# **Cadmium and copper interactions on the accumulation and distribution of Cd and Cu in birch** *(Betula pendula* **Roth) seedlings**

M. Gussarsson, S. Adalsteinsson, P. Jensén and H. Asp

*Department of Horticulture, Swedish University of Agricultural Sciences, P. O, Box 55, S-23053 Alnarp, Sweden* 

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# **Abstract**

The effect of different external cadmium (Cd) and copper (Cu) regimes on the concentration of Cd and Cu in roots and shoots of birch *(Betula pendula* Roth.) seedlings was investigated. The seedlings were grown for 12 days in a weak nutrient solution (containing all essential nutrient elements including  $0.025 \mu M$  Cu) at pH 4.2 with combinations of additional 0-2  $\mu$ M CdCl<sub>2</sub> and 0-2  $\mu$ M CuCl<sub>2</sub>. Root and shoot concentrations of Cu were decreased by Cd in all treatments which included  $0.1-2 \mu M$  of additional Cu in the treatment solution. When no extra Cu was added, only the shoot concentration of Cu was decreased by Cd whereas the root concentration was not affected. The shoot concentration of Cd was decreased by 0.5 and 2  $\mu$ M of additional Cu in the treatment solution. The root concentration of Cd was decreased by Cu only when the concentration of additional Cu in the treatment solution was equal to or exceeded the concentration of Cd.

# **Introduction**

Together with industrial and urban activities, low soil pH values increase the availability of heavy metals, including Cd, in the soil solution (Berggren, 1989). Cd may interfere with the uptake of positively charged ions in plant roots (Walker et al., 1977) and with element translocation to the shoot (Cunningham, 1977; Lamoreaux and Chaney, 1977). Heavy metal accumulation in plants is often restricted to the root tissue, with only small amounts transported to the shoot (Taylor, 1987 and references therein). Adsorption of heavy metals to negatively charged constituents of the cell wall may play an important role in the exclusion of metals from the symplast (Cumming and Taylor, 1990). Intracellular metal detoxification may include metal-induced synthesis of intracellular metal-binding organic compounds (Reese and Wagner, 1987; Weigel and Jäger, 1980).

The purpose of this work was to study the interaction of Cd and Cu on the accumulation and distribution of Cd and Cu in birch *(Betula pendula)* seedlings.

#### **Material and methods**

Seeds of birch *(Betula pendula* Roth.) were germinated in vermiculite soaked with distilled water. After 38 days 100 seedlings were transferred to styrofoamenclosed foam rubber discs floating on 30 L standard nutrient solution containing (in  $\mu$ *M*): 100 Ca(NO<sub>3</sub>)<sub>2</sub>, 100 KCI, 100 NH4NO3, 25 KH2PO4, 50 MgSO4, l0 Fe-EDTA, 3 MnSO<sub>4</sub>, 2 H<sub>3</sub>BO<sub>3</sub>, 0.025 ZnCl<sub>2</sub>, 0.025  $Na<sub>2</sub>MoO<sub>4</sub>$  and 0.025 CuCl<sub>2</sub>. The initial pH was 4.2. The nutrient solution was renewed gradually by adding 25 mL fresh solution every 6th min while keeping the level constant with an outlet for excess solution. After 22 days, 60 plants were selected for uniformity. Each plant was transferred to a PVC/PE holder floating on 350 mL standard nutrient solution in a 400 mL glass beaker. Each beaker was wrapped in black plastic sheets to keep light from the root system. The solutions were changed every second day. After 6 days Cd and the extra Cu were added to the nutrient solutions. Each concentration of CdCl<sub>2</sub> (0, 0.5 and 2  $\mu$ M) was complemented with additional CuCl $_2$  (0, 0.1, 0.5) and 2  $\mu$ M). There were five replicates (plants) in each treatment, and solutions continued to be changed every second day for another 12 days. At harvest (78 d old plants), the roots were rinsed for 10-15 sec in a standard nutrient solution to remove adhering treatment



*Fig. 1.* Concentration of Cu and Cd in shoots and roots of birch after a 12 day exposure to 0–2  $\mu$ M CuCl<sub>2</sub> combined with either 0 (empty columns), 0.5 (hatched), or 2  $\mu$ M CdCl<sub>2</sub> (filled). Values are means of 5 replicates. SE is given by vertical bars.

solution from the root system. Roots and shoots were separated and dried at 70 °C for 3 days prior to dry weight determination and wet-combusted in concentrated  $HNO<sub>3</sub>$  in a microwave oven (CEM, MDS 81) D). Cd and Cu were determined by atomic absorption spectrophotometry with a graphite furnace.

