Aluminum-induced exudation of organic acid anions from the DTZ of *Zea mays* (L.) root apices is mediated by an anion channel

Malte Kollmeier[1], P. Dietrich[2], C. S. Bauer[2], R. Hedrich[2] and Walter J. Horst[1]

[1]Institute of Plant Nutrition, University of Hannover, Herrenhäuser Str. 2, D-30419 Hannover

[2]Lehrstuhl für molekulare Pflanzenphysiologie und Biophysik, Julius-von-Sachs Institut für Biowissenschaften, University of Würzburg, Julius-von-Sachs-Platz 2, D-97082 Würzburg

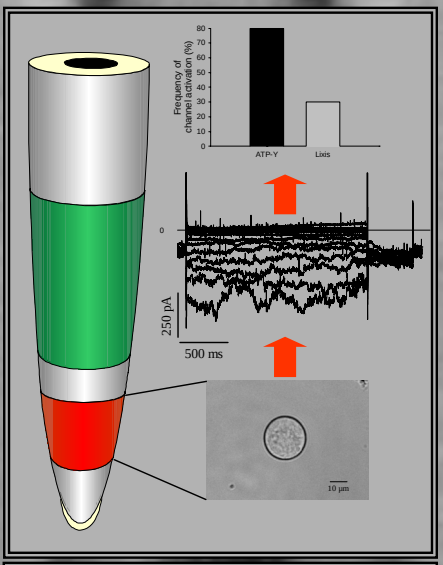
INTRODUCTION

Aluminum (Al) toxicity is a major crop yield limiting factor worldwide, particularly in the strongly weathered acid soils of the tropics. The exudation of organic acid anions such as oxalate, citrate and malate from the root has been reported to be a mechanism of Al resistance. This is due to the capacity of these molecules to chelate the most rhizotoxic monomeric Al species to non toxic complexes. Focusing on genotypical differences and the spatial Al-sensitivity of the maize root apex, two hypotheses were examined in this study: (a) Does Al stimulate the exudation of organic acid anions especially from the root apex? (b) Do anion channels play a role in the exudation process?

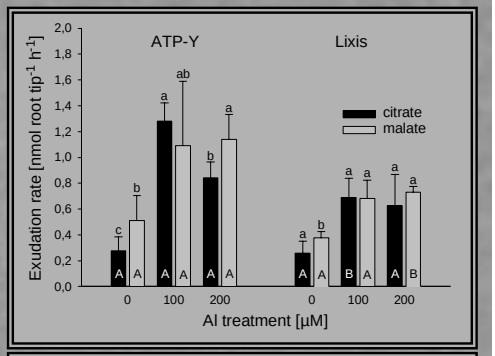
MATERIALS & METHODS

All experiments were conducted with selected three-day-old seedlings of the maize cultivars ATP-Y (Al-resistant) and Lixis (Al-sensitive). Five mm root tips of intact seedlings were incubated for 2 h in 4 mL of a solution containing 200 µM $CaCl_2$ 0, 50, 100 or 200 µM $AlCl_3$ (pH 4.3). The organic acid anions exuded were analyzed using HPLC.

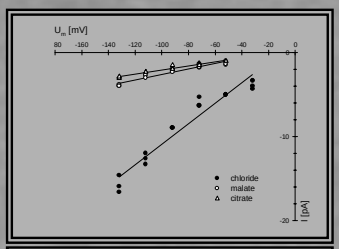
Protoplasts were enzymatically digested from two root zones: the DTZ (red on the right hand figure) and EZ (green) after pretreatment of the intact roots in an agarose gel containing NS ± 90 µM $AlCl_3$ for 1 h. All patch-clamp experiments were performed in the whole-cell configuration. The standard pipette solution consisted of (in mM) 100 TEACl, 2 $MgCl_2$, 2 MgATP, 2 EGTA, 10 Hepes/Tris (pH 7.2; 620 mosmol kg^{-1}), the standard bath solution was composed of (in mM) 20 TEACl, 1 $CaCl_2$, 5 MES, ± 0.05 $AlCl_3$ (pH 4.3; 600 mosmol kg^{-1}).



Preincubation of intact root apices with 90 µM Al for 1 h induced inward currents in 80 % of the DTZ protoplasts from the Al-resistant and in 30 % from the Al-sensitive cultivar



Incubation of 5 mm root apices of intact plants in 100 µM Al for 2 h stimulated the exudation rate of citrate and malate more in the Al-resistant than the Al-sensitive cultivar

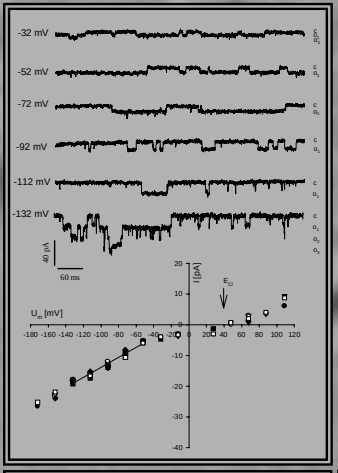


The Al-induced large conductance anion channel is permeable to citrate and malate as indicated by the single channel conductance after replacing the chloride in the pipette with citrate or malate

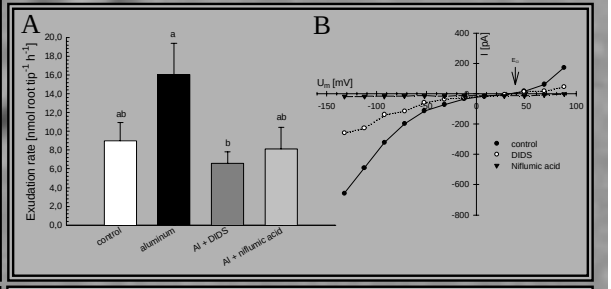
CONCLUSIONS

The results presented here clearly demonstrate a stimulative effect of Al on the exudation rate of organic acid anions from the root apex particularly in the Al-resistant cultivar.

From the results gained in whole plant and patch-clamp experiments, we conclude that an Al-induced large conductance anion channel in the plasma membrane of cortical DTZ cells accounts for the release of these anions capable of rendering Al non-phytotoxic.



Single channel fluctuations in the whole-cell configuration reveal a large conductance of 144 pS in the linear range between –132 and –32 mV



Application of the anion channel inhibitors niflumic acid or DIDS led to a considerable decrease in the total exudation rate of organic acid anions from intact root apices (A) as well as macroscopic inward currents (B) and single channel conductance (not shown)

We gratefully acknowledge the financial support by the German Research Foundation (DFG, SSP 717) to WJH