This is to certify that the

dissertation entitled

SOIL TRACE ELEMENTS LOADED IN
HIGH AMOUNTS FROM SEWAGE SLUDGE APPLICATIONS:
CHEMICAL FRACTIONATION, MOVEMENT, AND BIOAVAILABILITY

presented by

William Robert Berti

has been accepted towards fulfillment
of the requirements for

Ph.D. degree in Crop & Soil Sciences/
Institute of
Environmental Toxicology

Lee W. Jacobs
Major professor

Date 11/12/92

SOIL TRACE ELEMENTS LOADED IN HIGH AMOUNTS FROM SEWAGE SLUDGE APPLICATIONS: CHEMICAL FRACTIONATION, MOVEMENT, AND BIOAVAILABILITY

By

William Robert Berti

A DISSERTATION

Submitted to
Michigan State University
in partial fulfillment of the requirements
of the degree of

DOCTOR OF PHILOSOPHY

Department of Crop and Soil Sciences
Institute for Environmental Toxicology

1992

ABSTRACT

SOIL TRACE ELEMENTS LOADED IN HIGH AMOUNTS FROM SEWAGE SLUDGE APPLICATIONS: CHEMICAL FRACTIONATION, MOVEMENT, AND BIOAVAILABILITY

By

William Robert Berti

The protection of our soil resource requires an understanding of the fate of trace elements in the biosphere. Sequential extraction techniques, which chemically fractionate trace elements, should help ascertain their environmental availability in soil. Experimental comparisons of two techniques indicated that fractionation data was dependent on the method used. Method selection must be based on both theoretical and empirical information of the techniques and on the soils and trace elements of interest. From 1977 to 1986, municipal sludges containing Cd, Cr, Cu, Ni, Pb, and Zn were applied in greater than normal concentrations to research plots. Total elemental analysis of soils collected in 1989 and 1990 indicated some lateral movement of trace elements associated with physically moving soil particles during agronomic operations. These elements, however, had not leached below the 15 to 30 cm sample depth into which they were incorporated. Mass balance calculations of trace elements resulted in average recoveries from 45 to 114% of the total applied. These calculations were highly variable, indicative of the variable nature of sewage sludge composition, lack of totally uniform sludge applications, soil movement due to tillage, and sampling methods. Soil chemical fractionation demonstrated that Cd, Cu, and Zn resided primarily in the exchangeable

and acid-soluble fractions, Cr in the organic and Fe oxide fractions, Ni in the acid-soluble and Fe oxide fractions, and Pb in the residual fraction. Plant uptake of trace elements was variable from year to year, plant part, and crop. Results of soil chemical fractionation and plant analysis suggested that Cd, Ni, and Zn continued to be environmentally available, whereas Cr and Cu were relatively less available, and Pb was not environmentally available. Soil test methods for trace elements that were taken up by plants generally correlated well with the most labile soil fractions (i.e., water-soluble, exchangeable, and acid-soluble). Toxicity Characteristic Leaching Procedure (TCLP) was inappropriate to use when testing soils to access concentrations of trace elements that are toxic to plants.

ACKNOWLEDGMENTS

I wish to express my sincere thanks to Dr. Lee Jacobs for his support. Lee had been of immense help in the development of this dissertation, from his suggestions and discussions of research opportunities to his comments about the written dissertation. The time that he has given me and example he has set in helping me become a better scientist are appreciated.

My committee members have made a substantial contribution to my development as a scientist in the courses that I have taken from them and in our many discussions. My gratitude to Drs. Boyd Ellis, Stephen Boyd, and Matthew Zabik.

There have been many who have helped me in the laboratory and in the field. I wish to single out Sven Böhm, Ben Lentz, and Bruce McKellar whose help and friendship have been of special importance to me. Also, the assistance of Brian Graff and Dallas Hyde, who were never too busy to lend a hand, is gratefully acknowledged. To those who helped with establishing and maintaining the field plots on which this dissertation so heavily relied, I am in your debt.

Finally, my thanks to my family and many friends who have helped me in way immeasurable, with a special note of appreciation to Rhonda.

TABLE OF CONTENTS

CHAPTER ONE:

CHEMICAL FRACTIONATION OF SOILS: A COMPARISON OF TWO METHODS FOR SEQUENTIALLY EXTRACTING Cd, Cr, Cu, Ni, Pb, AND Zn

CHAPTER TWO:

MOVEMENT OF Cd, Cr, Cu, Ni, Pb, AND Zn FROM MUNICIPAL SEWAGE SLUDGES IN A SANDY LOAM SOIL

CHAPTER THREE:

CHEMICAL FRACTIONATION AND PLANT UPTAKE OF Cd, Cr, Cu, Ni, Pb, and Zn IN A SANDY LOAM SOIL FROM THE APPLICATION OF MUNICIPAL SEWAGE SLUDGES

LIST OF TABLES

LIST OF FIGURES

CHAPTER ONE:

CHEMICAL FRACTIONATION OF SOILS:
A COMPARISON OF TWO METHODS FOR SEQUENTIALLY EXTRACTING
Cd, Cr, Cu, Ni, Pb, AND Zn

ABSTRACT

Discovering the chemical forms in which elements in the biosphere occur should help determine their environmental availability. Sequential extraction techniques frequently have been used to chemically fractionate trace elements in soils, sediments, geologic materials, and water. Fractions measured have included water-soluble, exchangeable, carbonate, organic, Mn and Fe oxides, and residual components. In this chapter, theoretical issues and methodologies involved in the development of soil chemical fractionation techniques are reviewed. Two were selected for further study. Three soils, collected from field research plots to which municipal sewage sludges were applied, were sequentially extracted using the two methods. Fractionation data was dependent on the method used, and comparing results of the two methods alone did not resolve which technique was more appropriate. Rather, selection must be based on theoretical knowledge and empirical data of the techniques for specific soils and trace elements of interest. A method from a technique by Miller et al. (1986) was found more suitable for simultaneously extracting Cd, Cr, Cu, Ni, Pb, and Zn from sludge-treated soils.

INTRODUCTION

Trace elements are found in a variety of physicochemical forms, including those that are free or as complexed ions in soil solution; adsorbed at the surfaces of clays, Fe and Mn oxyhydroxides, or organic matter that are easily exchangeable; present in the lattice of secondary minerals such as phosphates, sulfides, or carbonates; occluded in amorphous materials such as Fe and Mn oxyhydroxides, Fe sulfides, or organic matter; and present in the crystal lattices of primary minerals (Lake et al., 1984; Tessier and Campbell, 1988). Sequential extraction methods have been used to fractionate the various forms of trace metals in soils, sediments, sludges, and dissolved solids in natural waters. These fractionation schemes are based both theoretically and experimentally on more than 100 years of research (Jackson, 1985) in many different scientific disciplines, such as soil science, geology, and chemistry.

A basic requirement of any selective extraction procedure should be to solubilize specific components of a soil or sediment (Chao, 1984). In much of the research literature, however, sequential fractionation techniques were employed with little or no reported explanation of their appropriateness or lack thereof, other than their utilization by other scientists. This may give one the sense that fractionation techniques have been fully elucidated, segregating various portions of soils or sediments into the desired fractions. In reality, the techniques are still controversial and uncertainty of the true fraction dissolved still exists.

No fractionation scheme is totally effective in dissolving each distinct form of a trace element. An extracting solution may dissolve less of the target fraction and more of a non-target fraction than desired, for example. In addition, when a fraction dissolves, metals in that portion may not remain in solution. Rather, the metals may resorb onto another fraction or can precipitate. The extraction procedure itself may cause a shift in elemental distribution so that the solids remaining after the extraction, rather than the reagent used, determine the trace element concentrations found in the extracting solution (Tessier and Campbell, 1988).

Nonetheless, each step in many of the schemes proposed will operationally recover a different soil fraction than either the step before or after it. However, one should not assume that each succeeding fraction will be less environmentally available than the previous one. Some of the extracting solutions appear to be highly selective for a particular soil fraction (e.g., NaOCl for organic matter), regardless of the order in which it is used.

Many different methods have been employed to fractionate trace elements depending on the composition of the substrate and the portions of the substrate considered most important. Several are listed in Table 1. Reviews of fractionation methods used to determine chemical forms of trace elements in natural waters (Florence and Batley, 1977), soils and sediments (Pickering, 1981), geochemical exploration (Chao, 1984), and sewage sludge and sludge-amended soils (Lake et al., 1984) have been written. Chao (1984) concluded that these techniques can be expected to

Table 1. Examples of sequential extraction procedures.

