Hyperaccumulators

Compiled by Mary Stevanus
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National Agricultural Library

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   Descriptors: alyssum, nickel, cobalt, zinc, metal-ions metal-tolerance ion-uptake roots, growth-rate translocation, shoots, mineral-content malic-acid serpentine-soils alyssum-bertolonii bioaccumulation
   Abstract: Alyssum bertolonii Desv., a crucifer which grows only in Tuscany on serpentine soil, is characterized by high Ni concentration in its leaves and a high tolerance to this element. In this research, the Co and Zn tolerance of A. bertol onii has
been tested and compared with that of Ni. Although the Zn concentration in the serpentine soil on which the plant grows is lower than that of Co, root growth showed no significant difference for these two elements. In excised roots, on the other hand, Co and Zn compete with Ni uptake and accumulation. Seedlings do not show any inhibition in the translocation of these three elements. They are all rapidly carried to and accumulate in the shoots, but the uptake and translocation of Zn does not show the saturation trend found for Ni and Co. The same gel filtration fractions contain the three elements in association with malic acid, involved in Ni tolerance.

2. **Al inhibits both shoot development and root growth in als3, an Al-sensitive Arabidopsis mutant.**
   Larsen, P. B., Kochian, L. V., and Howell, S. H.
   NAL Call #: 450-P692
   **Descriptors:** arabidopsis-thaliana shoots, plant-development roots, growth, inhibition, leaves, aluminum, phytotoxicity, injuries, stress-response callose, biosynthesis, mutants, wild-strains genotypes, genetic-variation phenotypes, symptoms, plant -morphology plant-anatomy ultrastructure-
   **Abstract:** In als3, an Al-sensitive Arabidopsis mutant, shoot development and root growth are sensitive to Al. Mutant als3 seedlings grown in an Al-containing medium exhibit severely inhibited leaf expansion and root growth. In the presence of Al, unexpanded leaves accumulate callose, an indicator of Al damage in roots. The possibility that the inhibition of shoot development in als3 is due to the hyperaccumulation of Al in this tissue was examined. However, it was found that the levels of Al that accumulated in shoots of als3 are not different from the wild type. The inhibition of shoot development in als3 is not a consequence of nonspecific damage to roots, because other metals (e.g. LaCl3 or CuSO4) that strongly inhibit root growth did not block shoot development in als3 seedlings. Al did not block leaf development in excised als3 shoots grown in an Al-containing medium, demonstrating that the Al-induced damage in als3 shoots was dependent on the presence of roots. This suggests that Al inhibition of als3 shoot development may be a delocalized response to Al-induced stresses in roots following Al exposure.

3. **The consequence of selection for copper tolerance on the uptake and accumulation of copper in Mimulus guttatus.**
   Tilstone, G. H. and Macnair, M. R.
   NAL Call #: 450-An7
   **Descriptors:** mimulus-guttatus copper, metal-tolerance mineral-uptake selective-breeding genotypes, concentration, roots, shoots, symplast, plant, plant-composition major-genes modifiers, california, apoplast-
   **Abstract:** Previous research has shown that copper tolerance in Mimulus guttatus Fischer ex DC. is controlled by a single major gene and can be enhanced by a number of minor genes (or modifiers). Here we report the uptake of copper by three lines which all possessed the major tolerance gene but differed in the modifier genes: the major gene only (IT), the major gene plus increased (HT6) and decreased (LT6) modifiers. HT6 showed the highest copper tolerance and IT the lowest. Copper uptake was investigated at five copper concentrations and over 30 d to analyse the concentration of copper
accumulated in roots and shoots and the partitioning of apoplastic and symplastic root copper. Significant differences were found for root copper concentration with the IT line accumulating the highest levels. Fifty-two per cent of the root copper in the IT line is symplastic and this increases to 60% in LT6 and 64% in HT6. Significant differences were recorded for shoot copper concentration with HT6 accumulating the highest and IT the lowest. At the highest external copper concentration the HT line accumulated nearly 800 micrograms g⁻¹ in its shoot, approaching levels reported for copper hyperaccumulation.

