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DESFERRIOXAMINE B vs. EDTA FOR IMPROVING THE PHYTOEXTRACTION OF METALS BY SUNFLOWER

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Introduction

Inoculation of siderophore-producing bacteria (SPB) has been proposed as a strategy to optimize metal phytoextraction (Rajkumar et al., 2010). The localized and continuous production of siderophore in the close vicinity of plant roots, where most of the SPB are established, would help in promoting metal phytoextraction with minimizing the risk of metal leaching. However, there is a need to dissect how siderophores interact with metals at the soil-root interface to better assess the potential of coupling phytoextraction with the inoculation of SPB. The present study focuses on desferrioxamine B (DFOB), the main siderophore produced by the actinobacteria *Streptomyces pilosus*. DFOB is characterized by a high selectivity for Fe(III) (10^{31} M^{-1}) but also by a good affinity for divalent metals including Cd(II) ($10^{7.9}$ M^{-1}), Cu(II) ($10^{14.1}$ M^{-1}) and Zn(II) ($10^{11.1}$ M^{-1}). The purpose of this work was to compare the efficiency of DFOB vs. EDTA for the phytoextraction of metals by sunflower. The main goal was to dissect the impact both chelators have on the mobility of metals in soil, their speciation in porewater and their accumulation in plant shoots, for two cultivars of sunflower.

Methods

Two cultivars of sunflower with contrasted pattern of metal repartition (Laporte et al., 2015) were grown for 28 days on an agricultural soil (pH 6.7) contaminated in Cu, Cd, Pb and Zn (339, 4.9, 1120 and 602 mg kg^{-1} soil, respectively). Four days after transplanting, DFOB was supplied at the concentration of 200 µmol kg^{-1} soil to mimic bacterial production. The same procedure was performed for EDTA. Porewater was extracted every week using Rhizon® soil moisture samplers. At harvest, xylem sap was collected by the "root pressure" method before roots and shoots were freeze-dried, weighed and digested in a mix HNO_3-H_2O_2. The concentrations of metals (Al, Fe, Cd, Cu, Ni, Pb, Zn) in xylem sap and plant tissues were assayed by ICP-MS. In porewater, the chemical speciation of Cd, Cu, Pb and Zn was addressed by pseudopolarography (Bravin et al., 2012) while the concentrations of DFOB and EDTA were determined using Chrome Azurol S reagents.

Results

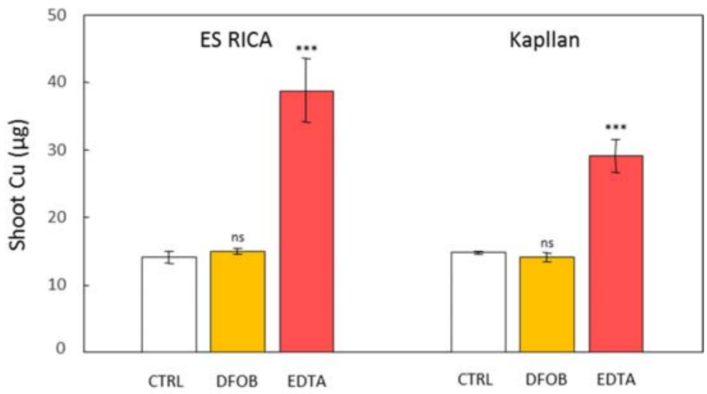
DFOB showed a stronger affinity for soil constituents than EDTA. DFOB supply increased selectively the mobility of Fe and Al in soil. Conversely, the supply of EDTA increased the mobility of Fe together with that of divalent metals. DPASV measurements highlighted how the presence of EDTA decreased strongly the free ionic fraction of metals in porewater, e.g. by 99% for Cd and 94% for Zn (Table 1). DFOB supply did not alter the accumulation of metals in sunflower shoots while the supply of EDTA enhanced selectively that of Cu and Ni (Figure 1). The promoting effect of EDTA supply towards the phytoextraction of Cu and

Ni was more pronounced for the cultivar ES RICA, whose ability to sequestrate metals in roots is known to be lower (Laporte et al., 2015).

Table 1. Total concentrations, chemical speciation and labile concentrations of Cd, Cu, Pb and Zn measured in porewater by differential pulse anodic stripping voltammetry (DPASV).

	Metal	Total conc. (µM)	Chemical speciation (%)			Labile conc. (µM)
			Labile	Dissociable	Inert	
Control	Cd	0.12 ± 0.02	100	0	0	0.12 ± 0.02
	Cu	2.2 ± 0.3	37 ± 4	18 ± 5	46 ± 8	0.81 ± 0.17
	Pb	nd	nd	nd	nd	nd
	Zn	46 ± 4	100	0	0	46 ± 4
EDTA	Cd	1.08 ± 0.26	1.3 ± 0.6	42 ± 6	56 ± 6	0.012 ± 0.005
	Cu	149 ± 15	6.1 ± 1.0	94 ± 1	0	9.1 ± 2.0
	Pb	1.25 ± 0.25	1.8 ± 0.6	51 ± 11	48 ± 10	0.022 ± 0.008
	Zn	353 ± 62	4.7 ± 0.6	10 ± 3	86 ± 4	13 ± 8

Figure 1. Amount of Cu recovered in shoots, in the sunflower cultivars ES RICA and Kapllan grown for 28 days on a metal contaminated soil supplied with DFOB or EDTA.



Conclusion

This works provides new insights on the mechanisms by which the supply of chelators (either natural or synthetic) may promote the phytoextraction of metals in soil. DFOB seems to exhibit a too strong affinity for Fe(III) to complex and thus mobilize divalent metals in soil. The specific increase in the phytoextraction of Cu and Ni observed when a low dose of EDTA was supplied is encouraging but raises questions. Further works are required to know if this specificity is linked to the kinetics of dissociation of Cu- and Ni-EDTA complexes at the root surface.

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