EFFECTS OF Pb IONS ON SLOWLY ACTIVATING CHANNELS IN THE RED BEET VACUOLAR MEMBRANE

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It is presently well established that two major ion currents in the vacuolar membrane of higher plants are mediated by non-selective cation channels, the so-called SV channel (slowly activating) and the FV channel (fast activating). The FV channel conducts various monovalent cations with poor selectivity, whereas the SV channel conducts K^+, Na^+, Mg^{2+}, Ba^{2+} and Ca^{2+}. Under conditions usually applied to patch-clamp experiments on vacuoles (symmetrical K^+, milimolar luminal Ca^{2+}) SV channel activates at positive potentials. Here we studied the effects of cytoplasmic Met_3PbCl (trimethyllead chloride) on SV channels in the red beet vacuolar membrane. Red beet (*Beta vulgaris* L.) vacuoles were isolated mechanically and transferred to the experimental chamber. The SV channels were studied by the patch-clamp technique in the whole-vacuole or the (cytoplasmic side-out) excised-patch configuration using EPC-7 amplifier (List-Medical-Electronic, Darmstadt, Germany). Data were analysed using software Pulse and PulseFit (HEKA Electronic, Lambrecht, Germany). Patch pipettes were prepared from Kimax-51 glass capilars and coated with Sylgard.

It was found that under symmetrical K^+ concentrations (100 mM K^+ on both sides of the vacuolar membrane) SV channels mediate outward currents only, which corresponds to K^+ uptake into the vacuole. Macroscopic whole-vacuole SV currents elicited by a series of positive voltages were in the range of a few hundred pA to 10 nA. The single channel records display a slow activating channel with a conductance in the order 70 pS. It was also found that cytoplasmic Met_3PbCl inhibits SV channels in the red beet vacuolar membrane. These results are discussed on the basis of cation-dependent gating of the SV channel.

This work was supported by the KBN grant 2 PO 4G 089 27.