300.16 aA	316.84 bcA	318.61 abA

Cultivars	RL (cm)*			
	-Al / -Al	-Al / +Al	+Al / -Al	+Al / +Al
IAC 62	6.49 aA	6.96 aA	6.87 bA	7.40 aA
IAPAR 59	6.20 aA	5.12 aA	6.93 bA	6.99 aA
Obatã IAC-1669/20	7.16 aAB	6.85 aB	9.33 aA	6.77 aB
Oeiras MG-6851	5.31 aB	5.26 aB	8.86 abA	7.67 aA

⁽¹⁾Means followed by the same uppercase letter in the line and lower case letter in the column do not differ by the Tukey test at 1% (***) and 5% (*), for the same characteristic evaluated. (- Al / - Al): germination and growth in the absence of Al; (- Al / + Al): only growth in the presence of Al; (+ Al / - Al): only germination in the presence of Al; (+ Al / + Al): germination and growth in the presence of Al.

Many studies have shown that root growth inhibition is the most rapid visible symptom of aluminum toxicity in the plants, resulting in root system reduction and injuries, likely leading to mineral deficiency and water stress (Degenhardt et al., 1998). In the coffee cultivars in the present study, a significant difference was observed in root length (RL) only in the treatment +Al / -Al, with the cultivars Obatã IAC-1669/20 and Oeiras MG-6851 presenting higher RL (Table 2). For these cultivars, the presence of aluminum in the solution during the germination might have stimulated the root growth, with the highest increase occurring in this phase, but not in the treatment +Al / +Al, where the presence of ion in the growth phase of the seedlings negatively affected RL, differing significantly only from the treatment +Al / -Al. In the remaining treatments (-Al / -Al and -Al / +Al), this cultivar's RL was lower, likely due to the absence of aluminum during germination, since, at

the concentration of 0.83 mmol L⁻¹. Al was not toxic for this cultivar during the germination and stimulated primary root growth. However, this cultivar was sensitive to the presence of Al during the growth phase, whose treatments (-Al / +Al and +Al / +Al) differed from the others with lower RL values (Table 2). This response was more evident in Table 3, showing a high percentage of negative variation in the root length of the cultivar Obatã IAC-1669/20 which germinated in the presence of aluminum, likely as a result of stimulus on the RL of the primary root during this cultivar's germination, promoting root elongation under hydroponics in the absence of Al while inhibiting it under hydroponics in the presence of the same cation. Mistro et al. (2007) observed that the relative tolerance index value of cultivar Obatã IAC-1669/20 was reduced, showing the sensitivity of this cultivar to aluminum, compared to cultivar Catuaí Amarelo IAC 62, suggesting tolerance of

this cultivar to aluminum at the concentration of 0.83 mmol L⁻¹. This concentration was equivalent to 45 mg L⁻¹ of Al³⁺ or 0.5 cmol_c dm⁻³, corresponding to the classification of the mean content of the element in soil, which varied from 0.4 to 1.0 cmol_c dm⁻³ (Fullin and Dadalto, 2001). In corn genotypes, the presence of toxic aluminum (100% of aluminum saturation) did not significantly reduce the diameter and height of stem, leaf area, dry matter of aerial parts, total dry matter and yield (Souza et al., 2000).

RL of cultivars Catuaí Amarelo IAC 62 and Iapar 59 did not differ significantly in the presence or absence of aluminum at different phases, showing that these cultivars presented some mechanism of tolerance to aluminum, since treatment +Al / +Al did not differ from the control (Table 2) and root growth of these cultivars was stimulated in the presence of aluminum (Table 3). Benin et al. (2004) evaluated oat genotypes and observed root growth retaking values that allowed a perfect discrimination between the sensitive and tolerant genotypes. Similar results were obtained by Freire et al. (1987) in rice, by Baligar et al. (1990) in sorghum and in wheat by Costa et al. (2003).

Cultivar Oeiras MG-6851 presented a higher RL in the treatments with the presence of aluminum during the germination, regardless of aluminum absence or presence during the seedling growth. Despite presenting a negative percentage of root length variation (Table 3), this cultivar presented some tolerance to aluminum at the concentration

of 0.83 mmol L⁻¹, due to the significant difference between the control and the +Al / +Al, treatments, with the latter presenting higher RL values. These results suggested that root system development during the seedling growth was influenced by the presence of aluminum during the germination phase for this cultivar.

