

Tu, Q., X.Q. Shan, J. Qian, and Z.M. Ni. 1994. Trace metal redistribution during extraction of model soils by acetic acid/sodium acetate. *Anal. Chem.* 66:3562–3598.

Ure, A., P.H. Quevauviller, H. Muntau, and B. Griepink. 1993. Speciation of heavy metals in soils and sediments, an account of the improvement and harmonisation of extraction techniques under-

taken under the auspices of the BCR of the commission of the European communities. *Int. J. Environ. Anal. Chem.* 51:135–151.

Woolson, E.A., J.H. Axley, and P.C. Kearney. 1971. The chemistry and phytotoxicity of arsenic in soils: I. Contaminated field soils. *Soil Sci. Soc. Am. Proc.* 35:938–943.

Amelioration of Nickel Phytotoxicity in Muck and Mineral Soils

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ABSTRACT

In situ remediation (phytostabilization) is a cost-effective solution for restoring the productivity of metal-contaminated soils and protection of food chains. A pot experiment with wheat (*Triticum aestivum* L.), oat (*Avena sativa* L.), and redbeet (*Beta vulgaris* L.) was conducted to test the ability of limestone and hydrous ferric oxide (HFO) to ameliorate Ni phytotoxicity in two soils contaminated by particulate emissions from a nickel refinery. Quarry muck (Terric Haplohemist; 72% organic matter) contained 2210 mg kg⁻¹ of total Ni. The mineral soil, Welland silt loam (Typic Epiacquoll), was more contaminated (2930 mg Ni kg⁻¹). Both soils were very strongly acidic, allowing the soil Ni to be soluble and phytotoxic. Nickel phytotoxicity of the untreated muck soil was not very pronounced and could be easily confused with symptoms of Mn deficiency that occurred in this soil even with Mn fertilization. Severe nickel phytotoxicity of the untreated mineral soil prevented any growth of redbeet, the most sensitive crop; even wheat, a relatively Ni-resistant species, was severely damaged. White banding indicative of Ni phytotoxicity was present on oat and wheat leaves grown on the acidic mineral soil. Soil Ni extracted with diethylenetriaminepentaacetic acid (DTPA) and 0.01 M Sr(NO₃)₂ was indicative of the ameliorative effect of amendments and correlated well with Ni concentrations in plant shoots. Making soils calcareous was an effective treatment to reduce plant-available Ni and remediate Ni phytotoxicity of these soils to all crops tested. The ameliorative effect of HFO was crop-specific and much less pronounced.

THE long-term deposition of Ni-bearing particulate emissions originating from a Ni refinery located in Port Colborne, southern Ontario, Canada resulted in contamination of soils and vegetation in the vicinity northeast (downwind) of the refinery. Nickel concentrations exceeding 10 000 mg kg⁻¹ in the 0- to 5-cm soil layer were reported but this extremely high level of contamination is confined to a very limited area (Temple and Bisessar, 1981). Much attention has been devoted to Ni contamination of muck soil farms in the vicinity of the refinery (Temple and Bisessar, 1981; Frank et al., 1982; Bisessar, 1989). Vegetable production, the primary use of the muck soil, was adversely affected by the emissions from the refinery. Toxicity symptoms occurring in various vegetable crops as well as losses of marketable yield were investigated by Frank et al. (1982). Marketable yield of radish (*Raphanus sativus* L.) in muck soil containing 4800 mg kg⁻¹ of total Ni was reduced by 93.2%. No marketable yield of cabbage

(*Brassica oleracea* L. var. *capitata* L.) and beetroot (*Beta vulgaris* L.) was obtained at 2075 and 4505 mg kg⁻¹ Ni in soil, respectively. On the muck soil containing 1200 mg kg⁻¹ Ni, the predicted marketable yield of celery [*Apium graveolens* L. var. *dulce* (Mill.) Pers.] was reduced by 16 to 40% depending on growing season. Yield of lettuce (*Lactuca sativa* L.) grown in soil with 1300 mg kg⁻¹ Ni was increased by 8% or reduced by 36% in comparison with predicted yields in various growing seasons. Nickel toxicity to agricultural crops grown in contaminated mineral soils located in the vicinity of the refinery as well as potential for remediation of these soils received very little attention.

In situ amelioration of heavy metal toxicity, a cost-effective alternative to the replacement of contaminated soil, may be achieved by amending soils with components that reduce metal solubility and hence phytoavailability. Depending on metal, soil, and resources available, a variety of amendments can be used including clay minerals, apatite, ferric and manganese hydroxy oxides, and limestone (Brown and Chaney, 2000; Mench et al., 1994; Chlopecka and Adriano, 1996). According to King (1988), soil pH, organic matter, and Fe oxides content were the most important factors controlling Ni sorption by soils. Among them, soil pH was the primary factor controlling Ni sorption, hence governing Ni solubility. Limestone has been successfully used for full or partial remediation of Ni phytotoxicity in serpentine soils rich in Ni of geogenic origin (Hunter and Vergnano, 1952; Crooke, 1956). An earlier study (Chaney and Kukier, 1998) demonstrated that Ni phytotoxicity to oat and redbeet in Quarry muck containing 3000 mg kg⁻¹ of total Ni was ameliorated by the application of a high rate of limestone. Amendment of ferric hydrous oxide has also shown some beneficial effect. However, the amendments that reduced Ni phytotoxicity induced severe Mn deficiency and prevented full remediation of the contaminated Lake Plain soils that lost Mn during genesis (Baldwin and Johnston, 1986).

Theoretically, if soil Ni is highly enriched by industrial contamination, the goal of remediation treatments should be to reverse Ni phytotoxicity in a persistent manner. If such soils were amended with limestone to correct phytotoxicity, and limestone were not regularly applied to correct acidity potential generated from applied N and P fertilizers and natural processes, soil pH

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Abbreviations: DTPA, diethylenetriaminepentaacetic acid; HFO, hydrous ferric oxide; limestone, a mixture of reagent-grade Ca and Mg carbonates (4.8:1, w/w).

would slowly fall and allow the sorbed Ni to be solubilized and again induce Ni phytotoxicity. A justifiable alternative to soil removal and replacement is to make the soil calcareous to inactivate soil Ni by application of a very high rate of limestone so that future pH decline is very unlikely in terms of centuries. Buffering soil pH at a high level with limestone is the single most effective amelioration treatment to convert soluble Ni into sorbed or occluded forms (nonphytotoxic) in the treated soil.

A further reduction of soluble Ni in soil solution may be achieved by addition of hydrous ferric oxide (HFO), which can increase Ni adsorption and enhance occlusion of Ni over time. Nickel sorption on laboratory-prepared Fe oxides has been shown to increase greatly with increase of pH (Bryce et al., 1994; Lo et al., 1994). Backes et al. (1995) demonstrated that metal sorption on HFO is affected by the specific surface area of the oxide, and is greater on amorphous than on crystalline minerals. Further, the crystalline Fe oxide, goethite, was found to occlude Ni over time in a diffusion-limited process (Bruemmer et al., 1988). Occlusion could be very significant over time as illustrated by the low fraction of DTPA-extractable Ni in serpentine soils that are simultaneously high in Fe oxides (L'Huillier and Edighofer, 1996).

The objective of the present experiment was to test the effectiveness of limestone and HFO, applied alone or in combination, in amelioration of Ni phytotoxicity in two soils widely differing in their properties. High rates of Mn fertilizer were supplied to prevent limestone and HFO from inducing Mn deficiency. Soil Ni extractability tests in relation to plant performance were evaluated as a tool for assessment of remediation effectiveness.

MATERIALS AND METHODS

Soil Collection

Remediation of an organic Quarry muck soil (Terric Haplohemist; Canadian classification, Orthic Humic Gleysol; 72% organic matter) and a mineral Welland silt loam soil (Typic Epiaquoll; Canadian classification, Terric Mesisol), both contaminated by particulate emissions from the Port Colborne, Ontario, nickel refinery, was examined in a greenhouse pot study. Soils were collected from the plow layer of previously cropped fields. The muck soil was collected at the site of Ni phytotoxicity evaluation studies conducted by Frank et al. (1982) and related studies by the Ontario Ministry of the Environment in the 1970s. Total soil Ni was determined according to the USEPA Method 3050 using boiling HNO_3 (USEPA, 1995). Organic matter content was determined as the loss on ignition by ashing the soil samples, previously dried at 105°C, in a muffle furnace at 450°C for 16 h. Soil pH was

measured in a 1:2 soil and water (by volume) slurry after 1 h of equilibration. Particle size distribution was determined by hydrometer method after removal of organic matter (Gee and Bauder, 1986). The properties of the soils used are presented in Table 1. Quarry and Welland are the predominant soil series contaminated by the Ni refinery. The total Ni levels in Quarry and Welland soils collected for our study were 2210 and 2930 mg kg^{-1} , respectively. This represents rather high extent of contamination compared with the wider area with recognized contamination (Kuja et al., 2001).

