INFLUENCE OF ARBUSCULAR MYCORRHIZAL FUNGI AND CHELATING AGENTS (EDTA AND EDDS) ON PHYTOEXTRACTION OF ZN FROM CONTAMINATED SOIL USING *SOLANUM NIGRUM*

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CHELATE-ENHANCED PHYTOEXTRACTION OF TOXIC METAL CONTAMINATED SOILS

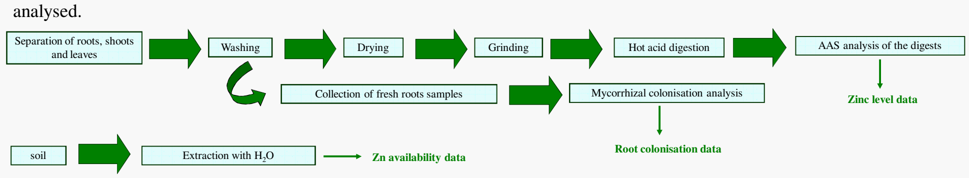
Phytoextraction appears as a low cost and environmental-friendly solution for the remediation of soils contaminated with heavy metals. Although several conditions must be met for the adequate effectiveness of phytoextraction to be achieved, the bioavailability of metals to plant roots is considered to be a critical requirement for metal uptake to occur. The addition of chelating agents, such as EDTA (ethylenediaminetetraacetic acid) and EDDS (S,S-ethylenediaminedissucinic acid) can increase metal availability and thus enhance the uptake and accumulation of heavy metals by the plant.

Biotic factors, such as symbiotic associations with arbuscular mycorrhizal fungi (AMF) can also affect metal uptake by the plants and enhance plant growth.

S. nigrum is an indigenous plant species to a metal polluted site in an industrialised area of Northern Portugal containing sediments with levels of metals above the limits established by EC Directive 86/278/EC, the main occurring metal in those sediments being zinc. Despite this contamination scenario, *S. nigrum* is abundant on the site. Previous studies with this plant showed that *S. nigrum* was able to accumulate up to 3800 mg Zn/kg dry tissue in the roots with no visible toxicity signs. In the present study, the influence of the addition of the chelating agents EDDS and EDTA, and their relationship with the presence of AMF isolates, on the bioavailability of Zn in contaminated soils, was assessed under greenhouse conditions. Metal uptake and Zn accumulation and localisation in *S. nigrum* is reported.

MATERIALS AND METHODS

The experiment was a factorial design with two matrix Zn levels [contaminated soil from the banks of Esteiro de Estarreja, with 426 mg Zn kg^{-1} dry soil (L) and the same soil after spiking with 500 more mg Zn kg^{-1} dry soil (L+Zn)], three mycorrhizal fungi treatments (no AMF, *Glomus claroideum* and *Glomus intraradices*) and three chelating agents treatments (no agent and EDTA or EDDS addition at a rate of 0.5 g kg^{-1} dry soil ten days before harvest). At harvest soil and plants were collected and analysed.



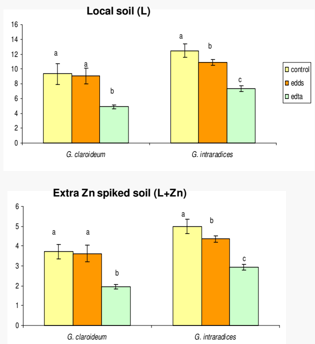


Figure 1: Percentage of mycorrhizal colonisation (%) for *G. claroideum* and *G. intraradices* in *S. nigrum*

Means with different letters for the same AMF are significantly different from each other (P < 0.05) according to Tukey test