I.	McLaren and Crawford, 1973	
	1. Soil solution and exchangeable	0.05*M* $CaCl_2$
	2. Specifically sorbed	2.5% acetic acid
	3. Organic	1.0*M* $K_4P_2O_7$
	4. Free oxides	0.175*M* $(NH_4)_2C_2O_4$ + 0.1*M* $H_2C_2O_4$
	5. Residual	HF
II.	Gupta and Chen, 1975	
	1. Interstitial water	Squeezed through 0.05 μ filter
	2. Soluble	Deaerated double distilled water
	3. Exchangeable	1*M* NH_4OAc (deaerated)
	4. Carbonate	1*M* HOAc
	5. Mn and amorphous Fe oxides	0.1*M* $NH_2OH\cdot HCl$ + 0.01*M* HNO_3
	6. Organic/sulfide	30% H_2O_2 @ 85oC
	7. Fe oxide (moderately reducible)	Sodium dithionite-citrate
	8. Residual	HNO_3/HF/$HClO_4$
III.	Stover et al., 1976	
	1. Exchangeable	1*M* KNO_3
	2. Sorbed	0.5*M* KF @ pH 6.5
	3. Organic	0.1*M* $Na_4P_2O_7$
	4. Carbonate	0.1*M* EDTA @ pH 6.5
	5. Sulfide	1*M* HNO_3
IV.	Tessier et al., 1979	
	1. Exchangeable	1*M* $MgCl_2$ @ pH 7.0
	2. Carbonates	NaOAc/HOAc @ pH 5.0
	3. Fe & Mn Oxides	$NH_2OH\cdot HCl$ in 25% HOAc @ pH~2
	4. Organic Matter	H_2O_2/HNO_3 @ pH~2
	5. Residual	HF and $HClO_4$
V.	Grove and Ellis, 1980	
	1. Water-soluble	D.I. Water
	2. Exchangeable	1*M* NH_4Cl
	3. Organically complexed	0.1*M* $CuSO_4$
	4. Amorphous precipitate	0.3*M* $(NH_4)_2C_2O_4$
	5. Crystalline precipitate	Na_3citrate/Na_2dithionite/ Na bicarbonate
	6. Total	HNO_3/$HClO_4$/HF
VI.	Emmerich et at., 1982	
	1. Exchangeable	0.5*M* KNO_3
	2. Adsorbed	Ion exchange water, extracted three times
	3. Organically bound	0.5*M* NaOH
	4. Carbonate	0.05*M* Na_2EDTA
	5. Residual	4.0*M* HNO_3

Table 1 (cont'd)

VI.	Shuman, 1985	
	1. Exchangeable	1*M* $Mg(NO_3)_2$ @ pH 7
	2. Organic	0.7*M* NaOCl @ pH 8.5
	3. Mn Oxides	0.1*M* $NH_2OH \cdot HCl$ + 0.01*M* HNO_3 @ pH 2
	4. Amorphous Fe Oxides	0.2*M* $(NH_4)_2$ $C_2O_4 \cdot H_2O$-0.2*M* $H_2C_2O_4$ @ pH 3
	5. Crystalline Fe Oxides	Amorphous Fe oxide solution plus 0.1*M* ascorbic acid
	6. Residual	$HF/HNO_3/HCl$ sequentially
VIII.	Miller et. al., 1986	
	1. Soluble	H_2O
	2. Exchangeable	0.5*M* $Ca(NO_3)_2$ @ pH 7
	3. Pb-displaceable	0.05*M* $Pb(NO_3)_2$ + 0.1*M* $Ca(NO_3)_2$
	4. Acid-soluble	0.44*M* CH_3COOH + 0.1*M* $Ca(NO_3)_2$
	5. Mn Oxide	0.1*M* $NH_2OH \cdot HCl$ + 0.01*M* HNO_3 @ pH 2
	6. Organically bound	0.1*M* $K_4P_2O_7$
	7. Amorphous Fe oxide	0.175*M* $(NH_4)_2C_2O_4$ + 0.1*M* $H_2C_2O_4$
	8. Crystalline Fe oxide	Amorphous Fe oxide solution plus UV radiation
	9. Residual	$HCl/HNO_3/HF$
IX.	Elliott et. al., 1990	
	1. Exchangeable	1*M* $MgCl_2$ @ pH 7.0
	2. Dilute acid-extractable	1*M* NaOAc @ pH 5
	3. Fe-Mn oxide bound	0.175*M* $(NH_4)_2C_2O_4$ and 0.1*M* $H_2C_2O_4$
	4. Organically bound	0.1*M* $Na_4P_2O_7$
X.	Sims and Kline, 1991	
	1. Exchangeable	0.5*M* KNO_3
	2. Sorbed	H_2O
	3. Organic	0.5*M* NaOH
	4. Carbonate	0.05*M* Na_2EDTA
	5. Sulfide	4*M* HNO_3

play a prominent role in the search for concealed ore bodies. Other reviewers gave less than sterling grades to these techniques. Pickering (1981) cautioned that the "careless" use of chemical fractionation techniques will result in the generation of erroneous or misleading data when their pitfalls and limitations are not appreciated. Lake et al. (1984) concluded that chemical extraction did not represent an analytical method of speciation (whether they meant "separation" or chemical speciation is not clear from their discussion). Rather, they suggested that the physical separation of trace metals tend to have less effect on the inherent speciation of a metal, and hence present an attractive approach for complex matrices.

Although Lake et al. (1984) presented no information detailing a physical separation technique, Shuman (1979, 1985) separated soils into sand, silt, and clay-sized fractions and determined total trace elements in each of these physical fractions. However, he did not relate the results to potential bioavailability of the trace elements.

In general, separation techniques have been developed and used to dissolve about six different chemical fractions: 1) water-soluble, 2) exchangeable, 3) carbonates, 4) organic, 5) sesquioxides, and 6) residual clay minerals not solubilized in first five. The following discussion attempts to review each of these fractions, including the separation techniques utilized.

Water-soluble Fraction

Trace elements, as defined by Mattigod et al. (1981), typically are present in the dissolved phase in soil solutions at concentrations less than 10^{-4}*M*. They can form soluble complexes with both organic and inorganic ligands that are present in the soil solution. In addition, at any time in a soil system that trace elemental solids are present, the concentration of trace elements in the solution phase is influenced by the rate of dissolution of unstable solid phases and the rate of precipitation of stable and metastable solid phases (Mattigod et al., 1981).

A water-soluble fraction should result in a measure of trace elements that are a portion of the so called "labile" pool, which consists of dissolved elements plus an amount of surface-bound elements that are in "reasonably" rapid equilibrium with the dissolved elements (Corey, 1990). In soils, trace elements can occur in a variety of solid phases. The labile fraction, however, would not include elements present as precipitates except for those at the surface of precipitates and sesquioxides that can react rapidly enough to establish an apparent equilibrium with the soil solution. Nor would it not include organic forms mineralized or immobilized by microorganisms (Corey, 1990) and inorganic forms occluded or coprecipitated with sesquioxides, or incorporated into the crystalline structure of clay minerals due to isomorphic substitution.

Ions occurring in the water extract as a result of dissolution of solid phases, especially those in relatively high concentrations (such as H^+, OH^-, Na^+, Ca^{2+}, and Mg^{2+}), can exchange for ions occupying charged sites on clays, organic matter, and hydrous oxides. Exchange reactions in the water-soluble fraction may result in greater concentrations of trace elements in solution. Trace elements that dissolve from the solid phase may subsequently exchange with other elements, reducing their own concentration in solution while increasing that of others. Additionally, they may form adducts (Sposito, 1981), affecting their activity but not necessarily their analytical concentrations in solution. A detailed discussion of macroscopic (thermodynamic) and microscopic ion exchange reactions follows in the section on the exchangeable fraction.

The significance of the water-soluble fraction is that it should represent those trace elements that are the most environmentally available. Compared to the other fractions, however, one would not expect large quantities of trace element in this fraction unless it is measured soon after the addition of soluble forms of the elements.

Cadmium, Cu, and Zn may occur in soils as oxides, hydroxides, and sulfates (Lindsay, 1979); Ni as hydroxides and carbonates (Kotrlý and Šůcha, 1985); and Pb as oxides and silicates (Lindsay, 1979). These solid phases, if present, may dissolve to produce measurable amounts of these five trace elements in a soil-water extract. Table 2 lists possible equilibrium reactions of Cd, Cu, Ni, Pb, and Zn containing minerals. Chromium can occur in soils as chromite ($FeCr_2O_4$) (Reisenauer, 1982) or chromic oxide (Cr_2O_3) (NAS, 1974), neither of which are soluble in water (CRC Press, 1987; and Merck, 1989).