Soils for the pot experiment were dried enough to allow them to be sieved through a 5-mm stainless steel sieve, homogenized, and stored moist in closed plastic containers at 4°C until amended with limestone and/or HFO and fertilizers. Maintaining the soil in a nondried condition allows preservation of microbial activity and minimizes redox reactions of Fe, Mn, and other elements that occur during soil drying (Bartlett and James, 1980). The moisture content of the soils was measured by oven-drying and was taken into account at treatment and fertilizer applications. All amendment rates are expressed on an oven-dry soil basis. Experiments were conducted in freely drained 1.5-L plastic pots holding 450 g of the oven-dry muck soil (510 g of air-dry) and 1070 g of the oven-dry (1120 g of air-dry) mineral soil; saucers were used to prevent loss of leachate, and plastic mesh was used to line the drainage hole to prevent soil loss from the pots.

Soil Amendments

A mixture of powdered reagent-grade amorphous CaCO_3 and MgCO_3 at 4.8:1 on a mass basis (hereafter referred to as "limestone") and hydrous ferric oxide (HFO) were used as amendments to counteract soil Ni phytotoxicity. Limestone was applied at 0 or 50 Mg ha^{-1} on the basis of the pot surface area (0 vs. 12.7% of oven-dry weight muck soil and 0 vs. 5.3% of oven-dry mineral soil). The combination of Ca and Mg carbonates was used because making a soil calcareous with only CaCO_3 may cause subsequent development of Mg deficiency. Under field conditions, dolomitic limestone should be used if one plans to make an acidic soil calcareous. Amorphous forms of pure chemicals were used to promote very rapid (<3 d) neutralization of soil acidity compared with commercial limestone, which may require more than 1 yr to reach equilibrium pH. Hydrous ferric oxide [freshly precipitated $\text{Fe}(\text{OH})_3$] rates were 0 and 10 Mg Fe ha^{-1} (2.53% Fe in the low-bulk density muck soil and 1.06% in the mineral soil). Treatments were combined in a factorial complete randomized block design with four replicates.

Hydrous ferric oxide was precipitated at room temperature by addition of NaOH to a solution of $\text{Fe}(\text{NO}_3)_3$ in a quantity exceeding the stoichiometric ratio [$\text{Fe}(\text{NO}_3)_3$ to NaOH molar ratio of 1:3] by 3%. Upon precipitation, the HFO was washed with several portions of deionized water to remove NaNO_3 . After leaching, electrical conductivity of the entrained solution was in the range of 0.05 to 0.06 mS cm^{-1} and the pH of the suspension was about 7.3.

Table 1. Properties of soils used in the pot study.

Soil	Total Ni mg kg^{-1}	pH in water, 1:2 (v/v)	Sand %	Clay %	Bulk density† kg dm^{-3}	Organic matter %
Quarry muck	2210	5.66	nd‡	nd	0.33	72
Welland loam	2930	5.24	14	31	0.72	17

† Bulk density of soil sieved through a 5-mm sieve and placed into a pot, dry-weight basis.

‡ Not determined.

Pot Experiment

Basal fertilizers (reagent-grade chemicals) were added to all pots as salt solutions at the following rates (expressed in milligrams per pot because of the differences in bulk density between the soils): 200 mg P as KH_2PO_4 and CaHPO_4 (added as a solid); 170 mg K as KH_2PO_4 ; 80 mg N as $\text{Ca}(\text{NO}_3)_2 \cdot 4\text{H}_2\text{O}$; 68 mg Mg as $\text{MgSO}_4 \cdot 7\text{H}_2\text{O}$; 0.90 mg Zn as $\text{ZnSO}_4 \cdot 7\text{H}_2\text{O}$; 0.45 mg Cu as $\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$; and 0.34 mg B as H_3BO_3 . Lake Plain soils are susceptible to Mn deficiency (Baldwin and Johnston, 1986). Both HFO and soil alkalization increase sorption of metals in soil including Mn; therefore, a higher Mn rate was applied to pots amended with limestone and/or HFO than to control pots. Our previous pot study (Kukier and Chaney, 2000) with muck soil showed that a very high rate of P fertilizer was also needed to avoid P deficiency caused by HFO and limestone application, although higher rates are required in pot experiments than in the field because of shorter root length in pots. The Mn fertilizer was added as $\text{MnSO}_4 \cdot \text{H}_2\text{O}$ at the rate of 22.6 mg per pot to control pots and at 113 mg per pot to the pots filled with soils treated with HFO, limestone, or limestone + HFO. These rates correspond to 20 and 100 kg ha^{-1} of Mn. Manganese sulfate was used because it was superior to MnO as a source of Mn to correct lime-induced Mn deficiency of soils amended with alkaline biosolids (Brown et al., 1997). After mixing with fertilizers and amendments, moist soil in the pots was incubated for 1 d prior to planting seeds.

'Grandin' hard red spring wheat, 'Ogle' oat, and 'Detroit Dark Red' redbeet were grown to test plant responses to soils and treatments. These species represent various degrees of sensitivity to Ni phytotoxicity. Wheat is relatively resistant to high soil Ni (Hunter and Vergnano, 1952), while oat is a sensitive species that exhibits very characteristic symptoms of Ni toxicity compared with other element phytotoxicities (Hunter and Vergnano, 1953). Redbeet, a very sensitive species to excessive soil Ni, was also grown because it was not a *Poaceae* species. The number of seeds planted per pot was 15 for all species. Polygerm seeds were used for redbeet. After emergence, plants were thinned to 10 per pot. The experiment was conducted in a greenhouse equipped with high-intensity sodium and incandescent lights capable of supplying $400 \mu\text{mol m}^{-2} \text{s}^{-1}$ of photosynthetically active radiation. Day length was set at 16 h. The temperature was 27°C during the day and 20°C during the night. Plants were harvested after 39 d of growth.

Soil and Plant Analysis

At harvest, plant shoots were cut about 1 cm above the soil surface. The lower parts of the oat and wheat shoots and the whole red beet shoots were rinsed in deionized water to remove adhering soil particles. Plants were oven-dried at 65° to a constant weight, ground in a stainless steel Wiley mill, weighed into Pyrex beakers and ashed in a muffle furnace at 450° for 16 h. Blanks were included every 10 samples. Ashed plant samples were digested with 2 mL of concentrated HNO_3 on a hot plate and then refluxed for 2 h with 10 mL of 3 mol L^{-1} HCl. Digested samples were filtered and diluted to 25 mL. Samples were analyzed for Ca, Mg, K, P, Mn, Fe, Zn, and Cu by inductively coupled plasma atomic emission spectrometry (ICP) using 40 mg L^{-1} Y as an internal standard. Nickel was analyzed by flame atomic absorption spectrometry with background correction. National Institute of Standards and Technology (NIST) Standard Reference Material 1573a (tomato leaves) was digested and analyzed (one per 20 samples) for quality control; results were within the listed deviation for the elements reported by a standard manufacturer.

After plant harvest, the soil in each pot was mixed and

representative soil samples were collected. All further measurements and analyses were performed on these air-dried soil samples. The pH was measured in water slurries at a soil to deionized water ratio of 1:2 (by volume) and 1 h equilibration time. The DTPA-extractable Ni, Mn, and Fe were determined using the method of Lindsay and Norvell (1978) but the soil to solution ratio was 1 g:30 mL. The original method developed for micronutrient deficiency diagnosis specifies a ratio of 10 g:20 mL, which assures that an excess of the chelating agent remains in the solution after all complexation reactions are completed. Our earlier studies demonstrated that a much lower ratio of soil to solution is necessary for metal-contaminated soils in order to avoid saturation of DTPA by the large pool of phytoavailable metals. Strontium-extractable Ni, Mn, Mg, and Ca were determined using $0.01 \text{ M Sr}(\text{NO}_3)_2$ at a soil to solution ratio of 1:4 (10 g:40 mL) and 2 h shaking time before filtering. It was a modification of the method described by Madden (1988). In both cases, pH of extracts was measured. Concentrations of Ni, Mn, and Fe in the extracts were determined by atomic absorption spectrometry (AAS) with deuterium background correction when necessary. Calcium and Mg were analyzed by ICP.

Statistical Analysis

The GLM procedure (SAS Institute, 1988) was used to test statistical significance of the treatment effects on plant yield and elemental composition as well as extractable metals in soils. The procedure was first applied to the pooled species data and then separately for each plant species and soil after species was found to significantly affect yield and elemental composition of the plant samples in response to the treatments. The Duncan multiple range test was used for separation of treatment means.

RESULTS AND DISCUSSION

Effect of Treatments on Soil pH and Nickel Extractability

Soil pH was modified mainly by application of the limestone. Hydrous ferric oxide addition resulted in a small but significant increase in pH (Table 2). Plant species did not have a significant effect on pH; therefore, only the average pH for each treatment and soil is presented.

