

## Ameliorating Effects of Aluminium on Low pH-induced Structure Damage to *Lotus corniculatus* Root Cells

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### ABSTRACT

Ultrastructural responses of *Lotus corniculatus* root cells to solutions with low pH 4.0 alone or with 2.0mM AlCl<sub>3</sub> were compared. The low pH induced special modifications of cell nuclei and disintegration of cytoplasmic organelles. Importantly, no cell wall thickenings were present. Al<sup>3+</sup> at the same pH did not induce as severe damage as the acidity alone. Increased vacuolation and irregular cell wall thickenings were most frequent. The effect of Al<sup>3+</sup> on cytoplasmic structure depended not only on severity of stress and on the position of a particular cell within the root, but also on the occurrence of stress-induced cell wall thickenings.

**KEYWORDS:** low pH, proton toxicity, aluminium, cell wall thickenings.

### INTRODUCTION

Acid soils are characterized by having excess H<sup>+</sup>, Mn<sup>2+</sup>, and Al<sup>3+</sup>, and concomitantly depletion of Ca<sup>2+</sup>, Mg<sup>2+</sup>, and PO<sub>4</sub><sup>3-</sup>. For vegetation it means nutrient deficiency and H<sup>+</sup>/Al<sup>3+</sup> toxicity. Primary target of proton toxicity may be linked to a disturbance of the stability in the pectic polysaccharide network (Koyama et al., 2001). Experiments in solution culture displayed statistically stronger negative correlation of relative root length with H<sup>+</sup> concentration than those associated with Al<sup>3+</sup> (Kinraide, 2003). This paper shows ameliorative effects of Al<sup>3+</sup> to H<sup>+</sup> at ultrastructural level of root cells.

### MATERIAL AND METHODS

Adventitious roots of *Lotus corniculatus* L. were grown in hydroponic culture with pH 5.5 (control), pH 4.0 without or with 2.0mM AlCl<sub>3</sub>, for 24h. For electron microscopy 2 mm long root apices were fixed with 3% glutaraldehyde and 1% OsO<sub>4</sub> both in 0.1M Na-cacodylate buffer, dehydrated in ethanol series and propylene oxide and embedded in Spurr's medium. Ultrathin sections were stained with uranyl acetate and lead citrate and investigated with EM Tesla BS 500.

### RESULTS AND DISCUSSION

Differences in the structural response of *Lotus corniculatus* roots exposed to pH 4 alone and to Al<sup>3+</sup> at the same pH in the root medium were found. Under the low pH the cell nuclei with dark central part reminding of nucleolus, surrounded by a transparent ring and a continuous envelope occurred in root cells particularly in cortex and stellar parenchyma (Fig. 1A). In the cytoplasm only fragments of membranes but no organelles were present. Importantly, there were no thickenings of the cell walls. To our best knowledge such structural responses have not been reported so far.

The effects of  $\text{Al}^{3+}$  in the solution with pH 4 were not as drastic as the same pH alone (Fig. 1B). Increased vacuolar volume was a general response. Frequently deposition of some material into the cell walls occurred (Figs. 1B, C), reminding of callose (Horst 1995, Vázquez et al. 1999). Structure of nuclei and cell organelles were quite well preserved in the cells provided with cell wall thickenings (Figs. 1B, cell 1 in C) while the structure of the cells lacking such wall modifications was damaged more severely (cell 2 in Fig. 1C). We suppose that these deposits induced by  $\text{Al}^{3+}$  might have protective role (Koyama et al., 2001) and can ameliorate a parallel effect of  $\text{H}^+$  on acid-labile coordinate bonds in the pectic polysaccharide network of cells walls and proton toxicity in general.

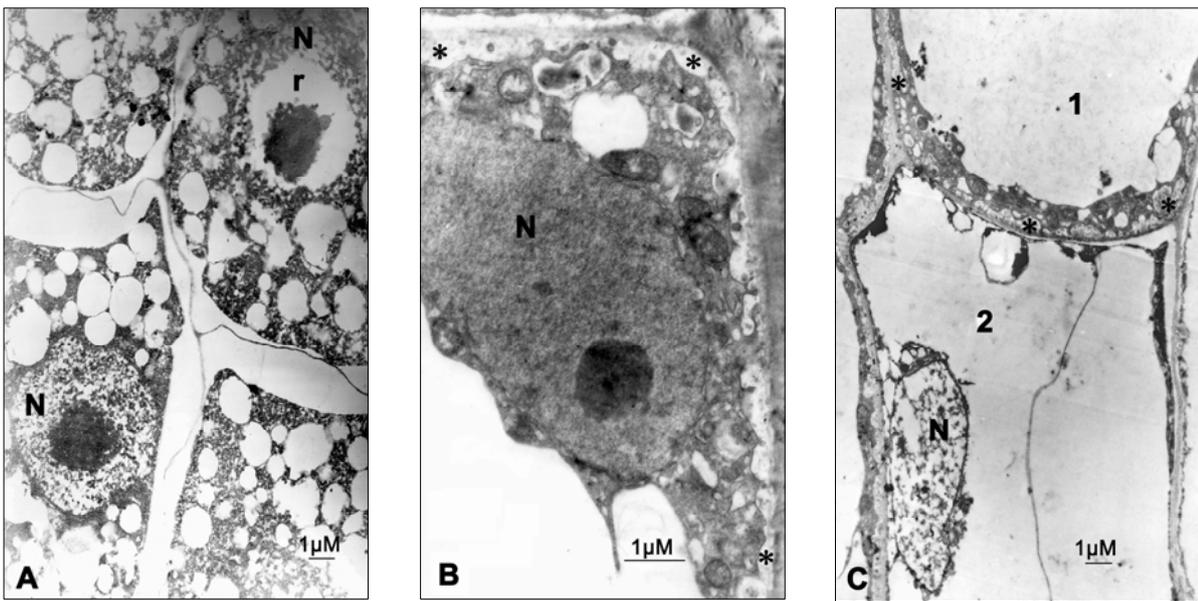


Figure 1. Root cells exposed to pH 4.0 without  $\text{AlCl}_3$  (A); and pH 4.0 with 2.0mM  $\text{AlCl}_3$  (B, C);  
N – Nucleus, r – transparent ring, \* - cell wall thickenings

#### REFERENCES

- Horst, W.J. 1995. The role of the apoplast in aluminium toxicity and resistance of higher plants: a review. *Z. Pflanzenernäh. Bodenk.* 158:419-428.
- Kinraide, T.B. 2003. Toxicity factors in acidic forest soils: attempts to evaluate separately the toxic effects of excessive  $\text{Al}^{3+}$  and  $\text{H}^+$  and insufficient  $\text{Ca}^{2+}$  and  $\text{Mg}^{2+}$  upon root elongation. *European Journal of Soil Science*, 54, 323–333.
- Koyama, H. Toda, T. and Hara, T. 2001. Brief exposure to low-pH stress causes irreversible damage to the growing root in *Arabidopsis thaliana*: pectin–Ca interaction may play an important role in proton rhizotoxicity. *Journal of Experimental Botany*, Vol. 52, No. 355, pp. 361-368.
- Vázquez, M.D. Poschenrieder, C. Corrales, I. and Barceló, J. 1999. Change in apoplastic aluminum during the initial growth response to aluminum by Roots of a Tolerant Maize Variety. *Plant Physiology*, 119, pp. 435–444.

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