For the characteristics evaluated such as height (H), dry mass of the aerial part (DMAP) and root fresh mass and dry mass (RFM and DRM, respectively), there was no interaction between the coffee cultivars and presence and absence of aluminum at different initial development phases (Table 4 A and B). In maize, a reduction was observed in the dry mass of the aerial part with increase of aluminum (Batista et al., 2009). Table 5 showed that the statistical difference between the means of the values obtained from the cultivars was significant for the four characteristics previously cited, pointing cultivar Catuaí Amarelo IAC 62 as presenting the lowest means, not differing statistically from cultivar Oeiras MG 6851, which presented the similar results. The seedlings of cultivars Iapar 59 and Obatã IAC 1669/20 presented, thus, more general development. Although showing less development, based on these characteristics, cultivar Catuaí Amarelo IAC 62 was the only one that effectively presented primary root growth increase (Table 3), indicating its tolerance to aluminum.

Table 3 - Root length (cm) of arabica coffee cultivar seedlings germinated in the absence and presence of aluminum and variation (%) in root length in response to absence (-Al) and presence (+Al) of aluminum in the nutritive solution.

Cultivars germinated in the absence of Al	Al (45 mg L ⁻¹)		Variation ⁽¹⁾ (%)
	-Al	+Al	
IAC 62	6.49	6.96	+ 7.24
Iapar 59	6.20	5.12	- 17.42
Obatã IAC-1669/20	7.16	6.85	- 4.33
Oeiras MG-6851	5.31	5.26	- 0.94
Cultivars germinated in the presence of Al	Al (45 mg L ⁻¹)		Variation ⁽¹⁾ (%)
	-Al	+Al	
IAC 62	6.87	7.40	+ 7.71
Iapar 59	6.93	6.99	+ 0.87
Obatã IAC-1669/20	9.33	6.77	- 27.44
Oeiras MG-6851	8.86	7.67	- 13.43

⁽¹⁾ Sign + indicates stimulus to root growth in the presence of aluminum and sign - indicates root growth negatively affected by the presence of aluminum.

Table 4 - Height (H), dry mass of the aerial part (DMAP), root fresh mass (RFM) and root dry mass (RDM) of *Coffea arabica*⁽¹⁾ seedlings. (A) different cultivars. (B) originated from seeds germinated in the absence and presence of aluminum and developed in the absence and presence of aluminum.

A				
Cultivars	H (cm)	DMAP (mg)	RFM (mg)	RDM (mg)
Catuaí Amarelo IAC 62	5.32 b	82.14 c	71.01 b	10.66 b
Iapar 59	6.17 a	109.28 a	84.30 a	13.04 a
Obatã IAC-1669/20	6.21 a	99.96 ab	73.15 ab	12.90 a
Oeiras MG-6851	5.77 ab	90.41 bc	61.80 b	10.82 b
B				
Aluminum	H (cm)	DMAP (mg)	RFM (mg)	RDM (mg)
- Al / - Al	5.97 ab	94.45 a	87.98 a	11.63 b
- Al / + Al	5.71 ab	99.38 a	58.37 b	11.01 b
+ Al / - Al	6.22 a	95.51 a	83.45 a	13.47 a
+ Al / + Al	5.57 b	92.45 a	60.47 b	11.31 b

⁽¹⁾Means followed by the same letter in the column do not differ by the Tukey test at 1%. (- Al / - Al): germination and growth in the absence of Al; (- Al / + Al): only growth in the presence of Al; (+ Al / - Al): only germination in the presence of Al; (+ Al / + Al): germination and growth in the presence of Al.

Except for DMAP, which was not sensitive, aluminum affected significantly all the characteristics evaluated (Table 4B). Vasconcelos et al. (2002) reported that DMAP was an insensitive parameter in detecting differential tolerance to aluminum. The lowest height value of the aerial part of the seedlings was observed in the treatment +Al / +Al, but this treatment did not differ from the control and from the -Al / +Al treatments. However, the absence of Al in the two phases of development (-Al / -Al) and its presence in the seedling development phase (-Al / +Al and +Al / +Al) affected H negatively. The highest RFM was observed in the control (-Al / -Al) and +Al / -Al treatments that differed from the others. On the other hand, the RDM of the seedlings was significantly higher only in treatment +Al / -Al.

The results obtained suggested that the presence of aluminum in the germination did not induce the cultivars to tolerate it, even at more advanced stages of development. Although many studies on the nutritive solution have been representative of field conditions, it could be suggested that these cultivars be evaluated under these conditions and at more advanced stages of development, since the response of the cultivars to aluminum toxicity could change from one stage to the other, as well as their nutritional needs.

CONCLUSIONS

The concentration of 0.83 mmol L⁻¹ of Al³⁺ resulted difference in the *arabica* coffee cultivars.

Based on the evaluation of the characteristic root length during the initial seedling growth, the cultivars Catuaí Amarelo IAC 62 and Iapar 59 could be considered tolerant to aluminum; cultivar Oeiras MG-6851 presented intermediary tolerance, and cultivar Obatã IAC-1669/20 was sensitive to the element.

The differential tolerance presented by the coffee cultivars in the presence of aluminum during the initial development of the seedlings occurred regardless of the presence of Al in the germination phase.

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