The advantage of using $\text{Sr}(\text{NO}_3)_2$ for determination of plant-available Ni is that soil pH remains almost unaltered during extraction, and the exchange reactions occur under the pH conditions representative of those in the bulk soil in the pots. On average, pH of the mineral soil $\text{Sr}(\text{NO}_3)_2$ extracts was only 0.086 pH unit lower than one measured in a water slurry. In the muck soil, the average pH decrease in $\text{Sr}(\text{NO}_3)_2$ extracts as compared with water was 0.19 pH unit.

The control mineral soil contained about 22 times

Table 2. Soil pH after plant harvest. Soil to water ratio 1:2 (v/v), equilibration time 2 h. HFO = hydrous ferric oxide.

Treatment	Muck soil	Mineral soil
Control	5.85a†	5.11a
HFO	6.11b	5.23b
Limestone	7.36c	7.47c
Limestone + HFO	7.61d	7.53d

† Means within a column followed by the same letter are not significantly different according to Duncan's multiple range test ($P < 0.05$).

Table 3. The effect of amendments on diethylenetriaminepentaacetic acid (DTPA)- and 0.01 M Sr(NO₃)₂-extractable elements in the muck and mineral soil. HFO = hydrous ferric oxide.

Treatment	DTPA-extractable			Sr(NO ₃) ₂ -extractable			
	Ni	Mn	Fe	Ni	Mn	Ca	Mg
	mg kg ⁻¹			g kg ⁻¹			
	Muck soil						
Control	358a [†]	4.78b	785a	2.48a	0.20a	1.73a	0.390c
HFO	302b	6.53a	740a	1.41b	0.12b	1.46b	0.323d
Limestone	224c	1.99c	399b	0.74c	0.06c	1.23c	0.801a
Limestone + HFO	195d	2.28c	351b	0.44d	0.04c	1.06d	0.644b
	Mineral soil						
Control	634a	13.5a	870b	54.2a	4.34a	1.27b	0.195c
HFO	605b	11.8b	943a	46.6b	4.09a	1.36a	0.206c
Limestone	239c	9.63c	387c	2.42c	0.26b	0.931d	0.646a
Limestone + HFO	221d	8.67c	379c	1.25d	0.09b	0.993c	0.618b

[†] Means within element and soil followed by the same letter are not significantly different according to Duncan's multiple range test ($P < 0.05$).

more Sr-extractable Ni than the control muck soil (Table 3). The lower extractability of Ni in the muck soil may be partially explained by the higher initial soil pH, the lower total Ni content, and higher quantities of organic matter capable of chelating metals in the muck soil (Dunemann et al., 1991). At acidic pH, humic acids in muck soil adsorb and chelate Ni strongly, while HFO adsorption of Ni is weak below pH 6 and sharply increases between pH 6 and 7. Hydrous ferric oxide applied as a single ameliorant significantly reduced Sr-extractable Ni in both soils. The reduction of Sr-extractable Ni by the HFO treatment was greater in the mineral than in the muck soil, 7.6 and 1.07 mg kg⁻¹ as compared with the control, respectively. However, in the mineral soil, the reduction was not sufficient to eliminate the strong Ni phytotoxicity. In both soils, limestone had a stronger ameliorative effect on Ni phytotoxicity and Sr extractability than HFO. Applied alone to the muck soil, limestone reduced Sr-extractable Ni by 70% in comparison with the control, while HFO reduced Sr-extractable Ni by only 43%. Limestone had a dramatic effect on Sr-extractable Ni in the mineral soil. The Sr-extractable Ni was reduced to about 2.5 mg Ni kg⁻¹ of dry soil, 22 times lower than the control, which enabled growth of the most sensitive crops. Application of both limestone and HFO further slightly reduced Sr-extractable Ni in both soils.

In contrast to 0.01 M Sr(NO₃)₂ extraction, which is conducted at actual soil pH conditions, the DTPA extractant is buffered at pH 7.3 by triethanolamine. The solution pH measured after extraction was in the range of 7.18 to 7.33 in this experiment. The lower pH values were found in the control and HFO treatments. Each ameliorant significantly decreased DTPA-extractable Ni in both soils (Table 3). Limestone was more effective than HFO, and the largest reduction in DTPA-Ni was obtained when both ameliorants were applied. Much greater quantities of Ni were extracted by DTPA than 0.01 M Sr(NO₃)₂, but the general tendency for decrease in extractable Ni with increasing soil pH was common for both methods. Within each soil, a strong correlation was obtained between DTPA- and Sr-extractable Ni. The correlation coefficients were 0.99 and 0.90 for the mineral and muck soil, respectively. Results of both extraction methods indicated that HFO and limestone

reduced plant-available Ni in soils. Calcium carbonate increases soil retention of metals by raising the pH, which in turn reduces proton competition with metal ions for adsorption sites. So far, there is no evidence that calcium carbonate itself can sorb or occlude significant amounts of metals (McBride, 1989), although this possibility should not be ruled out.

Theoretical considerations as well as experimental data led Sadiq and Enfield (1984a,b) to conclude that Ni carbonate and phosphate are too soluble to precipitate in soil. Formation of Ni hydroxide precipitate would require pH above 8.0. Comparing the theoretical solubility isotherm of Ni ferrite (NiFe₂O₄) with experimental data, they hypothesized that Ni ferrite may be a soil phase controlling Ni solubility in soils. Using X-ray absorption fine structure (XAFS) spectroscopy, Roberts et al. (1999) obtained direct evidence that a mixed Ni–Al hydroxide precipitated on a soil clay fraction even at pH of 6.8, although, at this pH, solution was undersaturated with respect to Ni(OH)₂. Therefore, it seems likely that at pH 7.5 attained by the soils amended with lime or HFO + lime, Ni precipitation, either in a form of a mixed Ni–Al hydroxide, Ni ferrite, or some other Ni compound, may have occurred. Others have considered slow reduction in soluble Ni in soils to be an occlusion process and inorganic Ni compounds have not been demonstrated to form in soils at the levels of contamination found in Port Colborne (C.J. Warren, ENPAR Technologies Inc., Guelph, ON, Canada, personal communication, 2000).

The capability of HFO to sorb metals is well known. Sorption of cations on HFO surfaces is highly dependent on pH. Retention of metal cations is low at pH below 4.5 to 6, depending on metal and properties of HFO. Retention dramatically increases at pH 6 and reaches a plateau above pH 7 (Stahl and James, 1991; Bryce et al., 1994; Lo et al., 1994). Reduction of extractable Ni associated with HFO application, especially pronounced in the mineral soil (by 7.6 mg kg⁻¹ in comparison with the control), is probably the result of metal sorption by HFO. However, a slight increase in soil pH induced by HFO amendment complicates the interpretation. The response curves of Ni extractability in relation to changes in soil pH are unknown for both soils. This makes it difficult to distinguish between Ni sorption on HFO and

increase in sorption on other components of soil induced by a slight increase in pH associated with application of HFO. Because only two pH levels (acid and calcareous) were tested, the design of this experiment did not allow for a full examination of the ameliorative potential of the HFO.

Effect of Treatments on Soil Iron, Manganese, Calcium, and Magnesium Extractability

Hydrous ferric oxide amendment increased DTPA-extractable Fe only in the mineral soil (Table 3), but the effect was not very pronounced. Making soil calcareous greatly reduced Fe extractability in both soils following the well-known lower Fe solubility and phytoavailability in calcareous soils (Römheld and Marschner, 1986). Similarly, Mn extractability was decreased by raising soil pH.

The $\text{Sr}(\text{NO}_3)_2$ -extractable Ca was decreased, and extractable Mg was increased by the amendment with Ca-Mg carbonate in such a manner that the sum of extracted Ca and Mg was approximately constant across all treatments within each soil. Calcium and Mg carbonates probably were the solid phases controlling solubility of both cations under alkaline conditions. The solubility of carbonates depends on their mineral form, but generally the solubility of Mg carbonates is much greater than solubility of Ca carbonates (Lindsay, 1979). In this situation, the decrease in $\text{Sr}(\text{NO}_3)_2$ -extractable, mostly exchangeable Ca in the limestone-amended muck and mineral soils may be related to a competition between Ca and Mg for adsorption sites.

Plant Growth and Symptoms Developed

Muck Soil

During the first four weeks after planting seeds in the muck soil, oat plants in all treatments looked healthy. Several days before harvest, a very mild chlorosis of the entire plant appeared in the control and HFO treatment. Interveneal chlorosis developed on the older leaves. Chlorosis progressed followed by appearance of grayish-green irregular spots, located near the leaf base in most cases. The spots were restricted to "middle age" leaves. These symptoms were recognized as *gray speck*, a classic symptom of Mn deficiency in oat (Mengel and Kirkby, 1982; Baldwin and Johnston, 1986), but due to

scheduled harvest of plants no attempt to correct the deficiency was made. The Mn concentrations in plants from the control and HFO-amended soil were less than 7 mg kg^{-1} , indicative of a severe Mn deficiency. At harvest, oat plants grown in the muck soil were at the flag leaf stage. Heads started to emerge in all treatments except for the control. Nickel concentrations in shoots of oat grown in the control and HFO-amended soil were above the toxicity threshold level for this species (Hunter and Vergnano, 1952) (Table 4), but the white banding diagnostic for Ni toxicity was not observed. It was not possible to determine with certainty whether the observed chlorosis was caused by Mn deficiency only or by a combination of Mn deficiency and Ni toxicity. Plants grown in soil amended with limestone without or with HFO looked healthy. Nickel concentrations in shoots were lowered by these treatments by more than 35% in comparison with the control and HFO treatment, while Mn levels were increased.