4. **Deterrence of herbivory by zinc hyperaccumulation in Thlaspi caerulescens (Brassicaceae).**
   Pollard, A. J. and Baker, A. J. M.
   NAL Call #: 450-N42
   Descriptors: thlaspi, zinc, mineral-uptake mineral-excess herbivores, schistocerca-gregaria deroceras-panormitanum pieris-brassicae foliage, defense-mechanisms nutrient-solutions feeding-preferences leaves-

5. **Direct measurement of 59Fe-labeled Fe2+ influx in roots of pea using a chelator buffer system to control free Fe2+ in solution.**
   *Plant physiol* 111: 1 pp.93-100. (May 1996).
   NAL Call #: 450-P692
   Descriptors: pisum-sativum roots, iron, nutrient-uptake measurement, chelating-agents buffers, nutrient-transport translocation, mutants, genetic-variation ferrozine-buffered-uptake-solutions
   **Abstract:** Fe2+ transport in plants has been difficult to quantify because of the inability to control Fe2+ activity in aerated solutions and nonspecific binding of Fe to cell walls. In this study, a Fe(II)-3-(2-pyridyl)-5,6-diphenyl-1,2,4 tria zine-4’4’’-disulfonic acid buffer system was used to control free Fe2+ in uptake solutions. Additionally, desorption methodologies were developed to adequately remove nonspecifically bound Fe from the root apoplasm. This enabled us to quantify unidirectional Fe2+ influx via radiotracer (59Fe) uptake in roots of pea (Pisum sativum cv Sparkle) and its single gene mutant brz, an Fe hyperaccumulator. Fe influx into roots was dramatically inhibited by low temperature, indicating that the measured Fe accumulation in these roots was due to true influx across the plasma membrane rather than nonspecific binding to the root apoplasm. Both Fe2+ influx and Fe translocation to the shoots were stimulated by Fe deficiency in Sparkle. Additionally, brz, a mutant that constitutively exhibits high ferric reductase activity, exhibited higher Fe2+ influx rates than +Fe-grown Sparkle. These results suggest that either Fe deficiency triggers the induction of the Fe2+ transporter or that the enhanced ferric reductase activity somehow stimulates the activity of the existing Fe2+ transport protein.

6. **The ecological significance of nickel hyperaccumulation: a plant chemical defense.**
   Martens, S. N. and Boyd, R. S.
   NAL Call #: QL750.O3
   Descriptors: cruciferae, nickel, uptake, defense-mechanisms insect-pests herbivores,
The hepatoprotective cytochrome P-450 enzyme inhibitor isolated from the Nigerian medicinal plant Cochlospermum planchonii is a zinc salt.

Aliyu, R., Okoye, Z. S. C., and Shier, W. T.


Descriptors: medicinal-plants rhizomes, plant-extracts cytochrome-p-450 enzyme-inhibitors zinc, formates-salts isolation, liver, nigeria, hepatoprotective-agents

Abstract: Aqueous extracts of Cochlospermum planchonii Hook.f. (Cochlospermacae) rhizomes are used by native medical practitioners in northern Nigeria to treat jaundice. An extract prepared by a laboratory adaptation of their method was hepatoprotective in carbon tetrachloride-treated rats (CCl4), and it inhibited cytochrome P-450 enzymes, which constitutes a plausible hepatoprotective mechanism. A crystalline inhibitor (0.3% of dry weight of rhizomes) was isolated using inhibition of two rat cytochrome P-450 enzymes, aminopyrine-N-demethylase and aniline hydroxylase, as bioassays to guide fractionation by solvent partitioning, polyamide column chromatography, preparative thin layer chromatography and fractional crystallization. The inhibitor was identified as zinc formate by inductively coupled plasma atomic emission spectroscopy, nuclear magnetic resonance spectroscopy and comparison with synthetic material by powder X-ray diffraction crystallography. Synthetic and plant-derived zinc formate were equally effective as inhibitors of cytochrome P-450 enzymes and as hepatoprotective agents in carbon tetrachloride-treated rats. Cochlospermum planchonii rhizomes contain unusually high levels of manganese and zinc, although much higher levels have been observed in plants considered to be hyperaccumulators of these metals.
9. **Hyper-accumulation of astaxanthin in a green alga Haematococcus pluvialis at elevated temperatures.**