Exchangeable Fraction

The cation exchange reaction between two cations A^{u+} and B^{v+} substituting on the exchange complex X^{-1}, is represented in Equation (1) (Sposito, 1981):

$$vAX_u(s) + uBCl_v(aq) \rightarrow uBX_v(s) + vACl_u(aq) \tag{1}$$

where Cl^- balances the cationic charge in the aqueous solution phase. The equilibrium constant that describes the exchange reaction in Equation (1) is:

$$K_{ex} = \frac{(BX_v)^u(ACl_u)^v}{(AX_u)^v(BCl_v)^u} \tag{2}$$

Parentheses in Equation (2) represent activities.

Equation (3) represents the anion exchange reaction and Equation (4) is the equilibrium constant that describes the anion exchange

Table 2. Equilibrium reactions† of Cd, Cu, Ni, Pb, and Zn minerals at 25°C.

Equilibrium Reaction	log K°
Cadmium	
$CdO + 2H^{+} \rightleftharpoons Cd^{2+} + H_2O$	15.14
$\beta\text{-}Cd(OH)_2(c) + 2H^{+} \rightleftharpoons Cd^{2+} + 2H_2O$	13.65
$CdSO_4 \cdot 2Cd(OH)_2(c) + 2H^{+} \rightleftharpoons 3Cd^{2+} + SO^{4-} + 4H_2O$	22.65
$CdCO_3(\text{octavite}) + 2H^{+} \rightleftharpoons Cd^{2+} + CO_2 + H_2O$	6.16
Copper	
$CuO(\text{tenorite}) + 2H^{+} \rightleftharpoons Cu^{2+} + H_2O$	7.66
$Cu(OH)_2(c) + 2H^{+} \rightleftharpoons Cu^{2+} + 2H_2O$	8.68
$CuCO_3(c) + 2H^{+} \rightleftharpoons Cu^{2+} + CO_2(g) + 2H_2O$	8.52
$CuSO_4(\text{chalcocyanite}) \rightleftharpoons Cu^{2+} + SO_4^{2-}$	3.72
$CuSO_4 \cdot 5H_2O(c) \rightleftharpoons Cu^{2+} + SO_4^{2-} + 5H_2O$	-2.61
$CuO \cdot CuSO_4(c) + 2H^{+} \rightleftharpoons 3Cu^{2+} + SO_4^{2-} + H_2O$	11.50
$Cu_2CO_3(c) + 2H^{+} \rightleftharpoons Cu^{2+} + CO_2(g) + H_2O$	8.52
Nickel	
$Ni(OH)_2(s) + 2H^{+} \rightleftharpoons Ni^{2+} + 2H_2O$	12.80
$NiCO_3(s) + 2H^{+} \rightleftharpoons Ni^{2+}\ CO_2 + H_2O$	11.28
Lead	
$PbO(\text{yellow}) + 2H^{+} \rightleftharpoons Pb^{2+} + H_2O$	12.89
$PbO(\text{red}) + 2H^{+} \rightleftharpoons Pb^{2+} + H_2O$	12.72
$Pb_3O_4(c) + 8H^{+} + 2e^{-} \rightleftharpoons 3Pb^{2+} + 4H_2O$	73.79
$PbO_2(c) + 4H^{+} + 2e^{-} \rightleftharpoons Pb^{2+} + 2H_2O$	49.68
$Pb_2SiO_4(c) + 4H^{+} \rightleftharpoons 2Pb^{2+} + H_4SiO_4$	18.45
$PbCO_3(\text{cerussite}) + 2H^{+} \rightleftharpoons Pb^{2+} + CO_2(g) + H_2O$	4.65
Zinc	
$Zn(OH)_2(\text{amorp}) + 2H^{+} \rightleftharpoons Zn^{2+} + 2H_2O$	12.48
$\alpha\text{-}Zn(OH)_2(\text{amorp}) + 2H^{+} \rightleftharpoons Zn^{2+} + 2H_2O$	12.19
$\beta\text{-}Zn(OH)_2(\text{amorp}) + 2H^{+} \rightleftharpoons Zn^{2+} + 2H_2O$	11.78
$\gamma\text{-}Zn(OH)_2(\text{amorp}) + 2H^{+} \rightleftharpoons Zn^{2+} + 2H_2O$	11.74
$\epsilon\text{-}Zn(OH)_2(\text{amorp}) + 2H^{+} \rightleftharpoons Zn^{2+} + 2H_2O$	11.53
$ZnO(\text{zincite}) + 2H^{+} \rightleftharpoons Zn^{2+} + H_2O$	11.16
$ZnSO_4(\text{zinkosite}) \rightleftharpoons Zn^{2+} + SO_4^{2-}$	3.41
$ZnO \cdot 2ZnSO_4(c) + 2H^{+} \rightleftharpoons 3Zn^{2+} + 2SO_4^{2-} + H_2O$	19.12
$ZnCO_3(\text{smithsonite}) + 2H^{+} \rightleftharpoons Zn^{2+} + CO_2(g) + H_2O$	7.91

†From Lindsay, 1979 and Kotrlý and Šůcha, 1985.

reaction (Sposito, 1981):

$$wCY_x(s) + xNa_wD(aq) \rightarrow xDY_w(s) + wNa_xC(aq) \tag{3}$$

$$K_{ex} = \frac{(DY_w)^x(Na_xC)_w}{(CY_x)^w(Na_wD)^x} \tag{4}$$

In these equations, C^{x-} and D^{w-} are the two anions exchanging on Y^+. Y^+ represents one equivalent of the anion exchange complex. The cation balancing the anionic charge in this case is Na^+. It follows from Equations (1) and (2) that in order to maximize the exchange of B^{v+} for A^{u+}, the cation of interest, the (BCl_v) can be increased by utilizing an exchange solution with high concentration, (AX_u) can be increased by using more exchange complex (increasing the amount of soil in the procedure), and increasing "v", the valence on the cation B^{v+}. The same general statements apply to the anion exchange reaction in order to maximize the exchange of D^{w-} for C^{x-}, the anion of interest. The concentration of $Na_wD(aq)$, $CY_x(s)$, and the charge of D^{w-} can be increased.

The choice of a solution to supply ions that can substitute for the cations and anions occupying surface sites would be simple if thermodynamic theory alone governed ion exchange. However, trace elements have different affinities for surface sites at a molecular level, resulting in a plethora of interactions between ions and exchange sites that ensures difficulty in selecting a single solution to work as well as Equations (1) to (4) may suggest. This is apparent from Table 1, which shows the various solutions and the combinations of cations and anions at different molar concentrations that researchers have used to extract the exchangeable fraction.

The concept of Hard and Soft Acids and Bases (HSAB) (Sposito, 1981) is an appropriate starting point in a discussion of ion-exchange site interactions. According to this concept, any chemical species that can accept electrons in a chemical reaction is a Lewis acid and one that can donate electrons is a Lewis base. Furthermore, these two groups are subdivided into hard, borderline, and soft Lewis acids and bases (Pearson, 1963). The HSAB theory states that hard acids and hard bases prefer to coordinate to one another while soft acids and soft bases exhibit an affinity for each other. The degree to which a metal cation displays these characteristics is thought to be proportional to the Misono softness parameter, Y (Misono et al., 1967):

$$Y = \frac{(10I_z r)}{(I_{z+1} z^{1/2})} \tag{5}$$

in which the electronegativity of the element, I_z, is the zth ionization potential of the cation, z is the charge on the cation, and r is the ionic radius. For a given oxidation state, as the ionic radius increases, the polarizability and the degree of softness increases.

Five of the six trace elements of interest occur most commonly in soils as the cationic species Cd^{2+}, Cu^{2+}, Ni^{2+}, Pb^{2+}, and Zn^{2+} (Bohn et al., 1979; Norvell, 1972). Chromium is most commonly present as the anionic species, CrO_4^{2-} (Bohn et al., 1979), but can also occur as Cr^{3+}, CrO_2^-, and $Cr_2O_7^{2-}$ at values of *pe* (-log of electron activity) and pH found in soils (Bartlett and Kimble, 1976a). The cations may form inner-sphere or outer-sphere complexes with surface functional groups on hydrous metal oxides, siloxane surfaces, and organic matter. They may also accumulate in the diffuse ion swarm or may be adsorbed as metal-ligand complexes. Generally speaking, Cd^{2+} is considered a soft Lewis acid and Cu^{2+}, Ni^{2+}, Pb^{2+} and Zn^{2+} are grouped as borderline Lewis acids. Misono softness parameters and values used to calculate them are listed in Table 3.