Initially, wheat grown in the muck soil looked healthy regardless of the treatments. About three weeks after planting seeds, wheat grown in the control and HFO-amended soil developed a very slight uniform chlorosis of the entire plant. With time, the uniform chlorosis progressed and interveinal chlorosis and yellowish-white discoloration somewhat resembling stripes developed on the older leaves. The younger leaves and stems were uniformly chlorotic with chlorosis being more pronounced in plants grown in HFO-treated soil. In control wheat, heads were almost fully emerged at the time of harvest. Growth of plants on HFO-treated soil was less advanced and heads had just started to emerge. The nutritional disorder experienced by plants grown in the control and HFO-amended soil was identified as Mn deficiency based on the postharvest shoot analysis (Table 5). Less advanced growth and stronger chlorosis in the HFO as compared with the control treatment corroborated a lower Mn concentration in shoots. Plants grown in the limestone and limestone plus HFO treatments had entered the flowering stage at the time of harvest. Both treatments resulted in healthy-looking plants. At harvest, Ni concentration in shoots of wheat grown in the control and HFO-amended muck soil (Table 5) was at the reported upper critical level (8 mg kg^{-1} ; Macnicol and Beckett, 1985), which is defined as the highest foliar concentration of the element not associated with yield reduction. Results of our related

Table 4. The effect of soil amendments on the elemental composition of oat shoots. HFO = hydrous ferric oxide.

Treatment	Ni	Mn	Fe	Zn	Cu	P	Mg	Ca	K	
	mg kg ⁻¹					g kg ⁻¹				
	Muck soil									
Control	62.9a†	5.92a	52.0a	77.2a	8.90a	5.20a	3.00a	7.40a	51.8a	
HFO	62.7a	6.56a	50.2ab	62.2b	8.38a	3.10b	3.36a	7.36a	48.3a	
Limestone	39.5b	16.5b	46.3ab	37.9c	6.26b	2.59bc	3.96b	3.35b	39.4b	
Limestone + HFO	40.8b	11.3c	43.2b	31.0c	6.10b	2.20c	3.91b	4.46b	42.4b	
	Mineral soil									
Control	692a	71.5a	48.7a	24.5a	6.52a	3.27a	4.19a	23.7a	3.2a	
HFO	399b	20.0b	27.0b	19.4b	4.84b	2.56b	3.00b	16.1b	12.8b	
Limestone	83.1c	95.1c	41.7ab	24.9a	9.26c	2.32b	4.96c	4.1c	30.8c	
Limestone + HFO	80.1c	108c	36.0ab	19.7b	9.47c	1.60c	3.67ab	3.8c	31.7c	

† Means within element and soil followed by the same letter are not significantly different according to Duncan's multiple range test ($P < 0.05$).

Table 5. The effect of soil amendments on the elemental composition of wheat shoots. HFO = hydrous ferric oxide.

Treatment	Ni	Mn	Fe	Zn	Cu	P	Mg	Ca	K	
	mg kg ⁻¹					g kg ⁻¹				
	Muck soil									
Control	7.98a†	7.25a	56.9a	65.3a	6.29a	4.68a	2.46a	6.74a	49.9a	
HFO	8.11a	6.04a	53.9a	71.1a	6.55a	3.01b	2.81ab	8.27b	57.9b	
Limestone	3.85b	12.3b	55.9a	47.6b	4.98b	2.65b	3.28b	3.87c	37.7c	
Limestone + HFO	2.82b	12.1b	46.4b	31.1c	4.50b	2.07c	3.12b	4.09c	42.6c	
	Mineral soil									
Control	271a	56.7a	29.7a	45.0a	8.15a	1.68a	2.44a	9.88a	21.0a	
HFO	81.4b	19.2b	28.5a	46.2a	6.12b	0.942b	1.64c	5.64b	21.9a	
Limestone	7.44c	90.1c	46.2b	24.1b	7.08b	2.39c	3.34b	3.23c	29.0b	
Limestone + HFO	7.54c	96.2c	44.0b	20.8b	6.55b	1.88a	2.87ab	2.92c	29.8b	

† Means within element and soil followed by the same letter are not significantly different according to Duncan's multiple range test ($P < 0.05$).

study (unpublished data, 1999) indicate that this variety of wheat is able to tolerate foliar Ni concentrations as high as 20 mg kg⁻¹ without any toxicity symptoms at later stages of growth or yield reduction. Strong chlorosis of plants, in this case, should instead be attributed to Mn deficiency, although synergistic effect of Mn deficiency on Ni toxicity should not be ruled out. Nickel levels in the shoots of wheat plants in limestone and limestone plus HFO-amended muck soil were well below the toxicity threshold. Shoot Mn concentrations were rather low but no deficiency symptoms were identified at any stage of plant growth.

Initially, all treatments resulted in healthy redbeet seedlings in the muck soil. The first very mild chlorosis appeared in plants grown in the control and limestone-treated soil after about three weeks of growth. However, in the limestone treatment, chlorosis disappeared at the later stages of growth. Hydrous ferric oxide applied alone or with limestone resulted in plant chlorosis at the later stages of growth. At harvest, plants had six or seven true leaves. There was no visible difference in plant size between treatments. Control plants had chlorotic older leaves, and some of the young leaves reached normal size but remained unfolded. Similarly, plants on the HFO treatment developed chlorosis of the older leaves. Hydrous ferric oxide amendment jointly with limestone induced the strongest chlorosis of entire plants. Redbeet grown in soil amended with limestone alone appeared completely healthy. Hydrous ferric oxide significantly reduced Ni concentration in the redbeet shoots but limestone alone was more effective in mitigating Ni uptake (Table 6). Joint application of limestone and HFO showed no additional mitigating effect compared

with limestone alone. Manganese acquisition by redbeet was adversely affected by both amendments. The lowest, strongly deficient Mn level in plant shoots was recorded at the joint application of limestone and HFO.

Symptoms exhibited by plants were probably the net effect of the alleviation of Ni toxicity and amendment-induced Mn deficiency. The threshold Ni phytotoxic concentration in redbeet shoots has not been reported in the literature. Symptoms developed by the control plants were attributed to Ni phytotoxicity, as no nutritional disorder other than elevated Ni concentration was revealed by analysis of plant tissue. Chlorosis observed in the HFO treatment was the result of low Mn status rather than Ni toxicity. This conclusion is supported by the observation that redbeet grown in the lime plus HFO-amended mineral soil looked completely healthy. Nickel concentrations in the shoots of redbeets grown in the HFO-amended muck soil and lime plus HFO-amended mineral soil were 24.4 and 25.9 mg kg⁻¹, respectively, and the only difference was the Mn status in plants (Table 6). Limestone apparently remediated Ni toxicity in the muck soil. Chlorosis developed by a joint limestone and HFO application should be attributed to severe Mn deficiency. This time the youngest leaves became chlorotic, which is typical of Mn deficiency (Mengel and Kirkby, 1982).

Mineral Soil

About 6 d after planting seeds, oat grown in the highly Ni phytotoxic control mineral soil exhibited classic symptoms of nickel toxicity. White banding perpendicular to leaf veins was present on cotyledonary leaves

Table 6. The effect of soil amendments on the elemental composition of redbeet shoots. HFO = hydrous ferric oxide.