NAL Call #: QR53.B56

*Descriptors*: chlorophyta, astaxanthin, biosynthesis, algae-culture carotenogenesis-

*Abstract*: When a green alga Haematococcus pluvialis was cultivated at 30 degrees C, astaxanthin production was 3-fold more increased than at 20 degrees C. With acetate supplementation to 30 degrees C culture, the alga synthesized over 2-fold more carotenoid than without addition. Tiron, a radical scavenger, however, severely blocked the stimulated carotenogenesis, suggesting that endogenously generated active oxygen was responsible for the highly stimulated carotenogenesis. From these results, possible roles of the elevated cultivation temperatures for hyperaccumulation of astaxanthin were discussed.

10. **Hyperaccumulation, complexation and distribution of nickel in Sebertia acuminata.**


NAL Call #: 450-P5622

*Descriptors*: sapotaceae, nickel, detoxification, plant-composition plant-tissues citric-acid nuclear-magnetic-resonance ion-balance nitrate, sap, insect-repellers drosophila-melanogaster mineral-uptake latex, phloem, xylem, iron, zinc, copper, latex-tubes plant-glands lactifers-

*Abstract*: The nickel content in different parts of the hyperaccumulating tree Sebertia acuminata was analysed by atomic absorption spectroscopy. Nickel was found to be mainly located in laticifers. The total nickel content of a single mature tree was estimated to be 37 kg. By gel filtration and NMR spectroscopy, citric acid was unequivocally identified as counter ion for about 40% of this metal present. Nitrate was assumed to be a further partner for a complete ionic balance. Phytochelatins were not found to be involved in nickel detoxification in Sebertia. The localization of nickel complexes inside the laticifers was demonstrated by light microscopy as well as by scanning electron microscopy in combination with an EDX system for the analysis of elements. A repellent effect of the plant sap was observed on the fruit fly Drosophila melanogaster indicating that in hyperaccumulating plants nickel functions as an agent to prevent predation.

11. **Hyperaccumulation of nickel by Arenaria rubella (Caryophyllaceae) from Washington State.**


NAL Call #: 450-M26

*Descriptors*: arenaria-caryophyllaceae species, plant-composition nickel, concentration, uptake, washington-

12. **Lead phytoextraction: species variation in lead uptake and translocation.**


NAL Call #: 450-N42 *Descriptors*: metal-ions. polluted-soils. bioremediation-. zea-mays. ambrosia-artemisiifolia. triticum-aestivum. thlaspi-. lead-. translocation-. nutrient-
13. **Negative control of heavy metal uptake by the Saccharomyces cerevisiae BSD2 gene.**
Liu, X. F., Supek, F., Nelson, N., and Culotta, V. C.
NAL Call #: 381-J824
Descriptors: saccharomyces-cerevisiae polypeptides, structural-genes copper, cadmium, heavy-metals metal-ions ion-uptake ion-transport endoplasmic-reticulum mutants-
Abstract: We have previously shown that mutations in the Saccharomyces cerevisiae BSD2 gene suppress oxidative damage in cells lacking superoxide dismutase and also lead to hyperaccumulation of copper ions. We demonstrate here that bsd2 mutant cells additionally accumulate high levels of cadmium and cobalt. By biochemical fractionation and immunofluorescence microscopy, BSD2 exhibited localization to the endoplasmic reticulum, suggesting that BSD2 acts at a distance to inhibit metal uptake from the growth medium. This BSD2 control of ion transport occurs independently of the CTR1 and FET4 metal transport systems. Genetic suppressor analysis revealed that hyperaccumulation of copper and cadmium in bsd2 mutants is mediated through SMF1, previously shown to encode a plasma membrane transporter for manganese. A nonsense mutation removing the carboxyl-terminal hydrophobic domain of SMF1 was found to mimic a smf1 gene deletion by eliminating the copper and cadmium toxicity of bsd2 mutants and also by precluding the bsd2 suppression of superoxide dismutase deficiency. However, inactivation of SMF1 did not eliminate the elevated cobalt levels in bsd2 mutants. Instead, this cobalt accumulation was found to be specifically mediated through the SMF1 homologue, SMF2. Hence, BSD2 prevents metal hyperaccumulation by exerting negative control over the SMF1 and SMF2 metal transport systems.