All cultivation solutions were continuously aerated. The plants were grown in a climate chamber at  $20\pm1$  $°C$ , 50 $\pm$ 5 % RH and a photoperiod of 16 h (fluorescent tubes,  $182 \pm 10 \ \mu$  mol m<sup>-2</sup> s<sup>-1</sup>).

## **Results and discussion**

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The concentrations of both heavy metals in shoots and roots increased with increasing Cd and Cu concentration in the ambient solution (Fig. 1). Both Cd and Cu were accumulated mainly in the root. This corroborates earlier studies on heavy metal distribution in birch (Gussarsson and Jensén, 1992). Both root and shoot concentrations of Cu were decreased by Cd in the treatments, which included  $0.1-2 \mu M$  of additional Cu in the treatment solution. However, when no extra Cu was added, only the shoot concentration was decreased while the root concentration of Cu was not affected. Ions with the same valency may compete for uptake sites, thereby limiting the uptake of each other (Walker et al., 1977). This hypothesis is strongly supported by the interactive effects of Cd on the root concentration of Cu reported in this paper but it does not explain the absence of Cd effects on the Cu concentration in roots not exposed to additional Cu (Fig. 1). On the contrary, since the concentration of Cd in the treatment solution greatly exceeded that of Cu, a strong inhibitive effect of Cd on Cu uptake would be expected. However, if the competitive effect of Cd on Cu uptake was counteracted by increased capacity for intracellular Cu accumulation in the root, the root concentration of Cu may have been maintained and decreased shoot concentration of Cu would solely reflect the interactions in heavy metal uptake (Fig. 1). Cd may induce synthesis of metal binding organic substances for metal detoxification in the cytoplasm (Reese and Wagner, 1987; Weigel and Jäger, 1980). A limitation in ion binding specificity of these compounds would result in increased accumulation of both metals in the root and decreased metal translocation to the shoot. At higher Cu concentrations in the nutrient solution, the intracellular accumulation of Cu in the root may have been overshadowed by the competitive uptake between Cu and Cd (Fig. 1).

Root and shoot concentrations of Cd were both decreased by Cu only when the concentrations of additional Cu in treatment solutions equalled or exceeded the concentration of Cd (Fig. 1). Decreased concentration of Cd in roots and shoots may be caused by interactive ion uptake described earlier. When the plants were exposed to 2  $\mu$ M Cd combined with 0.5  $\mu$ M additional Cu, only the shoot concentration was decreased whereas the root concentration was not affected. The results of this study support earlier suggestions that Cd can be accumulated specifically by roots.

## **Acknowledgement**

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## **References**

- Berggren D 1989 Speciation of aluminum, cadmium, copper, and lead in humic soil solutions - a comparison of the ion exchange column procedure and equilibrium dialysis. Int. J. Environ. Anal. Chem. 35, 1-15.
- Cumming J R and Taylor G J 1990 Mechanisms of metal tolerance in plants: physiological adaptations for exclusion of metal ions from the cytoplasm. *In* Stress Responses in Plants: Adaptation and Acclimation Mechanisms. Eds. R G Alsher and J R Cumming. pp 329-356. Wiley-Liss, New York, USA.
- Cunningham L M 1977 Physiological and biochemical aspects of cadmium in soybean: The effects of induced Cd toxicity on the
- uptake and translocation of Zn, Fe, Mg, Ca and K. *In* Trace Substances in Environmental Health. Ed. D D Hemphill. pp 133-147. University of Missouri, Colombia, USA.
- Gussarsson M and Jensén P 1992 Effects of copper and cadmium on uptake and leakage of K + in birch *(Betula pendula)* roots. Tree Physiol. 11, 305-313.
- Lamoreaux R J and Chaney W R 1977 Growth and water movement in silver maple seedlings affected by cadmium. J. Environ. Qual. 6, 201-205.
- Reese R N and Wagner G J 1987 Effects of buthionine sulfoximine on Cd-binding peptide levels in suspension-cultured tobacco cells treated with Cd, Zn, or Cu. Plant Physiol. 84, 574-577.
- Taylor G J 1987 Exclusion of metals from the symplasm: a possible mechanism of metal tolerance in higher plants. J. Plant Nutr. 10, 1213-1222.
- Walker W M, Miller J E and Hassett J J 1977 Effect of lead and cadmium upon the calcium, magnesium, potassium, and phosphorus concentration in young corn plants. Soil Sci. 124 145-151.
- Weigel H J and Jäger H J 1980 Subcellular distribution and chemical form of cadmium in bean plants. Plant Physiol. 65,480-482.