

## **Mercury uptake kinetics by white lupin roots**

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### **ABSTRACT**

Kinetics of Hg uptake by white lupin roots was studied at 20 °C and ice-cold conditions. At 20°C, Hg uptake showed two components, active influx of Hg and passive accumulation of Hg, which can be monitored at ice-cold conditions. Ice-cold Hg-uptake showed a linear behaviour. The difference between Hg uptake at 20 °C and ice-cold conditions was best fitted to a hyperbolic curve, which would indicate the presence of an active transport of Hg across the root membrane mediated by a carrier. Values obtained are  $K_m = 149 \mu\text{M}$  and  $V_{\max} = 3.6 \mu\text{mol Hg g}^{-1} \text{FW h}^{-1}$ . Data obtained at 20 °C were best fitted to a hyperbola with linear compound, giving the kinetic parameters the values of  $K_m = 218 \mu\text{M}$  and  $V_{\max} = 3.8 \mu\text{mol Hg g}^{-1} \text{FW h}^{-1}$ , quite similar to those obtained from the corrected hyperbolic function. On the other hand, Hg is somehow accumulating in plant material under ice-cold conditions. Further investigations are needed to elucidate whether Hg is entering inside plant cells at  $<2 \text{ }^\circ\text{C}$ , by way of an ionic channel or just an adsorption phenomenon is taking place.

### **1. INTRODUCTION**

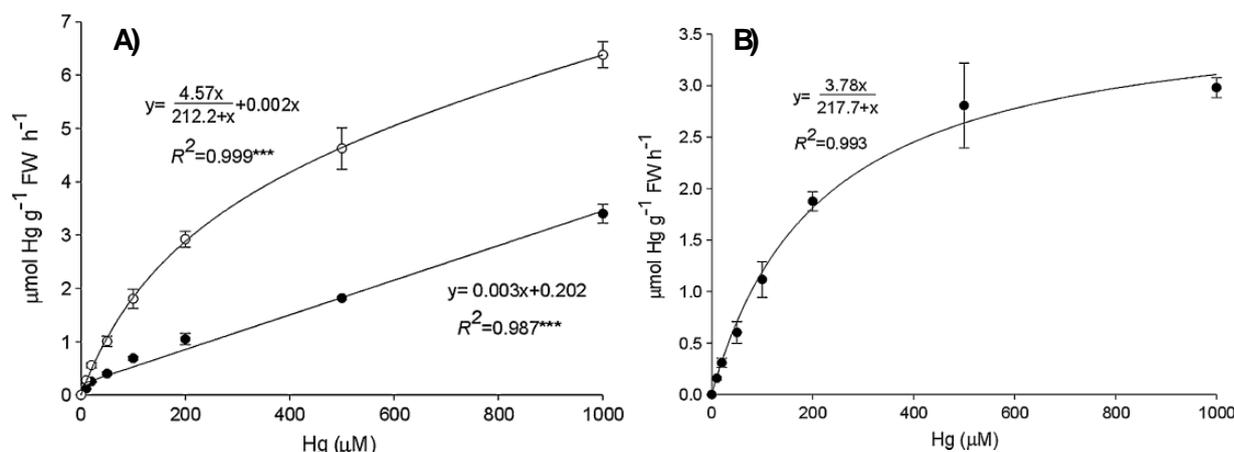
Mercury is a pollutant, whose levels in soil have been increased nowadays due to human activities (Moreno-Jiménez et al., 2006). It can induce toxicity symptoms in plants such as inhibition of plant growth or disturbances on water and nutrient uptake. *Lupinus albus* L. (white lupin) is a nitrogen-fixing legume that can live in degraded soils. This plant species has been reported as a candidate in the phytostabilisation of soils with trace elements, and its use have been evaluated in Hg-polluted soils (Rodríguez et al., 2003). There is a lack of knowledge about kinetics parameters of Hg uptake by higher plants. Ion transport across cellular membrane occurs by the activity of transporters. Transmembrane transporters are characterized by kinetic parameters, such as maximum rate of ion transport across the cellular membranes ( $V_{\max}$ ) and affinity for ion ( $K_m$ ). The objective of our study is to evaluate Hg uptake by lupin roots.

### **2. MATERIALS & METHODS**

Seedlings were grown for 2 weeks in a growth cabinet. Three replicate samples of whole roots (excised at the node) were used. Roots were incubated in aerated solution containing 0.5 mM  $\text{CaCl}_2$ , 2 mM MES and 0, 10, 20, 50, 100, 200, 500 or 1000  $\mu\text{M}$  Hg at pH 6.0 for 20 min both at 25 °C and ice-cold ( $<2 \text{ }^\circ\text{C}$ ) conditions. After root incubation, plant material was rinsed in a fresh ice-cold solution containing 5 mM  $\text{CaCl}_2$  and 5 mM MES at pH 6.0 for 2 min and incubated in a fresh ice-cold nutrient solution of the same composition during 15 min for desorbing out no-uptaken Hg. Fresh weights of the roots were determined before analysis. Each plant root was mineralised as Lozano-Rodríguez et al. (1995) described. Mercury in the extracts was measured by Advanced Mercury Analyser (AMA-254, LECO Company). Quality and Hg losses controls were carried out during sample processing.

### 3. RESULTS & DISCUSSION

At 20 °C, Hg<sup>2+</sup> influx was graphically resolved into a saturable (hyperbolic) component and a non-saturable linear component (Fig. 1A), while only a non-saturable linear component was best fitted for the ice-cold Hg influx. The saturable component was more clearly observed for the lower Hg external concentrations, while the linear component was more evident at the highest Hg concentrations. A significant reduction of Hg influx was observed in the ice-cold experiments, where metabolic energy-dependent uptake processes should be minimal. So the ice-cold influx could be attributed to apoplastic adsorption of Hg that remained bound to the cell walls after desorption, while Hg influx at 20 °C should include an active component in Hg influx, suggesting the existence of a root transporter that can be used for Hg uptake into root cells. When subtracting influx data obtained for 20 °C and ice-cold experiments, the contribution of the saturable component will only remain. These data were best fitted to a hyperbola (Fig. 1B), with  $K_m$  and  $V_{max}$  values for the saturable component of 218  $\mu\text{M}$  and 3.8  $\mu\text{mol Hg g FW}^{-1} \text{h}^{-1}$ .



**Figure 1.** Hg uptake by roots of *L. albus*: A) Uptake at different temperatures: 20 °C (open circles) or ice-cold (<2 °C, full circles); B) Saturable component of Hg uptake, obtained by subtracting Hg influx data at 20 °C and ice-cold. Vertical bars indicate standard error (n=3).

No previous information about the kinetic parameters of Hg uptake in plants has been reported. An active component on Hg influx was found in our experiment. Pandey and Singh (1993) described the Hg<sup>2+</sup> uptake pattern of a cyanobacterium, finding also an active component in Hg uptake. By examining the various essential nutrients as grouped by their chemical properties in the periodic table, we can identify the closest chemical relatives of Hg as Zn, Cu or Fe, so that their transporters could be possible candidates to facilitate Hg influx. Calcium channels or even aquaporines could also partly contribute to this high Hg uptake ability of white lupin. Further investigations are needed to elucidate whether Hg is entering inside plant cells at <2 °C, by way of an ionic channel or just an adsorption phenomenon is taking place.

### REFERENCES

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