14. **Nickel accumulation by serpentine species of Streptanthus (Brassicaceae): field and greenhouse studies.**
Kruckeberg, A. R. and Reeves, R. D.
NAL Call #: 450-M26
Descriptors: cruciferae, nickel, uptake, concentration, plant-tissues serpentine-soils hyperaccumulation-

15. **Nickel hyperaccumulation defends Streptanthus polygaloides (Brassicaceae) against pathogens.**
Boyd, R. S., Shaw, J. J., and Martens, S. N.
NAL Call #: 450-Am36
Descriptors: cruciferae, nickel, plant-composition disease-resistance erysiphe-polygoni xanthomonas-campestris-pv -campestris alternaria-brassicicola pathogenicity, infectivity- 
Abstract: The Ni-hyperaccumulating annual, Streptanthus polygaloides, may contain as much as 16,400 ppm Ni (dry weight) in its tissues. The function of Ni hyperaccumulation is not known. We tested the hypothesis that one function of Ni hyperac cumulation in S. polygaloides is defense against pathogens. Growth of pathogenic organisms on Ni-hyperaccumulating plants (averaging 5,630 ppm Ni, produced by growing plants on high-Ni soil) was compared to pathogen growth on nonhyperaccumulating plants ( averaging 124 ppm Ni, produced by growing plants on low-Ni soil). Plants containing
hyperaccumulated Ni were more slowly infected by a powdery mildew (Erysiphe polygoni) than low-Ni plants. Two strains of the bacterial pathogen Xanthomonas campestris pv. campestris (one a genetically engineered bioluminescent strain) grew in low-Ni plants but not high-Ni plants. Growth of X. campestris pv. campestris was markedly inhibited by Ni concentrations of 400 ppm in artificial media. Growth of the fungus Alternaria brassicicola, which was necrotrophic on S. polygaloides, also was inhibited on high-Ni leaves relative to low-Ni leaves. These results demonstrated negative effects of hyperaccumulated Ni on a taxonomically wide range of pathogenic organisms, supporting the hypothesis that Ni hyperaccumulation defends S. polygaloides against plant pathogens.

16. **Nickel-resistant bacteria from anthropogenically nickel-polluted and naturally nickel-percolated ecosystems.**

Stoppel, R. D. and Schlegel, H. G.


NAL Call #: 448.3-Ap5

*Descriptors*: soil-bacteria bacteria, isolation, waste-water-treatment industrial-wastes nickel, metal-tolerance dna-probes dna-hybridization identification, soil-chemistry plasmids, nickel, chlorides, new-caledonia zaire, europe, usa, nickel-containing-soils nickel-chloride

*Abstract*: DNA fragments harboring the nickel resistance determinants from bacteria isolated from anthropogenically polluted ecosystems in Europe and Zaire were compared with those harboring the nickel resistance determinants from bacteria isolated from naturally nickel-percolated soils from New Caledonia by DNA-DNA hybridization. The biotinylated DNA probes were derived from the previously described Alcaligenes eutrophus CH34, Alcaligenes xylosoxidans 31A, Alcaligenes denitrificans 4a-2, and Klebsiella oxytoca CCUG 15788 and four new nickel resistance-determining fragments cloned from strains isolated from soils under nickel-hyperaccumulating trees. Nine probes were hybridized with endonuclease-cleaved plasmid and total DNA samples from 56 nickel-resistant strains. Some of the New Caledonian strains were tentatively identified as Acinetobacter, Pseudomonas mendocina, Comamonas, Hafnia alvei, Burkholderia, Arthrobacter aurescens, and Arthrobacter ramosus strains. The DNA of most strains showed homologies to one or several of the following nickel resistance determinants: the cnr and ncc operons of the strains A. eutrophus CH34 and A. xylosoxidans 31A, respectively, the nre operon of strain 31A, and the nickel resistance determinants of K. oxytoca. On the basis of their hybridization reactions the nickel resistance determinants of the strains could be assigned to four groups: (i) cnr/ncc type, (ii) cnr/ncc/nre type, (iii) K. oxytoca type, and (iv) others. The majority of the strains were assigned to the known groups. Among the strains from Belgium and Zaire, exclusively the cnr/ncc and the cnr/ncc/nre types were found. Among the New Caledonian strains all four types were represented. Homologies to the nre operon were found only in combination with the cnr/ncc operon. The homologies to the cnr/ncc operon were the most abundant and were detected alone or together with homologies to the nre operon. Only the DNA of the strains isolated from soil in Scotland and the United States and that of five of the New Caledonian strains did not show any detectable homologies to any of our probes. The nickel resistance fragment isolated from Burkholderia strain 32W-2 was studied in some detail. This 15-kb BamHI fragment
conferred resistance to 1 to 5 mM NiCl₂ to Escherichia coli and resistance to up to 25 mM NiCl₂ to A. eutrophus. It showed strong homologies to both the cnr/ncc operon and the nre operon and conferred strictly regulated (inducible) nickel resistance to A. eutrophus.