Treatment	Ni	Mn	Fe	Zn	Cu	P	Mg	Ca	K	
	mg kg ⁻¹					g kg ⁻¹				
	Muck soil									
Control	32.6a†	25.3a	70.0a	85.0a	9.75a	3.58a	10.6a	18.8a	59.0a	
HFO	24.4b	14.8b	77.3a	68.4bc	10.9ab	2.96b	11.7a	18.4a	54.9a	
Limestone	12.2c	14.8b	72.7a	75.9ab	14.2c	3.09b	15.9b	12.4b	58.0a	
Limestone + HFO	9.74c	9.74c	88.0b	59.7c	11.8b	3.14b	16.5b	12.7b	58.2a	
	Mineral soil									
Control	no plants survived									
HFO	no plants survived									
Limestone	32.8a	121a	93.3a	26.3a	11.6a	3.47a	18.6a	14.9a	50.7a	
Limestone + HFO	25.9b	68.7a	103b	25.7a	10.3a	2.61b	16.3a	12.2b	54.2a	

† Means within element and soil followed by the same letter are not significantly different according to Duncan's multiple range test ($P < 0.05$).

that did not unfold completely and had a characteristic needle shape. Seedlings were smaller than in other treatments. Amendment with HFO did not cure Ni toxicity; severity of toxicity symptoms was similar to that of the control plants, but cotyledonary leaves were slightly bigger than in the control plants. Limestone applied without or with HFO completely alleviated Ni phytotoxicity symptoms. This pattern of response persisted to the end of the experiment. At harvest, oat grown in the control soil was severely damaged by Ni toxicity. Nickel level in shoots was 692 mg kg^{-1} , and many plants were dead (Table 4). Surviving plants were stunted. White chlorosis of leaves was accompanied by a brownish necrosis. The white and green banding pattern was very clear after germination but became diffuse at harvest. Green bands turned pale green or yellowish, while white bands became broader. Application of HFO significantly decreased shoot Ni concentration (399 mg kg^{-1}), but improvement of plant condition was minor. Flag leaves were produced by most plants in this treatment. All kinds of chlorosis were observed, including yellow-white uniform chlorosis, interveinal chlorosis, and the banded chlorosis, which indicates Ni phytotoxicity. Fe deficiency, observed only in this treatment, may have contributed to the development of the extremely strong chlorosis. Plants were severely stunted, the height of the tallest plants was about 10 cm. Limestone amendment without or with HFO made a dramatic difference in Ni uptake by oat and plant growth. Oat whole shoot Ni levels were reduced to 80 mg kg^{-1} . Although this Ni concentration was reported to be toxic to oat (Hunter and Vergnano, 1952), both treatments resulted in healthy plants with only a few white stripes observed on single leaves of single plants indicating some residual effect of Ni phytotoxicity. The nature of symptoms was so mild and scattered that they would be overlooked under field conditions. Heads started to emerge at harvest.

Six days after seeding, wheat seedlings in the control mineral soil were stunted and developed symptoms resembling Ni toxicity in oat, but banding was rather grayish than white and not as regular as in oat. Leaf tips were the most damaged. Similar symptoms plus an interveinal white chlorosis were observed in plants grown on soil amended with HFO. Limestone applied alone or with HFO produced healthy wheat seedlings. While conditions of plants grown in the control and HFO-amended soil deteriorated in the course of the experiment, plants grown in soil amended with limestone or limestone and HFO remained healthy. At harvest, the Ni level in wheat shoots in the control treatment was 271 mg kg^{-1} , and plants were severely stunted. Some of them produced flag leaves but no heads emerged. All kinds of chlorosis, including a uniform yellowing and interveinal and banded chlorosis, were present. Especially clear white and green banding perpendicular to leaf veins developed on flag leaves. Results of tissue analysis confirmed that all these symptoms should be attributed to Ni toxicity and Ni-toxicity-induced Fe deficiency (Table 5). Hydrous ferric oxide amendment lowered shoot Ni concentration to 81 mg kg^{-1} and visibly improved plant condition, but

the effect was far from being satisfactory. Plants were stunted. Heads emerged but their size was strongly reduced. Stems, younger leaves, and flag leaves were fully green. Older leaves had brownish necrosis starting from the tips and progressing to the middle of leaf blades. Limestone applied without or with HFO lowered shoot Ni level well below toxicity threshold and fully cured toxicity symptoms. Plants looked healthy and heads had begun to flower at the time of harvest.

The phytotoxic effect of Ni on redbeet in the control and HFO-amended mineral soil was already evident at the germination stage. Seed germination percentage was adversely affected on the control mineral soil. Only a few plants germinated in each control pot, and all of them died shortly after germination. A similar situation was observed in the pots amended with HFO. Initially, no toxicity symptoms were recorded in the seedlings grown in the limestone-treated pots as well as pots that received limestone and HFO. The first appearance of chlorosis in the limestone treatment was observed about three weeks after plant seeding. At harvest, strong chlorosis developed on all except the youngest leaves in this treatment. It was probably a manifestation of Ni toxicity, which was greatly alleviated or completely cured by a joint application of limestone and HFO, as most plants in this treatment treatment looked very healthy and only a few exhibited very mild chlorosis. Joint application of both ameliorants not only significantly decreased shoot Ni concentration but also improved acquisition of Fe (Table 6), which has a mitigating effect on Ni toxicity (Crooke et al., 1954).

Elemental Composition of Plants

In both soils and all species tested, making soil calcareous significantly reduced Ni concentrations in plant shoots. Hydrous ferric oxide applied alone to acid-contaminated soils was ineffective. The redbeet results indicate that HFO applied jointly with limestone may have a beneficial effect in protecting crops in the short term. Studies involving more species representing dicot and *Poaceae* families are necessary to answer the question of whether the beneficial effect of HFO is species- or rather plant family-specific. In the longer term, aging of Ni sorbed and/or occluded to HFO should reduce soluble Ni significantly (Bruemmer et al., 1988).

Despite application of high rates of MnSO_4 , the Mn concentrations in the shoots of plants grown in the muck soil were barely sufficient or clearly deficient (Tables 4–6). The most severe Mn deficiency occurred in wheat and oat grown in the control and HFO-amended muck soil, while Mn status in plants grown in limestone-amended soil was much better. In contrast to the grass family, redbeet (representing dicot crops) suffered the strongest Mn deficiency when the muck soil was amended with limestone plus HFO. Manganese deficiency symptoms were absent in all species at the early stage of growth when Mn applied as MnSO_4 was still in plant-available form. As Mn fixation by soil progressed and seed Mn was diluted, deficiency symptoms appeared in wheat as a very slight uniform chlorosis of the entire

plant about three weeks after seed planting. This early warning was not initially interpreted by us as Mn deficiency as it was assumed that the high rate of Mn fertilizer applied would have prevented Mn deficiency. In oat plants, Mn deficiency symptoms appeared much later. The mineral soil did not induce Mn deficiency even when made calcareous. Neither strontium nitrate–nor DTPA-extractable Mn provided a good prediction of the plant availability of soil Mn, except for redbeet; but both methods confirmed a high binding capacity of the muck soil for Mn as compared with mineral soil. Manganese deficiency has been recognized as a very common nutritional disorder in the farmed organic soils on the Lake Erie lake plain in Ontario; therefore, Mn fertilization is recommended in agricultural practice on these soils irrespective of Ni contamination (Baldwin and Johnston, 1986).

Hydrous ferric oxide application did not increase Fe concentrations in either of the *Poaceae* species tested. The only statistically significant increase in plant Fe concentration compared with the control was observed in redbeet grown in either muck or mineral soil amended with limestone and HFO. Plants normally limit Fe to <100 mg kg⁻¹ dry wt. unrelated to soil Fe levels (Chaney, 1984). Any increase in shoot Fe would be beneficial for plants because the toxic effect of Ni may be mitigated by increased Fe concentrations in plant tissue (Crooke and Knight, 1955; Crooke et al., 1954). Instead, it seems that Ni successfully competed with Fe for uptake by oat and wheat grown in the control and HFO-amended acidic mineral soil. Alleviation of Ni toxicity by limestone enabled these plant species to increase shoot Fe concentration to healthy levels despite decrease in plant-available Fe in soil as indicated by DTPA test (Table 3).

Magnesium level in all crops was significantly increased by the application of Mg-containing limestone, while plant Ca levels were significantly lowered. Two alternative or simultaneous hypotheses can be proposed to explain this phenomenon. The plausible explanation is a decrease in plant-available soil Ca and increase in Mg, as indicated by Sr(NO₃)₂ extraction, and a competition between Ca and Mg for plant uptake. This seems to be a valid explanation for the plants that did not suffer any Ni toxicity or experienced a minor Ni phytotoxicity, like wheat and redbeet grown in the muck soil. The Ca and K uptake pattern by oat in mineral soil (Table 4) suggests that extreme Ni toxicity may also affect an acquisition of essential nutrients in a manner independent of the element status in the soil. Similarly, the increase of Ca acquisition induced by high Ni

levels was observed by Brune and Dietz (1995) in a nutrient solution study. The pattern of amendment-induced changes in P status in plant shoots was uniform among species and soils. Both amendments significantly reduced P availability to plants as indicated by plant tissue analyses, but the high rate of P fertilizer needed for valid pot experiments prevented any deficiency from occurring except for the wheat grown in HFO-amended mineral soil. The severe P deficiency observed in this case should instead be attributed to the toxic effect of Ni.

Plant Yield

Manganese deficiency experienced by all crops grown in a muck soil confounds the evaluation of Ni toxicity and the effectiveness of ameliorants in relation to plant yield. There was a statistically significant increase in oat yield, in comparison with the control, associated with application of both amendments, but typical symptoms of Ni toxicity could not be identified at any stage of growth (Table 7). A similar situation was repeated in the case of wheat. Application of limestone with and without HFO significantly increased yield in comparison with the control and HFO treatments. This yield difference is consistent with the potentially phytotoxic Ni concentrations in shoots of plants grown in the control and HFO-amended soil. On the other hand, yield reduction could have been caused by severe Mn deficiency. While Ni concentration in the shoots of redbeet grown in the muck soil was significantly lowered by both amendments, the yield remained unaffected, possibly because of amendment-induced Mn deficiency. Manganese levels in redbeet shoots paralleled Ni levels, which means that beneficial effect of decreased Ni uptake on yield could have been counteracted by Mn deficiency, even when 100 kg Mn ha⁻¹ was applied.