	AMF species	Zn (mg kg^{-1} dry tissue) leaves	stems	roots
Local soil				
Control	No fungi	531 ± 21 a	1431 ± 157 a	1356 ± 359 a
	G. claroideum	656 ± 35 b	1938 ± 134 b	1798 ± 148 b
	G. intraradices	851 ± 69 c	2482 ± 40 c	2687 ± 49 c
EDDS	No fungi	764 ± 55 a	3068 ± 615 a	2769 ± 541 a
	G. claroideum	1016 ± 20 b	2855 ± 343 a	3409 ± 561 a
	G. intraradices	1020 ± 52 b	2899 ± 205 a	3095 ± 40 a
EDTA	No fungi	506 ± 55 a	2190 ± 293 a	2461 ± 343 a
	G. claroideum	1191 ± 214 b	3747 ± 912 b	3069 ± 58 b
	G. intraradices	978 ± 108 b	2809 ± 183 ab	3150 ± 277 b
Local + Zn soil				
Control	No fungi	1434 ± 161 a	3690 ± 441 a	5628 ± 388 a
	G. claroideum	2542 ± 525 b	5446 ± 490 b	7465 ± 1662 a
	G. intraradices	2082 ± 180 b	4423 ± 464 a	6399 ± 429 a
EDDS	No fungi	3435 ± 455 a	8267 ± 1250 a	6463 ± 83 a
	G. claroideum	3627 ± 228 a	7317 ± 439 a	7363 ± 150 a
	G. intraradices	3221 ± 84 a	4787 ± 501 b	7140 ± 997 a
EDTA	No fungi	4735 ± 480 a	7028 ± 414 a	7948 ± 514 a
	G. claroideum	3444 ± 430 b	6236 ± 415 b	6953 ± 53 b
	G. intraradices	3070 ± 248 b	6474 ± 215 ab	6816 ± 359 b

Table 1: Effects of the addition of chelating agents and AMF species on *S. nigrum* Zn accumulation

Means with different letters in the same treatment group are significantly different from each other (P < 0.05) according to Tukey test

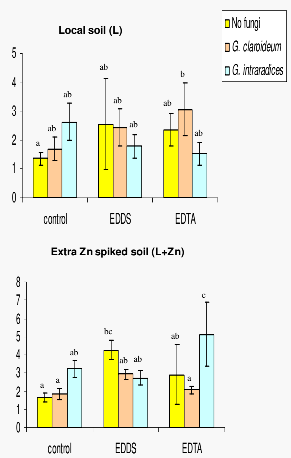


Figure 2: Percentage of Zn (mass) uptake to the plant stems from the tested soils

Means with different letters are significantly different from each other (P < 0.05) according to Tukey test

Figure 3: Plant samples grown in local (L) and extra Zn spiked soil (L+Zn)

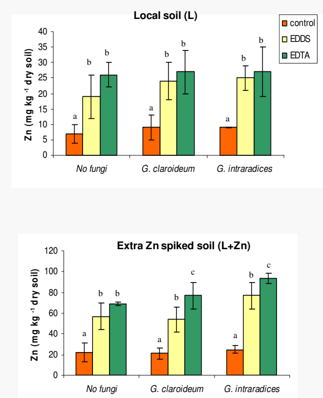


Figure 4: Effects of the addition of chelating agents on the Zn availability in tested soils

Means with different letters for the same AMF are significantly different from each other (P < 0.05) according to Tukey test

RESULTS AND CONCLUSIONS

- Soil contamination and the addition of chelating agents influenced negatively mycorrhizal colonisation in *S. nigrum*
- Plants biomass was negatively influenced by the Zn contamination in the soil
- AMF colonisation did not influence the biomass yield of *S. nigrum* neither did the addition of EDTA or EDDS
- Inoculation with *Glomus claroideum* or *Glomus intraradices* enhanced the uptake and accumulation of Zn by *S. nigrum*
- Zinc concentration in water-extracts of the soils collected at the time of harvest was increased in soils with added EDTA or EDDS by up to four and three folds, respectively → the accumulation of zinc in all plant tissues increased: *S. nigrum* plants accumulated up to 4735, 8267 and 7948 mg Zn kg^{-1} in the leaves, stems and roots, respectively
- Neither EDTA nor EDDS had an effect on the root to shoot translocation in *S. nigrum*
- All the tested treatments improved the decontamination abilities of the plant, with the addition of EDTA to plants inoculated with AMF inducing higher amount of zinc removal from both tested soils
- Considering the best scenario of metal removal from the soils (application of EDTA and inoculation with AMF), the obtained offtakes would imply a minimum sequence of 13 to 14 crops to achieve soil clean-up (compared with 23 to 42 crops without chelating agents addition)

Inoculation with AMF and application of chelating agents enhanced the phytoextraction abilities of *S. nigrum*

ACKNOWLEDGEMENTS

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