Magnesium ions (Mg^{2+}) and Ca^{2+}, which are hard Lewis acids (Sposito, 1981), are among the cations often used to extract the exchangeable fraction of trace elements in soils (Table 1). Calcium, however, is softer than Mg, having a Misono softness parameter more similar to those of Cd, Cr, Cu, Ni, Pb, and Zn. Therefore, Ca would be the better choice for extracting this fraction for this group of trace elements. Other techniques to determine an exchangeable fraction in soil include the use of monovalent cations such as K^+ and NH_4^+ (Table 1). These are less satisfactory than divalent cations from a thermodynamic perspective. However, K has a softness parameter more similar to the trace elements of interest than does Ca. Better choices of exchange cations based on their softness include Fe^{2+} and Mn^{2+}. Miller et al. (1986) used this idea in the selection of Pb^{2+} in 0.05*M* $Pb(NO_3)_2$ to determine Cu, Fe, and Mn in a lead-displaceable fraction. Our desire to measure Pb in this and subsequent fractions precludes its use in an extracting solution.

The counter anions in solutions used to determine an exchangeable fraction have consisted primarily of NO_3^- and Cl^- (Table 1). From HSAB theory, NO_3^- is a hard Lewis base and Cl^- is a borderline Lewis base (Sposito, 1981). Consequently, one would expect Cl^- to more easily coordinate with the trace elements, which are borderline Lewis acids, than NO_3^-. Shuman (1985) modified his procedure from a solution of $MgCl_2$ to $Mg(NO_3)_2$ because of the work of Doner (1978) showing that the chloride

Table 3. Misono softness parameters for metal cations in aqueous solutions.

Ion^{z+}	Ionization Potential z	z+1	Ionic radii	Misono Softness Parameter
	- kJ mol^{-1} -		- nm -	
H^{+}	1312	NA†	0.154	NA
Ag^{+}	731	2073	0.126	0.444
Cs^{+}	375	2229	0.167	0.281
Cu^{+}	745	1958	0.096	0.365
K^{+}	419	3052	0.133	0.183
Li^{+}	520	7298	0.074	0.053
Na^{+}	496	4563	0.097	0.105
Rb^{+}	403	2632	0.147	0.225
Tl^{+}	589	1971	0.147	0.439
Ba^{2+}	965	NA	0.134	NA
Be^{2+}	1757	14848	0.035	0.029
Ca^{2+}	1145	4912	0.100	0.165
Cd^{2+}	1631	3616	0.097	0.309
Co^{2+}	1646	3232	0.072	0.259
Cu^{2+}	745	1958	0.073	0.196
Fe^{2+}	1561	2957	0.074	0.276
Mg^{2+}	1451	7732	0.072	0.096
Mn^{2+}	717	1509	0.080	0.269
Ni^{2+}	1753	3393	0.069	0.252
Pb^{2+}	1450	3081	0.120	0.399
Ra^{2+}	979	NA	0.143	NA
Sr^{2+}	1064	4207	0.112	0.200
Zn^{2+}	1733	3833	0.074	0.237
Al^{3+}	2745	11577	0.051	0.070
Co^{3+}	3232	4950	0.072	0.271
Cr^{3+}	2987	4737	0.063	0.229
Fe^{3+}	2957	5287	0.064	0.207
Ga^{3+}	2963	6175	0.062	0.172
In^{3+}	2704	5210	0.081	0.243

†NA = information not available

ion can complex metals. The soluble complexes formed between Ni^{2+}, Cu^{2+}, and Cd^{2+} resulted in greater mobility of the metals in a Cl^{-} solution than in a ClO_4^{-} solution (Doner, 1978). The soluble complexes that may form between Cl^{-} and the trace elements appeared to reduce resorption of the trace elements onto surfaces. In addition, soluble complex formation should not affect the measurable concentration of the trace elements in solution if Direct Current Plasma-Atomic Emission Spectrometry (DCP-AES) or atomic absorption analytical techniques are used. Doner (1978) did not address the effects of a NO_3^- salt solution on the formation of complexes. The work by Doner (1978), therefore, did not fully explain Shuman's (1985) decision to switch from Cl^- to NO_3^-.

Another concern in the selection of a solution used to determine an exchangeable fraction is the choice of an anion to displace anionic forms of Cr from positively charged surface sites. The work of Eary and Rai (1991) demonstrated that SO_4^{2-} and $H_2PO_4^-$ displaced $HCrO_4^-$ more effectively from positively charged sorption sites than Cl^- and ClO_4^-. Sulfate, PO_4^{3-}, and NO_3^- are considered hard Lewis bases (Sposito, 1981). This would infer that NO_3^- may also be more effective at displacing $HCrO_4^-$ than Cl^-. Sulfate and PO_4^{3-} may not be as acceptable as NO_3^- however, because they are more likely to form precipitates with Cd, Cu, Ni, Pb, and Zn (Table 2). Specific information on the apparent hardness or softness of anionic Cr species is not available.

Previous work cited by Tessier et al. (1979) and some of their own results indicated that NH_4OAc at pH 7 and NaOAc at pH 8.2, which have been used frequently to extract exchangeable elements, also appeared to attack carbonates. Based on this, a salt of the acetate anion should not be used to remove the exchangeable fraction when a carbonate fraction also is desired.

Carbonate Fraction

Three of the five fractionation techniques cited in Table 1 used a solution containing EDTA to extract carbonates. The work of Stover et al. (1976) demonstrated that oxalate, citrate, and NH_2OH resulted in inconsistent and incomplete recovery of metals from both carbonate and sulfide precipitates and considered them unsatisfactory in a sequential fractionation procedure. However, these researchers found that EDTA removed greater than 91% of Pb, Zn, and Cu carbonates and 68% of $CdCO_3$. Additionally, EDTA recovered less than 10% of Cd, Cu, and Zn sulfides and 29% of PbS.

On the basis of research done by others, Tessier et al. (1979) chose 1*M* NaOAc at pH 5 to remove metal carbonates from sediment. This technique was adapted from the work of Grossman and Millet (1961) who recommended 1*M* NaOAc at pH 5 to dissolve carbonates in the pretreatment of soils for mineralogical analysis. They reported that the NaOAc did not affect particle size, CEC, organic C, N, or free Fe values in carbonate free soils (Grossman and Millet, 1961).

Gupta and Chen (1975), in a technique adapted from the work of Chester and Hughes (1967), utilized 1*M* acetic acid as an extractant. Carbonate and some Fe and Mn oxides were considered the likely geochemical phases solubilized by the 1*M* acetic acid solution (no data presented). Miller et al. (1986) used a 0.44*M* CH_3COOH [2.6% (w/v) acetic acid] solution at pH 2.5 to determine an "acid-soluble" fraction. They hypothesized that the protons (hard Lewis acid) supplied by the acetic acid desorb inner-sphere complexed elements. It is unlikely, however, that this technique is completely successful in removing only specifically bound elements from soil surfaces. In addition, the low pH may solubilize previously solid mineral forms of the trace metals. (See Table 2 for K° values of sparingly soluble mineral forms of the trace elements.)

A 2.5% acetic acid solution also was used by McLaren and Crawford (1973) to remove Cu which was thought to be bound mainly by inorganic sites on oxides and clay minerals. Reducing the amount of soil extracted with the acetic acid to less than 5 g considerably increased the proportion of Cu removed (no data presented). This was attributed to greater solution of oxide material and to greater desorption of Cu from organic sites. However, 20-g samples were considered more appropriate to provide a reliable estimate of the predominantly inorganically bound Cu. The results of the correlation tests comparing the fractionation of Cu to various soil properties showed that the Cu extracted with acetic acid was not significantly correlated with percent organic C, percent clay, pH, or free Mn and Fe oxides.

McLaren and Crawford (1973) concluded that the acid-soluble fraction "is probably best considered as originating from more weakly binding specific sites (specific sorption) on all types of constituents, both organic and inorganic." Whether this acid-soluble fraction extracts Cd, Cr, Ni, Pb, and Zn, as it is thought to extract Cu, was not specifically addressed by either Miller et al. (1986) or McLaren and Crawford (1973). Interesting enough, McLaren and Crawford (1973) and Miller et al. (1986) made no comments in their reports as to the appropriateness of acetic acid to solubilize the carbonate fraction, probably because carbonates were not of particular concern in their soils.