In the highly phytotoxic mineral soil, in an absence of Mn deficiency, yield response to treatments was very clear. Making the soil calcareous dramatically increased yield and was somewhat better than joint application of limestone and HFO. Wheat and oat plants in the limestone + HFO treatment looked as healthy as plants in limestone-only treated soil. Under this circumstance there is no reason to attach any negative meaning to this yield reduction.

The Relationship between Soil-Extractable Metals and Plant Metals

Metals are extracted from soils by various means and solutions in order to predict metal toxicity in soil and to evaluate ameliorative effects of various treatments.

Table 7. Dry matter of plants grown in Quarry muck and Welland loam amended with limestone and/or hydrous ferric oxide (HFO).

Treatment	Quarry muck			Welland loam		
	Oat	Wheat	Redbeet	Oat	Wheat	Redbeet
	g dry weight per pot					
Control	4.98a†	3.56a	6.58a	0.20a	0.98a	0.0a
HFO	6.81b	2.96a	6.61a	0.25a	1.42a	0.0a
Limestone	6.33b	5.34b	6.35a	6.82b	6.01b	4.14b
Limestone + HFO	8.12c	5.95b	6.48a	4.78c	5.04c	4.12b

† Means within column followed by the same letter are not significantly different according to Duncan's multiple range test ($P < 0.05$).

In our study, two methods based on quite different extraction principles were used. The extraction method has a predictive value if there is a correlation between the extractable pool of metal in a soil and the uptake of metal by a plant. Ideally, such a relationship should be linear, which would mean that plant uptake is proportional to the extractable pool of metal in the soil. This is usually not the case under conditions of extreme toxicity of metals. In these extreme conditions, plant physiological processes are strongly affected, which leads to an exponential increase of the contaminant concentration in plant tissue and an abnormal uptake of other essential and nonessential elements. Oat and wheat are an example of such a situation. When all treatments in both soils were combined in one graph, the relationship between $\text{Sr}(\text{NO}_3)_2$ -extractable soil Ni and Ni concentration in oat and wheat shoots followed an exponential pattern (Fig. 1). Very similar trends were obtained for DTPA-extractable soil Ni, with R^2 values of 0.89 and 0.88 for oat and wheat, respectively. The control and HFO treatments in the Welland soil, which resulted in extremely high Ni levels in oat and wheat shoots, will be omitted in the further discussion on predictive value of both extractants.

The DTPA method is probably the most commonly used test for Ni-contaminated soils (Brown et al., 1989; Sauerbeck and Hein, 1991; Sheets et al., 1982) and is considered to be indicative of soil Ni phytotoxicity for soils with a small range in pH. When Haq and Miller (1972) tested soils with a wide range of pH, Zn and Mn concentration in corn (*Zea mays* L.) shoots was predicted well only when soil pH was included in the regression equation. Haq et al. (1980) found a similar relationship for soil Ni uptake by Swiss chard [*Beta vulgaris* L. subsp. *cicla* (L.) W.D.J. Koch var. *flavescens* (Lam.) DC.].

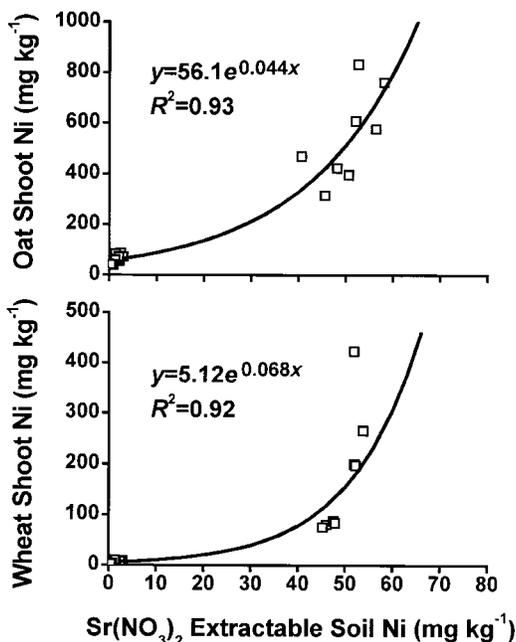


Fig. 1. The relationship between $\text{Sr}(\text{NO}_3)_2$ -extractable soil Ni and Ni concentrations in oat and wheat shoots.

There was no correlation between DTPA-extractable soil Ni and its concentration in oat shoots (Fig. 2a), but the DTPA test had a somewhat better predictive value for wheat and redbeet (Fig. 2b,c). Improvement of the predictive value of DTPA-extractable Ni for all crops was achieved when concentrations of extractable Ni were expressed in mg dm^{-3} soil rather than in mg kg^{-1} soil (Fig. 2d,e,f). This approach takes into account differences in bulk densities of the soils. Usually, differences in soil bulk densities can be ignored, but comparison of two soils having such different bulk densities comprises a special problem. The difference in total Ni concentration between mineral and muck soil (2930 versus 2210 mg kg^{-1}) was 720 mg kg^{-1} when concentrations were expressed as mg kg^{-1} . When Ni concentrations were expressed in mg dm^{-3} soil, the difference between the mineral and muck soil was much greater (2110 versus 729 mg dm^{-3}). Soil volume-based comparisons better illustrate the conditions plant roots are exposed to. Undoubtedly, there are many factors affecting Ni transfer between soil solid phase, soil solution, and plant roots, including specific surface area of the soil solid phase and pore size distribution, which in turn affect the interface between solid and liquid phase. Expression of total and extractable metal in milligrams per unit of soil volume oversimplifies the problem but seems to help in comparing the phytotoxicity of soils.

Strontium-extractable Ni was relatively well correlated with metal concentrations in plant shoots (Table 8). For oat, the correlation was further improved when concentration of extractable soil Ni was corrected for soil bulk density. This correction did not improve the correlation for redbeet and even further decreased correlation for wheat. The predictive value of both soil tests was similar when concentrations of extractable Ni were expressed in mg dm^{-3} , but the $\text{Sr}(\text{NO}_3)_2$ extractant was superior when the mg kg^{-1} unit was used.

Further improvement of predictive value of both soil tests was achieved when soil pH was incorporated into equations as an independent variable (Table 8). While the $\text{Sr}(\text{NO}_3)_2$ extractant predicted plant availability of soil Ni relatively well, the DTPA method required soil pH and bulk density to be taken into account to obtain meaningful results. The DTPA test combines two aspects of metal availability to plants: the intensity factor, which is related to the activity of metal in soil solution; and the capacity factor, characterizing the ability of the soil solid phase to replenish metal ions as they are absorbed by roots (Lindsay and Norvell, 1978). The significant improvement of the predictive value of the DTPA test by correction of the extraction results for soil bulk density may be related to the soil capacity factor.

The role of HFO in alleviation of Ni phytotoxicity remains unclear, perhaps because of the short growth period used in this study. Soil tests indicated that a reduction of extractable Ni occurred in both soils when HFO was applied alone or with limestone. This trend corroborates experimental data as well as theoretical principles of metal sorption on HFO (Bruemmer et al., 1988; Lo et al., 1994; Stahl and James, 1991). Oat and wheat, representing the *Poaceae* (grass) family, re-