17. **Physiological characterization of root Zn²⁺ absorption and translocation to shoots in Zn hyperaccumulator and nonaccumulator species of Thlaspi.**

NAL Call #: 450-P692

Descriptors: thlaspi, thlaspi-arvense zinc, metal-tolerance ion-uptake roots, absorption, plasma-membranes translocation, symplast, saturation, shoots, transport-processes kinetics, nutrient-availability thlaspi-caerulescens

Abstract: Radiotracer techniques were employed to characterize 65Zn²⁺ influx into the root symplasm and translocation to the shoot in Thlaspi caerulescens, a Zn hyperaccumulator, and Thlaspi arvense, a nonaccumulator. A protocol was developed that allowed us to quantify unidirectional 65Zn²⁺ influx across the root-cell plasma membrane (20 min of radioactive uptake followed by 15 min of desorption in a 100 micromolar ZnCl₂ + 5 mM CaCl₂ solution). Concentration-dependent Zn²⁺ influx in both Thlaspi species yielded nonsaturating kinetic curves that could be resolved into linear and saturable components. The linear kinetic component was shown to be cell-wall-bound Zn²⁺ remaining in the root after desorption, and the saturable component was due to Zn²⁺ influx across the root-cell plasma membrane. This saturable component followed Michaelis-Menten kinetics, with similar apparent Michaelis constant values for T. caerulescens and T. arvense (8 and 6 micromolar, respectively). However, the maximum initial velocity for Zn²⁺ influx in T. caerulescens root cells was 4.5-fold higher than for T. arvense, indicating that enhanced absorption into the root is one of the mechanisms involved in Zn hyperaccumulation. After 96 h 10-fold more 65Zn was translocated to the shoot of T. caerulescens compared with T. arvense. This indicates that transport sites other than entry into the root symplasm are also stimulated in T. caerulescens. We suggest that although increased root Zn²⁺ influx is a significant component, transport across the plasma membrane and tonoplast of leaf cells must also be critical sites for Zn hyperaccumulation in T. caerulescens.

18. **Quantitative genetics of zinc hyperaccumulation in Thlaspi caerulescens.**

Pollard, A. J. and Baker, A. J. M.
NAL Call #: 450-N42

Descriptors: thlaspi, zinc, plant-composition shoots, leaves, concentration, size, plant-communities individual-characteristics heritability, genetic-variation quantitative-genetics metal-tolerance polluted-soils bioremediation, england, sib-families

19. **Responses of six crop species to solution zinc²⁺ activities buffered with HEDTA.**