The desire to use acetic acid as a selective extracting solution appeared to have its beginnings in the work of Chester and Hughes (1967) who were searching for a technique to dissolve the Fe and Mn oxide phases of a ferro-manganese nodule in pelagic sediments. They reported on the work of Ray et al. (1957) who pointed out that dilute acetic acid dissolved the carbonates of most rocks, excluding dolomite, but will not attack lattice structures of clay minerals. Hirst and Nicholls (1958) used 25% (v/v) acetic acid to separate detrital and non-detrital fractions of marine sediments from carbonate rocks. Hodgson (1960) demonstrated that 2.5% (v/v) acetic acid strips the exchangeable fraction of adsorbed Co ions from the surface of the clay mineral montmorillonite. From their own work, Chester and Hughes (1967) discovered that 25% (v/v) acetic acid will partially dissolve Fe oxide and practically leave untouched Mn oxide of ferro-manganese nodules.

In our soils, carbonates also are not expected to represent an appreciable fraction in which the trace elements would naturally reside. However, it is possible that sludge applied to the soil contains carbonates used for stabilization. Therefore, it may be appropriate to at least consider a carbonate fraction. Both acetic acid and EDTA solutions appear to be valid techniques for selectively extracting the carbonate fraction. As for the "acid-soluble" fraction determined in the work of Miller et al. (1986) and McLaren and Crawford (1973), their technique hypothesizes a soil fraction that they would like to extract. Their data were inconclusive in the success of the procedure for solubilizing a "specifically adsorbed" or "weakly adsorbed" fraction. In reality, the acidic acid probably dissolves mineral forms of the trace elements, including carbonates, in addition to those associated with sesquioxides and organic matter.

Organic Fraction

Several different solutions have been used by researchers to fractionate the organic portion of soils and sediments from the mineral fractions (Table 1). Solutions include H_2O_2/HNO_3, NaOH, $CuSO_4$, NaOCl, and $K_4P_2O_7$ in various aqueous concentrations. One-tenth *M* $CuSO_4$ was used by Grove and Ellis (1980) to remove organically complexed forms of added Cr, Fe, and Mn; specifically, trace elements bound to carboxyl groups in the organic matter would exchange with the Cu^{2+} of the $CuSO_4$ solution. Bartlett and Kimble (1976a, 1976b) reported that $Na_4P_2O_7$, NH_4OAc @ pH 4.8, and 0.1*M* NaF also can be used to extract organically bound Cr.

Research by Anderson (1963) compared the effectiveness of a 5 to 6% solution of NaOCl at pH 9.5 with H_2O_2 and H_2O_2 at pH 5. His data showed that the NaOCl solution extracted as much or more organic matter in soil samples from Indiana and Michigan than the other two techniques. X-ray diffraction analysis after extraction with NaOCl indicated that sesquioxides, silica coatings, and crystalline clay components were still intact. Lavkulick and Wiens (1970) attributed the effectiveness of a NaOCl solution over H_2O_2 to its greater electrode potential, resulting in a more powerful oxidizing agent. Adjusting the pH of the NaOCl solution from 9.5 to 4.5 increased the electrode potential and the amount of Fe in the extracts and increased the carbon remaining in the residue after treatment (Lavkulick and Wiens, 1970). They concluded that three successive NaOCl treatments removed up to 98% of the oxidizable soil carbon. For mineralogical or particle size investigations, soil samples receiving this treatment are largely free from organic cementing agents, Na-saturated, and in the dispersed state.

Shuman (1983) also examined the usefulness of 5.3% NaOCl [about the same concentration of NaOCl used by Lavkulick and Weins (1970) and Anderson (1963)] to remove trace elements associated with soil organic matter. Based on preliminary experiments (data not reported), Shuman (1983) used NaOCl at pH 8.5 because low amounts of Zn were found in the extracts at pH 9.5. He reasoned that solution pH of 9.5 may induce precipitation of the released metals, causing them to remain in the soils.

Shuman (1983), however, did not present a strong case for lowering the pH of NaOCl to 8.5 (basing it only on Zn concentrations in the extracts). The data reported by Lavkulick and Wiens (1970) demonstrated the negative effects of decreasing pH, i.e., leaving a higher C content in the residue after treatment and obtaining higher Fe concentrations in the resulting extracts. Anderson (1963) was concerned with his decision to adjust NaOCl to pH 9.5. When NaOCl is titrated with HCl, pH 9.5 is an inflection point at which a small increase or decrease in the amount of acid added resulted in a large change in solution pH. Additionally, he showed that NaOCl at pH 8.5 has a greater oxidation potential ($E_{ox} \approx$ -1100 mV) than at pH 9.5 ($E_{ox} \approx$ -1010 mV) (Anderson, 1963). Consequently, Shuman's (1983) decision to buffer his NaOCl solution at pH 8.5 rather than 9.5 may be correct.

Comparisons Shuman (1983) made with H_2O_2 and $Na_4P_2O_7$ indicated that 5.3% NaOCl at pH 8.5 extracted less Mn and Fe than the other two methods, but generally extracted more Cu and Zn than H_2O_2 and less Cu and Zn than $Na_4P_2O_7$. Shuman (1983) concluded that two extractions with NaOCl were necessary to completely dissolve the organic fraction in soils with organic matter content ranging from 1.0 to 2.8%. No significant amounts of trace elements were dissolved from the soils after two treatments. Increasing the time for extraction from 15 to 30 minutes dissolved significantly more Mn and Zn but had less effect on Cu and Fe (Shuman, 1983).

Other techniques used to solubilize organic matter from soils and sediments have included strong bases such as NaOH and Na_2CO_3, neutral salts including NaF and $K_4P_2O_7$, and organic acid salts. Extraction with these solutions generally recovers 80% or less of the soil organic matter, and the strong bases also can have undesirable effects on other soil components, e.g., dissolving silica and other mineral fractions (Stevenson, 1982). Potassium pyrophosphate ($K_4P_2O_7$) reportedly dissolves Fe oxides (Bascomb, 1968) and Mn oxides (McLaren and Crawford, 1973). Sodium pyrophosphate, however, solubilized less than 30% of metal carbonates and sulfides (Stover et al., 1976). Of the methods used to remove organic matter from soils, multiple extractions using 0.7*M* NaOCl at pH 8.5 or 9.5 appears to be the most effective for removing organic matter while not dissolving appreciable amounts of other mineral components in the soil.

Mineral Fractions

Free Fe-Al oxides and hydroxides, occurring as discrete particles or coatings, can be extracted from soils using a sodium citrate dihydrate/$NaHCO_3$/$Na_2S_2O_4$ solution. The solution works well for this purpose because it fulfills a basic requirement for removal of free Fe hydrous oxides and hydroxides: it has a high oxidation potential

($Na_2S_2O_4$ is a good reducing agent). Also, it contains sodium citrate to act as a chelating agent for isolating Fe^{2+} and Fe^{3+} and is buffered at pH 7.3 by $NaHCO_3$ (Jackson, 1985).

Shuman (1982), however, found two major faults with this method. The $Na_2S_2O_4$ is often contaminated with Zn and can form sulfides that precipitate metals. Consequently, he conducted an experiment to find an acceptable alternative for dithionite to solubilize crystalline Fe oxides. His objective was to develop a sequential extraction scheme to solubilize Mn oxides, noncrystalline (amorphous) Fe oxides, and crystalline Fe oxides. In his study he examined 0.1*M* $Na_4P_2O_7$ at pH 10, 0.2*M* $(NH_4)_2C_2O_4$ in 0.2*M* $H_2C_2O_4$ at pH 3 (oxalate), 0.1*M* $NH_2OH \cdot HCl$ at pH 2, 1.0*M* $NH_2OH \cdot HCl$ in 25% acetic acid, 0.1*M* ascorbic acid in the oxalate solution, 0.1 g $SnCl_2$ per gram of soil in the oxalate solution, and 1.0 g dithionite per gram of soil in citrate buffer (Shuman, 1982).

These were solutions others had reported useful when solubilizing oxides (Chao, 1972; McKeague and Day, 1966; McKeague et al., 1971; and McLaren and Crawford, 1973). The $Na_4P_2O_7$ solution, often used to extract the organic fraction, extracted amounts of Fe similar to that of the oxalate solution and appreciable amounts of Mn and Al. The two $NH_2OH \cdot HCl$ solutions solubilized little Fe, but $NH_2OH \cdot HCl$ without acetic acid solubilized as much Mn as most of the other extractants indicating that it was specific for Mn oxides. Of the noncrystalline Fe-oxide extractants, the oxalate solution solubilized the most Zn and Cu. Ascorbic acid-oxalate solubilized the greatest amounts of Zn and Cu of the crystalline Fe oxide extractants.