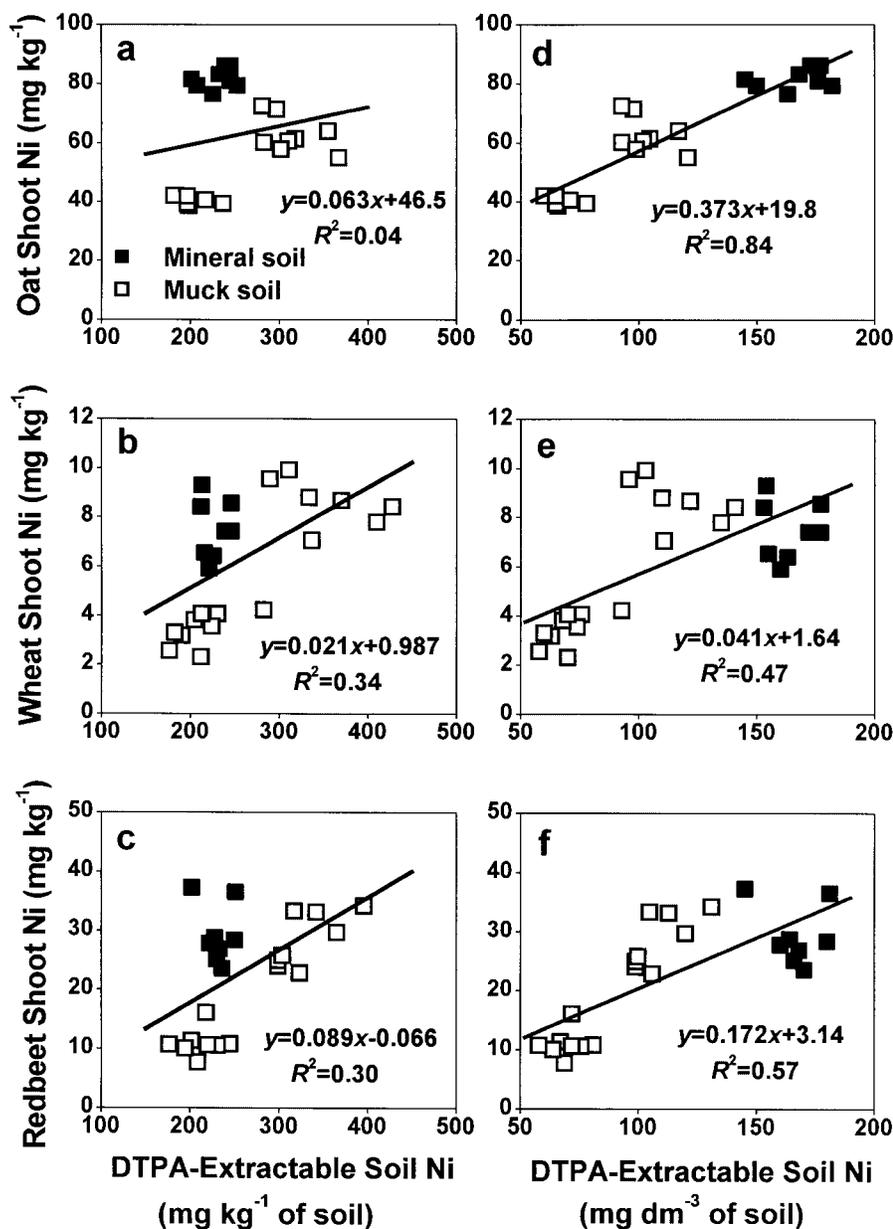


Fig. 2. The relationship between DTPA-extractable soil Ni (in mg kg^{-1} and mg dm^{-3} soil) and plant shoot Ni.

sponded to HFO amendment with decreased Ni uptake in only one case. In the mineral soil, HFO applied alone markedly reduced Ni concentrations in both species in relation to control plants. Furthermore, HFO amendment did not improve Fe acquisition by either *Poaceae* species, which could have been beneficial, as it is believed that increased Fe level in oat shoots can significantly mitigate the toxic effect of Ni. In contrast, the dicot redbeet always responded to HFO application with decrease in Ni uptake and increase in Fe acquisition. It is possible that the mechanism used by *Poaceae* species to obtain soil Fe contributed to this result. These species secrete phytosiderophores (avenic acid by oat and deoxymugeinic acid by wheat), which chelate soil Fe, and after diffusion back to the root the intact ferric chelate is absorbed (Römheld and Marschner, 1986). As noted by Chambers et al. (1998), unlike microbial

siderophores, chelation of ferric iron by phytosiderophores is not highly selective, because the ligands of phytosiderophores do not include phenolic or hydroxamate functional groups. Thus, Ni probably can displace Fe from Fe-phytosiderophore chelates. Further studies are needed to characterize the chemistry and physiology of Fe-Ni-phytosiderophore interactions in the rhizosphere of Ni-phytotoxic soils. Dicots such as redbeet use a reductive mechanism for releasing soil Fe for uptake as ferrous ion, and Ni^{2+} is known to inhibit uptake of Fe^{2+} by roots. It appears that the increase in reducible HFO aided redbeet obtain Fe strongly enough so that HFO contributed to reduction of Ni phytoavailability and phytotoxicity to redbeet.

Neither DTPA nor Sr extraction allowed for estimation of phytoavailable Mn for oat and wheat. The effect of limestone and HFO treatments on soil-extractable

Table 8. Values of coefficient of determination (R^2) for multiple linear regression equations describing the effect of soil-extractable Ni (expressed in mg kg^{-1} or mg dm^{-3} of soil) and pH on Ni levels in plant shoots. DTPA = diethylenetriaminepentaacetic acid.

Plant	Model: Plant Ni = $a + b(\text{soil Ni}) + c(\text{soil pH})$ †					
	Partial R^2 ‡, soil Ni (mg kg^{-1})	Partial R^2 , soil pH	Model R^2 §	Partial R^2 , soil Ni (mg dm^{-3})	Partial R^2 , soil pH	Model R^2
Extractant: 0.01 M $\text{Sr}(\text{NO}_3)_2$						
Oat	0.55	0.14	0.69	0.79	ns¶	0.79
Wheat	0.48	0.05	0.53	0.29	0.32	0.61
Redbeet	0.69	ns	0.69	0.65	0.2	0.85
Extractant: DTPA						
Oat	0.04	ns	0.04	0.84	0.03	0.87
Wheat	0.34	ns	0.34	0.47	0.26	0.73
Redbeet	0.3	ns	0.3	0.57	0.27	0.84

† Stepwise linear regression analysis was used for fitting equations. a , b , and c are coefficients in the linear equation. Soil Ni, soil pH, and plant Ni are variables. Soil Ni = $\text{Sr}(\text{NO}_3)_2$ - or DTPA-extractable soil Ni expressed in mg kg^{-1} or mg dm^{-3} soil. Soil pH = soil pH measured in a water slurry at plant harvest (soil to water ratio 1:2 (v/v), equilibration time 2 h). Plant Ni = Ni concentration in wheat, oat, or redbeet shoots in mg kg^{-1} of dry matter.

‡ Partial R^2 indicates the amount of variance explained by each variable included in the model.

§ Model R^2 is the coefficient of determination for the linear model including both variables soil Ni and soil pH.

¶ Variable did not meet 0.1500 significance level for entry into the model.

Mn was opposite to their effect on Mn concentrations in oat and wheat shoots. Both soil extraction methods had only qualitative value for oat and wheat in the sense that they confirmed a better Mn status of the mineral soil. In contrast to other crops, Mn concentrations in the redbeet shoots paralleled changes in soil-extractable Mn [$R^2 = 0.69$ and 0.71 for DTPA and $\text{Sr}(\text{NO}_3)_2$, respectively]. This suggests that different species may use different strategies for Mn acquisition. Both soil tests better predicted Mn availability to redbeet when extractable Mn was expressed in mg per dm^{-3} of soil [$R^2 = 0.88$ and 0.89 for DTPA and $\text{Sr}(\text{NO}_3)_2$, respectively].

The levels of $\text{Sr}(\text{NO}_3)_2$ -extractable Ca and Mg correlated with uptake of these elements by plants. Higher concentrations of Mg in shoots of plants grown in limestone-amended soils corresponded with higher levels of soil-extractable Mg. Calcium exhibited an opposite tendency. Lower levels of extractable Ca and, accordingly, lower shoot concentrations were found in calcareous soils. As discussed earlier, Ni toxicity in the control and HFO-amended soils may have also contributed to increased Ca uptake by plants under acid pH conditions.

CONCLUSIONS

This pot study demonstrated that an extreme Ni phytotoxicity in industrially contaminated soil can be successfully ameliorated with inexpensive soil amendments. Limestone applied at the rate equivalent to 50 Mg ha^{-1} completely cured Ni toxicity in the moderately phytotoxic muck soil and almost completely alleviated Ni toxicity in the mineral soil, which was so highly phytotoxic that it prevented any growth of redbeet and almost killed oat and wheat. High rates of Mn and P fertilizers were required to prevent amendment-induced deficiencies of these elements. Hydrous ferric oxide applied to an acid mineral soil somewhat alleviated Ni toxicity in wheat and oat but its ameliorative effect was rather minor. There was no beneficial effect of HFO on these crops for either soil when HFO was applied in combination with limestone. In contrast, redbeet responded to HFO application with decreased Ni and increased Fe uptake; high levels of Fe in plant tissue are believed to

have a protective function against Ni toxicity. It is likely that the difference in response to HFO application is plant family-specific. A study involving numerous species representing both *Poaceae* and dicot families is needed to explore this hypothesis. Further study is needed to understand how soil pH modifies ameliorative effect of HFO. Soil extraction with unbuffered salt solution [$0.01 \text{ M Sr}(\text{NO}_3)_2$] satisfactorily predicted plant uptake of Ni. The DTPA test was equally effective when soil pH and bulk density were taken into account while interpreting extraction results.