Parker, D. R.
NAL Call #: 56.9-So3

Descriptors: zea-mays triticum-aestivum elymus-elongatus medicago-sativa lycopersicon-esculentum glycine-max seedlings, zinc, mineral-deficiencies ion-activity
Abstract: Zinc contamination has long hampered the use of conventional nutrient solutions for attaining reproducible and severe Zn deficiencies. Chelator-buffered solutions show promise for precisely imposing micronutrient deficiencies, but further studies are needed to fully evaluate the method and to develop methods to avoid Zn-deficiency-induced P toxicity. In a greenhouse study, seedlings of maize (Zea mays L. 'Golden Cross Bantam'), wheat (Triticum aestivum L. 'Yecora Rojo'), tall wheatgrass (Elytrigia pontica [Podp.] Holub 'Orbit'), alfalfa (Medicago sativa L. 'Germain's WL-320'), soybean (Glycine max [L.] Merr. 'Vinton 81'), and tomato (Lycopersicon esculentum L. 'Jackpot') were grown for 16 to 29 d in solutions containing 0.4 to 12 micromolar total Zn. Computed Zn2+ activities were buffered at 4 to 123 pmol L-1 by a 50 micromolar excess of HEDTA, and solution P was maintained at 10 +/-, 3 micromolar. Zinc deficiencies ranged from mild to severe, dry matter yield minima ranged from 32 to 89% of controls, and critical Zn2+ activities for the onset of deficiency ranged from approximately 10 to 65 pmol L-1. The relative sensitivity of the six species to low Zn was maize > tomato > wheat > alfalfa approximately equal to tall wheatgrass > soybean. Shoot Zn concentrations were consistent with the widely reported critical range of 15 to 20 mg kg-1. Zinc deficiency in the three grass species led to hyperaccumulation of P to phytotoxic levels, and leaf symptoms largely reflected this toxicity. Leaf P concentrations in the three dicots tended to be lower, but may have been within the phytotoxic range. Thus, even with P concentrations that approach soil solution values, the P toxicity that often accompanies Zn deficiency in solution-culture experiments was not eliminated. The effects of Zn deficiency on shoot concentrations of nutrients other than P were variable, although divalent cations tended to be elevated in Zn-deficient shoots.

20. Scientist using plants to clean up metals in contaminated soil.
Bernstein, E.
NAL Call #: 286.8-N488
Descriptors: apocynum, ambrosia, genetic-engineering waste-treatment heavy-metals hyperaccumulators, bioremediation-

21. Shoot-to-root signal transmission regulates root Fe(III) reductase activity in the dgl mutant of pea.
Grusak, M. A. and Pezeshgi, S.
NAL Call #: 450-P692
Descriptors: pisum-sativum shoots, roots, iron, reduction, proton-pump ion-transport signals, ferroxidase, enzyme-activity developmental-stages mutants, genetic-variation cultivars, genotypes-
Abstract: To understand the root, shoot, and Fe-nutritional factors that regulate root Fe-acquisition processes in dicotyledonous plants, Fe(III) reduction and net proton efflux were quantified in root systems of an Fe-hyperaccumulating mutant (dgl) and a parental (cv Dippes Gelbe Viktoria [DGIV]) genotype of pea (Pisum sativum). Plants were grown with (+Fe treated) or without (-Fe treated) added Fe(III)-N,N'-ethylenebis[2-(2-hydroxyphenyl) -glycine] (2 micromolar); root Fe(III) reduction was measured in solutions containing growth nutrients, 0.1 mM Fe(III)-ethylenediaminetetraacetic acid,
and 0.1 mM Na2-bathophenanthrolinedisulfonic acid. Daily measurements of Fe(III) reduction (d 10-20) revealed initially low rates in +Fe-treated and -Fe-treated dgl, followed by a nearly 5-fold stimulation in rates by d 15 for both growth types. In DGV, root Fe(III) reductase activity increased only minimally by d 20 in +Fe-treated plants and about 3-fold in -Fe-treated plants, beginning on d 15. Net proton e fflux was enhanced in roots of -Fe-treated DGV and both dgl growth types, relative to +Fe-treated DGV. In dgl, the enhanced proton efflux occurred prior to the increase in root Fe(III) reductase activity. Reductase studies using plants with reciprocal shoot:root grafts demonstrated that shoot expression of the dgl gene leads to the generation of a transmissible signal that enhances Fe(III) reductase activity in roots. The dgl gene product may alter or interfere with a normal component of a signal transduction mechanism regulating Fe homeostasis in plants.

22. Uptake and transport of zinc in the hyperaccumulator Thlaspi caerulescens and the non-hyperaccumulator Thlaspi ochroleucum.

Shen, Z. G., Zhao, F. J., and McGrath, S. P.