Shuman (1983) rejected the method of using oxalate with UV light to solubilize crystalline Fe oxides because he felt it would be difficult to standardize the technique from one laboratory to the next. In order to find a chemical reductant to replace the UV light he tried both ascorbic acid and stannous chloride, both common reductants in chemical procedures. Both performed well and he concluded that either would be appropriate, except that the ascorbic acid-oxalate solution had fewer analytical difficulties when using atomic absorption spectrometry than the stannous chloride reductant. Therefore, he suggested the use of the $NH_2OH \cdot HCl$ solution for Mn oxides, oxalate solution for noncrystalline Fe oxides, and ascorbic acid-oxalate for crystalline Fe oxides. Finally, it should be noted that Shuman (1983) did not specify on what soil particle size these solutions were used, whereas in later work (Shuman, 1985), a 0.5 mm-mesh was used to screen the samples after grinding. Also, he did not indicate whether the soil samples were pretreated to remove other fractions, such as exchangeable and organic fractions prior to using these solutions to remove oxides from soil.

Residual Fraction and Total Elements

Once the other fractions have been extracted, the residual solids should contain mainly primary and secondary minerals, which may hold trace elements within their structure. These elements are not expected to become soluble (environmentally available) except over a relatively long (geologic) time span under natural weathering conditions.

Chromium, Cu, Ni, and Zn in the residual fraction present no special problems and can be dissolved with any of the techniques that utilize strong mineral acids. Cadmium and Pb offer distinct difficulties, since they can volatilize from the sample at high temperatures. The sample should not be heated to a temperature greater than 450°C for Cd (Baker and Amacher, 1982). No minimum temperature above which Pb would volatilize was noted by Burau (1982). Although he cites the work of others who heated samples to 450°C for Pb determination, he does not think heating samples even to this temperature is a particularly good idea. Additionally, the use of H_2SO_4 should be avoided when preparing samples to measure Pb because $PbSO_4$ precipitates can form (Burau, 1982).

The method outlined by Tessier et al. (1979) that used HF, $HClO_4$, and HCl in which the sample is not allowed to dry is relatively simple and also can be used for total trace element analysis. The method of Shuman (1979) may also be appropriate because, even though the solution is brought to dryness, the crucible is not heated above 120°C. However, the resulting solution is diluted by a factor of 50, whereas the method of Tessier et al. (1979) results in a solution with only a 25 fold dilution.

Sequential Extraction Techniques

No standard method of sequentially fractionating soils has been developed for polluted soils and sediments that completely separates specific soil fractions from one another. The works of Stover et al. (1976) and Tessier et al. (1979) have been used as starting points from which others have developed similar procedures. This has occurred not because they were the first to develop methods or because their techniques were better than ones already available, but probably because their work appeared in journals associated with pollution control and analytical chemistry, rather than a specific discipline such as geology or soil science.

More recently developed procedures, such as those of Shuman (1985) and Miller et al. (1986), utilize solutions that are appropriate for the particular fractions they attempt to isolate. Many of the solutions and techniques used in these two methods are similar. There are several interesting differences between these two methods, however, in the choice of extracting solutions, fractions to remove, the order in which the fractions are removed, differences in solution to soil ratio, shaking times, and particle size of the soil sample.

The work that Shuman (1979, 1982, 1983, 1985, 1988) has done to develop a fractionation scheme for Cu, Fe, Mn, and Zn is based both on a review of the literature and on his own laboratory experiments. His research has included various agricultural soils from the southeastern U.S., some of which have properties similar to soils in Michigan, i.e., sandy mineral soils that contain no carbonates or appreciable sulfides.

The technique developed by Shuman (1985) is outlined in Table 1. His first attempts at fractionating soils (Shuman, 1979) used $MgCl_2$ at pH 7 to remove exchangeable Zn, Mn, and Cu; 30% H_2O_2 to extract those micronutrients associated with organic matter; 0.2*M* ammonium oxalate and 0.2*M* oxalic acid, pH 3, for micronutrients associated with hydrous Fe oxides; and a multi-step procedure using $HCl/HNO_3/HF$ to dissolve residual micronutrients. In subsequent publications, he further refined his technique to remove Mn oxides using NH_2OH-HCl (Shuman, 1982); separated amorphous Fe oxides from crystalline Fe oxides using oxalate and ascorbic acid-oxalate solutions, respectively (Shuman, 1982); dissolved organic matter with sodium hypochlorite in order not to dissolve Mn and Fe oxides as can happen when using H_2O_2, sodium pyrophosphate (Shuman, 1983), or potassium pyrophosphate (Bascomb, 1968;

McLaren and Crawford, 1973); and substituted $Mg(NO_3)_2$ for $MgCl_2$ to remove exchangeable ions because, according to Doner (1978), Cl^- can complex metals (Shuman, 1985).

A weakness of this fractionation scheme is that Shuman had not specifically examined the ability of his method to extract several trace metals that are of environmental concern, namely Cd, Cr, Ni, and Pb. The reagents that he used, however, are ones that others have suggested when examining polluted soils (Elliott et al., 1990; Miller et al., 1986). Another problem in his technique may be the use of sodium hypochlorite for extracting trace elements associated with soil organic matter (Shuman 1983). This technique must be repeated at least twice to completely dissolve the organic fraction. He concludes from his own work that more than two extractions may be required for soils with greater than 3% organic matter. Additionally, NaOCl forms insoluble salts with Cr, Cd, Cu, and Zn unless it is acidified (personal observations).

A third concern of his technique is that the soils in his experiments contain concentrations of trace elements that one would "normally" find in soils of this type. Abnormally "high" concentrations of Cu, Mn, Zn and other trace elements were not a concern in his studies as they may be in research on polluted soils. Finally, Shuman (1979) determines residual and total elements using several mineral acids used in sequence. Each time the mixture is heated to dryness, there is a risk of losing Pb and Cd by volatilization at high temperature (Baker and Amacher, 1982; Burau, 1982).

Another sequential extraction procedure that holds promise for assessing trace element forms and environmental availability is that of

Miller et al. (1986). They used a nine step sequential extraction procedure to characterize three topsoils from southeastern U.S. (See Table 1 for an outline of this method.) Major differences between their technique and that of Shuman (1985) include the extraction of a water-soluble fraction, an exchangeable fraction using 0.5*M* $Ca(NO_3)$ at pH 7, Pb-displaceable and acid-soluble fractions, and a Mn oxide fraction extracted prior to the removal of organically bound metals using $K_4P_2O_7$. Also, the two schemes have differences in their soil-to-solution ratios. However, the six-step technique of Shuman (1985) sequentially extracts Mn oxide, noncrystalline (amorphous) Fe oxide, crystalline Fe oxide, and residue fractions utilizing similar extracting solutions and techniques to that of Miller et al. (1986), although there was a slight difference as to the order in which they were taken.

The objective of this study was to compare slightly modified methods of Shuman (1985) and Miller et al. (1986) to evaluate their applicability and usefulness to two Michigan soils, which had received high levels of trace elements via applications of municipal sewage sludge.

MATERIALS AND METHODS

Soil Samples

Three soils from experimental field plots, to which varying amounts of municipal sewage sludges were applied, were sequentially extracted using the modified techniques of Shuman (1985) and Miller et al. (1986). The first soil (Metea 1) is a composite sample from the 15 to 30-cm depth of a Metea sandy loam, 2 to 6% slope (loamy, mixed, mesic Typic Hapludalf) to which sludges had been surface-applied and incorporated over a ten-year period ("Campus" study). The second soil (Metea 2) is a surface sample of a Metea loamy sand, 2 to 6% slope, located in an experimental area which has had a more limited application of municipal sewage sludge than did Metea 1 ("St. Johns" study). The third soil (Capac) is a surface sample of a Capac loam, 0 to 3% slope (fine-loamy, mixed, mesic Aeric Ochraqualf). It was also from an experimental plot to which municipal sewage sludges have been applied ("W-170" study). These soil samples were chosen to examine the effectiveness of two sequential extraction techniques on soils with different levels of trace elements due to the application of sewage sludges.

Laboratory Analyses

Method 1 is a fractionation procedure that was modified from Shuman (1985). It was used to determine the concentrations of water-soluble, exchangeable, organic, Mn oxides, amorphous Fe oxides, crystalline Fe oxides, and residual forms of Cd, Cr, Cu, Ni, Pb, and Zn. Method 2 fractionation procedure was modified from Miller et al. (1986) and was used to determine the amounts of water-soluble, exchangeable, acid-soluble, Mn oxides, organic, amorphous Fe oxides, crystalline Fe oxides, and residual forms of Cd, Cr, Cu, Ni, Pb, and Zn.