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REFERENCES

- Backes, C.A., R.G. McLaren, A.W. Rate, and R.S. Swift. 1995. Kinetics of cadmium and cobalt desorption from iron and manganese oxides. *Soil Sci. Soc. Am. J.* 59:778-785.
- Baldwin, C.S., and R.W. Johnston. 1986. Manganese in soybean and small grain production. Ministry of Agriculture and Food, Ontario, Canada.
- Bartlett, R.J., and B.R. James. 1980. Studying dried, stored soil samples—Some pitfalls. *Soil Sci. Soc. Am. J.* 44:721-724.
- Bisessar, S. 1989. Effects of lime on nickel uptake and toxicity in celery grown on muck soil contaminated by a nickel refinery. *Sci. Total Environ.* 84:83-90.
- Brown, P.H., L. Dunemann, R. Schulz, and H. Marschner. 1989. Influence of redox potential and plant species on the uptake of nickel and cadmium from soils. *Z. Pflanzenernähr. Bodenk.* 152:85-91.
- Brown, S.L., J.S. Angle, and R.L. Chaney. 1997. Correction of limed-biosolid induced manganese deficiency on a long-term field experiment. *J. Environ. Qual.* 26:1375-1384.
- Brown, S.L., and R.L. Chaney. 2000. Combining by-products to achieve specific soil amendment objectives. p. 343-360. *In* J.F. Power et al. (ed.) Land application of agricultural, industrial and municipal by-products. SSSA Book Ser. 6. SSSA, Madison, WI.
- Brummer, G.W., J. Gerth, and K.G. Tiller. 1988. Reaction kinetics

- of the adsorption and desorption of nickel, zinc, and cadmium by goethite. I. Adsorption and diffusion of metals. *J. Soil Sci.* 39:37–52.
- Brune, A., and K.-J. Dietz. 1995. A comparative analysis of element composition of roots and leaves of barley seedlings grown in the presence of toxic cadmium, molybdenum, nickel, and zinc concentrations. *J. Plant Nutr.* 18:853–868.
- Bryce, A.L., W.A. Kornicker, A.W. Elzerman, and S.B. Clark. 1994. Nickel adsorption to hydrous ferric oxide in the presence of EDTA: Effects of component addition sequence. *Environ. Sci. Technol.* 28:2353–2359.
- Chambers, D.B., R.L. Chaney, B.R. Conard, N.C. Garisto, U. Kukier, H.A. Phillips, and S. Fernandes. 1998. Risk assessment for nickel in soil, with a critical review of soil-nickel phytotoxicity. White Paper Rep. to Ontario Ministry of the Environ. for Inco Ltd. SENES, Richmond Hill, ON, Canada.
- Chaney, R.L. 1984. Diagnostic practices to identify iron deficiency in higher plants. *J. Plant Nutr.* 7:47–67.
- Chaney, R.L., and U. Kukier. 1998. Remediating Ni-phytotoxicity of contaminated muck soil using lime and hydrous iron oxide. INCO Ltd., Toronto.
- Chlopecka, A., and D.C. Adriano. 1996. Mimicked in-situ stabilization of metals in a cropped soil: Bioavailability and chemical form of zinc. *Environ. Sci. Technol.* 30:3294–3303.
- Crooke, W.M. 1956. Effect of soil reaction on uptake of nickel from a serpentine soil. *Soil Sci.* 81:269–276.
- Crooke, W.M., J.G. Hunter, and O. Vergnano. 1954. The relationship between nickel toxicity and iron supply. *Ann. Appl. Biol.* 41:311–324.
- Crooke, W.M., and A.H. Knight. 1955. The relationship between nickel-toxicity symptoms and the absorption of iron and nickel. *Ann. Appl. Biol.* 43:454–464.
- Dunemann, L., N. von Wiren, R. Schulz, and H. Marschner. 1991. Speciation analysis of nickel in soil solutions and availability to oat plants. *Plant Soil* 133:263–269.
- Frank, R., K.I. Stonefield, P. Suda, and J.W. Potter. 1982. Impact of nickel contamination on the production of vegetables on an organic soil, Ontario, Canada, 1980–1981. *Sci. Total Environ.* 26:41–65.
- Gee, G.W., and J.W. Bauder. 1986. Particle size analysis. p. 383–411. *In* A. Klute et al. (ed.) *Methods of soil analysis. Part 1. Physical and mineralogical methods.* 2nd ed. Agron. Monogr. 9. ASA and SSSA, Madison, WI.
- Haq, A.U., T.E. Bates, and Y.K. Soon. 1980. Comparison of extractants for plant-available zinc, cadmium, nickel, and copper in contaminated soils. *Soil Sci. Soc. Am. J.* 44:772–777.
- Haq, A.U., and M.H. Miller. 1972. Prediction of available soil Zn, Cu, and Mn using chemical extractants. *Agron. J.* 64:779–782.
- Hunter, J.G., and O. Vergnano. 1952. Nickel toxicity in plants. *Ann. Appl. Biol.* 39:279–284.
- Hunter, J.G., and O. Vergnano. 1953. Trace-element toxicities in oat plants. *Ann. Appl. Biol.* 40:761–777.
- King, L.D. 1988. Retention of metals by several soils of the Southern United States. *J. Environ. Qual.* 17:239–246.
- Kuja, A., R. Jones, and W. McIlveen. 2001. Phytotoxicology soil investigation: INCO–Port Colborne (1999), Ontario Ministry of the Environment, July 2000. Rep. SDB-031-3511-2000. Queen's Printer for Ontario, Toronto.
- Kukier, U., and R.L. Chaney. 2000. Remediating Ni-phytotoxicity of contaminated Quarry muck soil using limestone and hydrous iron oxide. *Can. J. Soil Sci.* 80:581–593.
- L'Huillier, L., and S. Edighoffer. 1996. Extractability of nickel and its concentration in cultivated plants in Ni-rich ultramafic soils of New Caledonia. *Plant Soil* 186:255–264.
- Lindsay, W.L. 1979. *Chemical equilibria in soils.* John Wiley & Sons, New York.
- Lindsay, W.L., and W.A. Norvell. 1978. Development of a DTPA soil test for zinc, iron, manganese, and copper. *Soil Sci. Soc. Am. J.* 42:421–428.
- Lo, S.L., S.H. Shiu, C.F. Lin, and D.Y. Lee. 1994. Adsorption of metals on oxides formed from $\text{Fe}(\text{NO}_3)_3$, FeCl_3 and $\text{Fe}(\text{ClO}_4)_3$ solutions. p. 89–99. *In* D.C. Adriano et al. (ed.) *Biogeochemistry of trace elements.* Environ. Geochem. and Health 16. Science and Technol. Letters, Northwood.
- Macnicol, R.D., and P.H.T. Beckett. 1985. Critical tissue concentrations of potentially toxic elements. *Plant Soil* 85:107–128.
- Madden, M.S. 1988. Adapting the $\text{Sr}(\text{NO}_3)_2$ method for determining available cations to a routine soil testing procedure. M.Sc. thesis. University of Wisconsin, Madison.
- McBride, M.B. 1989. Reactions controlling heavy metal solubility in soils. *Adv. Soil Sci.* 10:1–56.
- Mench, M.J., V.L. Didier, M. Löffler, A. Gomez, and P. Masson. 1994. A mimicked in-situ remediation study of metal-contaminated soils with emphasis on cadmium and lead. *J. Environ. Qual.* 23:58–63.
- Mengel, K., and E.A. Kirkby. 1982. *Principles of plant nutrition.* International Potash Institute, Worblauen-Bern, Switzerland.
- Roberts, D.R., A.M. Scheidegger, and D.L. Sparks. 1999. Kinetics of mixed Ni–Al precipitate formation on a soil clay fraction. *Environ. Sci. Technol.* 33:3749–3754.
- Römheld, V., and H. Marschner. 1986. Mobilization of iron in the rhizosphere of different plant species. p. 155–204. *In* B. Tinker and A. Läuchli (ed.) *Advances in plant nutrition.* Volume 2. Praeger Publ., New York.
- Sadiq, M., and C.G. Enfield. 1984a. Solid phase formation and solution chemistry of nickel in soils: 1. Theoretical. *Soil Sci.* 138:262–270.
- Sadiq, M., and C.G. Enfield. 1984b. Solid phase formation and solution chemistry of nickel in soils: 1. Experimental. *Soil Sci.* 138:335–340.
- SAS Institute. 1988. *SAS/STAT user's guide.* Release 6.03 ed. SAS Inst., Cary, NC.
- Sauerbeck, D.R., and A. Hein. 1991. The nickel uptake from different soils and its prediction by chemical extractions. *Water Air Soil Pollut.* 57–58:861–871.
- Sheets, P.J., V.V. Volk, and E.H. Gardner. 1982. Plant and soil reactions to nickel ore processed tailings. *J. Environ. Qual.* 11:446–451.
- Stahl, R.S., and B.R. James. 1991. Zinc sorption by iron-oxide-coated sand as a function of pH. *Soil Sci. Soc. Am. J.* 55:1287–1290.
- Temple, P.J., and S. Bisessar. 1981. Uptake and toxicity of nickel and other metals in crops grown on soil contaminated by a nickel refinery. *J. Plant Nutr.* 3:473–482.
- USEPA. 1995. *Test methods for evaluating soils and wastes.* Vol. IA: Laboratory manual physical/chemical methods. SW 846 3rd ed. U.S. Gov. Print. Office, Washington, DC.