NAL Call #: QK710.P55

Descriptors: thlaspi, zinc, metal-ions ion-uptake species-differences translocation, metal-tolerance plant-composition roots, shoots, malic-acid phytotoxicity, citric-acid chlorosis-

Abstract: Growth and zinc uptake of the hyperaccumulator species Thlaspi caerulescens J. & C. Presl and the non-hyperaccumulator species Thlaspi ochroleucum Boiss. & Heldr. were compared in solution culture experiments. T. caerulescens was was able to tolerate 500 mmol m-3 (32.5 g m-3) Zn in solution without growth reduction, and up to 1000 mmol m-3 (65 g m-3) Zn without showing visible toxic symptoms but with a 25% decrease in dry matter (DM) yield. Up to 28 g kg-1 of Zn in shoot DM w as obtained in healthy plants of T. caerulescens. In contrast, T. ochroleucum suffered severe phytotoxicity at 500 mmol m-3 Zn. Marked differences were shown in Zn uptake, distribution and re-distribution between the two species. T. caerulescens had much higher concentrations of Zn in the shoots, whereas T. ochroleucum accumulated higher concentrations of Zn in the roots. When an external supply of 500 mmol m-3 Zn was withheld, 89% of the Zn accumulated previously in the roots of T. caerulescens was trans transported to the shoots over a 33 d period, whereas in T. ochroleucum only 32% was transported. T. caerulescens was shown to have a greater internal requirement for Zn than other plants. Increasing the supply of Zn from 1 to 10 mmol m-3 gave a 19% increase i n the total DM of this species. Even the shoots from the 1 mmol m-3 Zn treatment which showed Zn deficiency contained 10 times greater Zn concentrations than the widely reported critical value for Zn deficiency to occur in many other plant species. The results obtained suggest that strongly expressed constitutive sequestration mechanisms exist in the hyperaccumulator T. caerulescens, which detoxify the large amount of Zn present in shoot tissues and decrease its physiological availability in the cytosol. Both T. caerulescens and T. ochroleucum had constitutively high concentrations of malate in shoots, which were little affected by different Zn treatments. Although malate may play a role in Zn chelation because of the high concentrations present, it can not explain the species specificity of Zn tolerance and hyperaccumulation.

23. Use of elemental sulphur to enhance a cadmium solubilization and its vegetative removal from contaminated soil.
24. **Use of wild type and nickel resistant Neurospora crassa for removal of Ni2+ from aqueous medium.**

Shravan Kumar, C., Sivarama Sastry, K., and Maruthi Mohan, P.

**Descriptors:** neurospora-crassa nickel, metal-ions resistance, mycelium, ion-uptake culture-media pollution

**Abstract:** A novel nickel resistant, hyperaccumulating N. crassa nir-2 mutant, isolated by us, sequestered 90% of Ni2+ from medium with 120 mg/l Ni2+. The parent wild strain showed comparable efficiency only at much lower concentrations (< 10 mg/l). The initial rapid rate and efficiency of Ni2+ removal could be maintained beyond 2 h by fresh addition of mycelial biomass. The results have been discussed from the standpoint of the utility of metal resistant fungi in the control of environmental pollution.

25. **Zinc hyperaccumulation in Thlaspi caerulescens. II. Influence on organic acids.**

Tolra, R. P., Poschenrieder, C., and Barcelo, J.

**Descriptors:** thlaspi, zinc, mineral-uptake nutrient-uptake nutrient-availability malic-acid citric-acid succinic-acid oxalic-acid biosynthesis, plant-composition shoots, roots, metal-tolerance

**Abstract:** The influence of different zinc (Zn) concentrations (1.5 to 1500 micromolar) on organic acid levels in roots and shoots of the Zn-hyperaccumulator plant Thlaspi caerulescens was investigated. In shoots, malate was the most abundant organic acid (164 to 248 micromol/g f.w.), followed by citrate, succinate, and oxalate. A significant correlation between soluble Zn and both malate and oxalate was observed in shoots, but not in roots. In shoots, a significant correlation between inorganic cation equivalents and organic acid anion equivalents was found. These observations and the finding, that organic acid concentrations were high even under suboptimal Zn supply (1.5 micromolar)
suggest that in *T. caerulescens* the high organic acid concentration in shoots is a constitutive property. The variation of the organic acid concentrations seem to be a consequence of the cation-anion balance rather than a specific Zn tolerance mechanism. The constitutively high organic acid concentration may be responsible for the high Zn and iron (Fe) tissue concentrations required for optimum growth in *T. caerulescens*. 

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