Extractions, unless otherwise specified, were performed at laboratory room temperatures on a reciprocating shaker on which centrifuge bottles were placed on their sides, long axis perpendicular to the direction of shaker movement. The shaker speed was set to the minimum necessary to wash the walls of the centrifuge bottle, about 140 revolutions per minute (rpms). The solutions was centrifuged at about 5860 × g for 15 min. The supernatant was then decanted from the centrifuge bottle. Deionized water (Method 1) or 0.025*M* $Ca(NO_3)_2$ (Method 2) was used to wash the soil between extractions (except between Step 1 and 2 of the methods) to remove occluded solutions by shaking on a reciprocating shaker for 5 min and then centrifuging. Trace elements measured in the washings, if any, were added to the concentration of elements determined in the preceding extract. Unless specified, samples were not dried between steps. Solutions were stored at 4°C until analyzed. Trace element concentrations in centrifuged and digestion solutions were measured using DCP-AES.

Chemicals used to make the extracting solutions may be contaminated with trace elements. Extracting solutions were purified prior to use by passing them through a 50-g column of Chelex 100 (100 to 200 mesh) at a flow rate of about 5 mL min^{-1}. This purification procedure is time-consuming and will not improve solution quality when trace elements are not a contamination problem. In order to determine whether purification of an extracting solution was necessary, atomic emission spectra of a sample of the purified solution was compared to that of the unpurified solution. If there were no differences between the spectra of the solutions, purification was unnecessary.

<u>Fractionation Method 1</u> (modified from Shuman, 1985)

1. Water-soluble: Ten mL of deionized water (resistivity greater than 16.7 megohms) were added to a 5-g sample of air-dried 2-mm soil in a 50-mL centrifuge bottle and shaken for 16 h.

2. Exchangeable: Twenty mL of 1*M* $Mg(NO_3)_2$ (256.432 g $Mg(NO_3)_2 \cdot 6H_2O$ L^{-1} adjusted to pH 7 using $Mg(OH)_2$ or HNO_3) were added to the 5-g soil sample from the previous step and shaken for 2 h.
3. Organic matter: Ten g of 0.7*M* NaOCl (~5.2% NaOCl solution with 10.07% [w/v] Cl^- L^{-1}) adjusted to pH 8.5 immediately prior to use with 0.01*M* NaOH were added to the soil from Step 2. The bottles were placed in a boiling water bath for 30 min and the solutions were periodically shaken. This treatment was repeated four more times on the same sample with no washings between treatments. The resulting solutions for each soil were analyzed separately and the results were subsequently added together.
4. Mn oxides: The soil from Step 3 was air-dried, crushed, and passed through a 500-μm sieve. One gram of soil and 50 mL of 0.1*M* $NH_2OH \cdot HCl$ (hydroxylamine hydrochloride) solution prepared in 0.01*M* HNO_3 at pH 2 [6.949 g $NH_2OH \cdot HCl$ and 0.65 mL 69% HNO_3 (15.4*M*) diluted to 1 L. Note: $NH_2OH \cdot HCl$ is hygroscopic. Water can be removed by heating to 100°C.] were mixed and shaken for 30 min.
5. Noncrystalline (amorphous) Fe oxides: Fifty mL 0.2*M* $(NH_4)_2C_2O_4 \cdot H_2O$ (ammonium oxalate) - 0.2*M* $H_2C_2O_4 \cdot 2H_2O$ (oxalic acid) at pH 3 (28.42 g $(NH_4)_2C_2O_4 \cdot H_2O$ and 25.214 g $H_2C_2O_4 \cdot 2H_2O$ in 1 L) were added to the soil from Step 4 and shaken in the dark for 4 h.
6. Crystalline Fe oxides: Fifty mL 0.2*M* $(NH_4)_2C_2O_4 \cdot H_2O$ + 0.2*M* $H_2C_2O_4$ at pH 3 plus 0.1*M* $C_6H_8O_6$ (ascorbic acid; solution from step 5 plus 17.61 g $C_6H_8O_6$ in 1 L) were added to the soil from Step 5, placed in a boiling water bath for 30 min and periodically hand shaken.

7. Residual: Determined by using the total analysis method outlined below (Shuman, 1979) for the sample remaining from Step 6 after air drying, grinding, and passing through a 35 mesh sieve.

Fractionation Method 2 (modified from Miller et al., 1986)

1. Water-soluble: Twenty mL deionized water (resistivity greater than 16.7 megaohms) were added to a 0.5-g sample of air-dried 2-mm soil in a 50-mL centrifuge bottle and shaken for 16 h.
2. Exchangeable: Twenty mL 0.5*M* $Ca(NO_3)_2$ (118.08 g $Ca(NO_3)_2 \cdot 4H_2O$ L^{-1}) adjusted to pH 7 with CaO were added to the soil from Step 1 and shaken for 16 h.
3. Acid-soluble: Twenty mL 0.44*M* CH_3COOH + 0.1*M* $Ca(NO_3)_2$ (25.29 mL acetic acid + 23.61 $Ca(NO_3)_2 \cdot 4H_2O$ L^{-1}) were added to the soil from Step 2 and shaken for 8 h.
4. Mn oxide: Twenty mL 0.1*M* $NH_2OH \cdot HCl$ + 0.01*M* HNO_3 (6.949 g $NH_2OH \cdot HCl$ + 0.65 mL concentrated HNO_3 L^{-1}) were added to the soil from Step 3 and shaken for 30 min.
5. Organic matter: Twenty mL 0.1*M* $Na_4P_2O_7$ (44.607 g $Na_4P_2O_7$ L^{-1}) were added to the soil from Step 4 and shaken for 24 h.
6. Noncrystalline (amorphous) Fe oxide: Twenty mL 0.175*M* $(NH_4)_2C_2O_4$ + 0.1*M* $H_2C_2O_4$ [24.869 g $(NH_4)_2C_2O_4$ + 12.607 g $H_2C_2O_4$ L^{-1} (oxalate reagent)] were added to the soil from Step 5 and shaken in the dark for 4 h.
7. Crystalline Fe oxide: Twenty mL oxalate reagent were added to the soil from Step 6 and placed in a boiling water bath under ultraviolet irradiation for 3 h. Samples were shaken periodically.

8. Residual: Determined by using the total analysis method outlined below (Shuman, 1979) for the sample remaining from Step 7 after air drying, grinding, and passing through a 35 mesh sieve.

In addition to the complete sequential procedures listed above, separate samples of various soil-to-solution ratios were individually extracted, and the order of extraction was varied to evaluate the effect on amounts of extractable metals.

Total Elemental Analysis (Shuman 1979)

Half-gram air-dried soil, ground to pass a 35-mesh sieve, was weighed into a 50 mL Teflon beaker, and 1 mL of aqua regia (1 part concentrated HNO_3 to three parts concentrated HCl) was added to wet the sample. Eight mL of concentrated HF were added and the sample digested in a sand bath/hot plate for 3 h at 80°C. The temperature was raised to 120°C and the sample was evaporated to dryness. Five mL concentrated HNO_3 were added, the sample was left overnight at room temperature, and then evaporated to dryness at 100°C. Five mL of concentrated HCl were added and the above procedure repeated. Residual salts were dissolved by warming with about 10 mL of 1*M* HNO_3 (64.94 mL 69% HNO_3 L^{-1}), transferred with rinsing using 1*M* HNO_3 into a 25-mL volumetric flask, and taken to volume with 1*M* HNO_3. The digestion solution was analyzed by DCP-AES.

RESULTS AND DISCUSSION

Total Elemental Concentration

Total content of Cd, Cr, Cu, Ni, Pb, and Zn in the soil samples are given in Table 4. The choice of these three sludge-treated soils resulted in diverse concentrations for most of the six elements of interest, providing a good range of metal concentrations to compare the two sequential extraction methods. A fourth soil is listed in the table that was not included in the sequential analysis. This Metea soil sample was collected from an untreated area adjacent to the sludge-treated Metea samples and should contain background levels initially present in the Metea soils.

Chemical Fractionation of Cd, Cr, Cu, Ni, Pb, and Zn

The intent of this study was to select and use a sequential extraction technique for which the selection of extracting solutions and their order of use are based on good chemical reasoning, strong empirical evidence, appropriateness for the soil types with which we are working, and effectiveness in fractionating Cd, Cr, Cu, Ni, Pb, and Zn in the concentration ranges expected. Based on a review of the literature, Methods 1 and 2 (from techniques developed by Shuman (1985) and Miller et al. (1986), respectively) appeared to fit most of these selection criteria. Method 1 had not been demonstrated for the so called "heavy" metals, Cd, Cr, Ni, and Pb. Additionally, the effectiveness of both methods had not been established on the soils of Michigan.

Table 4. Total content of Cd, Cr, Cu, Ni, Pb, and Zn in the 2-mm fraction of three sludge-amended soils and an untreated soil.

Cd	Cr	Cu	Ni	Pb	Zn
			$mg\ kg^{-1}$		
			Metea 1		
4.8±0.4	636±8	366±4	397±5	180±8	1545±19
			Metea 2		
<2.5	43±0	16±1	14±0	56±0	42± 8
			Capac		
6.3±0.0	115±1	46±0	33±1	89±0	123± 5
			Metea (no sludge)		
<2.5	26±1	7±0	11±0	40±1	45± 6

Water-soluble Fraction

The values in the first column of Table 5 and 6 indicate that Cr, Cu, Ni, and Zn occurred in measurable concentrations in the water-soluble fraction of the three soils. Of these four elements, however, only Cu and Ni appeared in measurable quantities in half or more of the samples. The 10-g water to 5-g soil extraction (2:1) ratio resulted in less Cu and Ni solubilized compared to the 20-g water to 0.5-g soil (40:1) ratio when a comparison could be made. However, measurable quantities of soluble elements occurred more often with the 2:1 ratio of Method 1, compared to the 40:1 ratio of Method 2. The lower ratio also provided greater analytical sensitivity, as indicated by the lower detection limits (Table 5 versus Table 6).

Table 5. Chemical fractionation of Cd, Cr, Cu, Ni, Pb, and Zn using Method 1.

Soil	Cd	Cr	Cu	Ni	Pb	Zn
			mg kg^{-1}			
			Water-soluble			
Metea 1	<0.1†	<0.04	0.8±0.1	0.9±0.1	<0.2	0.7±0.0
Metea 2	<0.1	<0.04	0.1±0.0	<0.04	<0.2	<0.12
Capac	<0.1	0.04±0.0	0.2±0.0	0.1±0.0	<0.2	<0.12
			Exchangeable			
Metea 1	<0.2	0.3±0.0	3.0±0.0	23±2	2.2±0.1	37±1
Metea 2	<0.2	0.1±0.0	0.7±0.6	0.8±0.5	1.7±0.4	1.3±0.0
Capac	0.4±0.1	0.2±0.2	1.5±0.0	2.5±0.1	1.7±0.1	4.8±2.6
			Organic			
Metea 1	1.0±0.3	360±40	110±10	63±1	8.2±0.1	375±55
Metea 2	0.2±0.2	8.9±1.5	6.2±0.0	0.8±0.1	4.6±0.1	5.9±0.8
Capac	1.6±0.3	52±1	29±1	9.6±1.1	14±1	27±2
			Mn Oxide			
Metea 1	<2.5	10±2	105±6	38±2	29±4	248±22
Metea 2	<2.5	<1	2.0±0.3	<1	<5	<3
Capac	<2.5	<1	3.7±0.1	1.7±0.0	9.6±0.1	10.8±0.4
			Amorphous Fe Oxide			
Metea 1	<2.5	96±22	61±12	80±14	48±2	290±15
Metea 2	<2.5	<1	2.0±0.5	<1	<5	8.6±3.0
Capac	<2.5	3.4±1.8	4.9±0.3	4.3±2.4	19±17	17±2

Table 5 (cont'd)

Soil	Cd	Cr	Cu	Ni	Pb	Zn
	mg kg^{-1}					
			Crystalline Fe Oxide			
Metea 1	<2.5	5.1±0.1	5.1±0.5	5.8±0.4	9.9±4	35±11
Metea 2	<2.5	10.7±0.9	20±1	37±0	17±0	123±6
Capac	<2.5	3.5±0.4	3.2±0.1	3.2±0.1	10.5±2.0	13±4
			Residue			
Metea 1	<1.4	22±2	9.6±3.7	18±1	58±0	76±6
Metea 2	<1.4	16±1	3.0±0.4	7.7±1.7	22±2	34±0
Capac	<1.4	27±2	3.1±0.2	14±1	23±0	40±7
			Sum of individual fractions			
Metea 1	1.0±0.3	495±17	297±22	230±18	155±6	962±87
Metea 2	0.2±0.2	36±2	34±2	46±1	47±3	177±11
Capac	1.9±0.2	86±6	45±2	35±2	79±14	113±10

†"<" signifies that the value is below analytical detection indicated.

Table 6. Chemical fractionation of Cd, Cr, Cu, Ni, Pb, and Zn using Method 2.

Soil	Cd	Cr	Cu	Ni	Pb	Zn
	$mg\ kg^{-1}$					
			Water-soluble			
Metea 1	<2†	<0.8	2.6±0.1	2.5±0.2	<4	<2.4
Metea 2	<2	<0.8	<0.8	<0.8	<4	<2.4
Capac	<2	<0.8	0.8	<0.8	<4	<2.4
			Exchangeable			
Metea 1	<2	<0.8	2.1±0.3	39±1	<4	180±12
Metea 2	<2	<0.8	<0.8	<0.8	<4	<2.4
Capac	2.1	<0.8	<0.8	1.7	<4	<2.4
			Acid-soluble			
Metea 1	2.4±0.6	35.8±0.2	190±8	142±2	19±3	1040±12
Metea 2	<2	<0.8	4.4	<0.8	4.8	7.3
Capac	<2	7.4	14	6.4	8.8	46
			Manganese Oxide			
Metea 1	<2	<2	33±4	23.3±0.6	13.1±0.4	47±4
Metea 2	<2	<2	<0.8	<0.8	<4	10
Capac	<2	<2	2.4	0.9	<4	7
			Organic			
Metea 1	<2	187±29	52±9	28±4	26±3	53±8
Metea 2	<2	6.4	1.4	<0.8	6.0	9.2
Capac	<2	30	7.8	1.1	6.2	<2.4
			Amorphous Fe Oxide			
Metea 1	<2	135±25	36±6	94±14	13±7	72±7
Metea 2	<2	2.5	1.4	<0.8	<4	9.2
Capac	<2	10	6.7	3.3	11	<2.4

Table 6 (cont'd)

Soil	Cd	Cr	Cu	Ni	Pb	Zn
	- - - - - - - - - - - - -	- - mg kg^{-1} - -	- - - - - - - - - - - - - -			
			Crystalline Fe Oxide			
Metea 1	<2	195±50	23±3	47±6	8.7±2.6	125±7
Metea 2	<2	9.6	3.8	3.4	<4	20
Capac	<2	21	4.5	6.3	7.4	44
			Residue			
Metea 1	<2	35±4	62±79	14±3	45±10	51±18
Metea 2	<2	26	5.9	11	33	21
Capac	<2	29	4.7	9.4	26	28
			Sum of individual fractions			
Metea 1	2.8	600±105	400±110	390±30	125±1	1580±50
Metea 2	<2	46	17	14	44	84
Capac	2.1	99	41	29	64	140

†"<" signifies that the value is below analytical detection indicated.

That measurable quantities of some trace elements occurred using both methods indicated the value of a water-soluble fraction. However, concentrations were either less than primary and secondary regulations of the Safe Drinking Water Act of the U.S. Environmental Protection Agency (i.e., Cr, Cu, and Zn) or below analytical detection limits of DCP-AES (i.e., Cd and Pb; Ni is not regulated). Water-soluble trace elements should be the most available for plant uptake and leaching, representing the greatest potential risk.

Exchangeable Fraction

There is a considerable difference in the concentrations extracted in the exchangeable fraction between the two methods. For those samples in which concentrations were greater than analytical detection limits, 1*M* $Mg(NO_3)_2$ of Method 1 extracted greater amounts of Cu and lower concentrations of Ni and Zn than 0.5*M* $Ca(NO_3)_2$ used in Method 2 (Table 5 versus Table 6). However, because of the more favorable solution to soil ratio we were able to determine lower concentrations of all six trace elements with greater levels of confidence using Method 1 than with Method 2. This is most apparent with Cr, Cu, Pb, and Zn. All three measurements of exchangeable Cr and Pb and two of three measurements of Cu and Zn were below detection limits in solutions of Method 2, whereas in Method 1 all were greater than analytical detection limits.

To further evaluate methodologies, concentrations of the six trace elements extracted with 1*M* $Mg(NO_3)_2$, 1*M* $MgSO_4$, and 0.5*M* $Ca(NO_3)_2$ (all adjusted to pH 7 prior to use) in various solution to soil ratios are listed in Table 7. The amount of each trace element extracted was dependent not only on the extracting solution, but also on sample size. For example, where detected, 0.5*M* $Ca(NO_3)_2$ extracted more or the same quantity of Cd and Cr and less or equal amounts of Cu, Ni, Pb and Zn than did 1*M* $Mg(NO_3)_2$.

These extracting solutions work by exchange reactions wherein the cation (or anion) of the salt solution is preferentially exchanged for the ion on clays, sesquioxides, and organic matter held by electrostatic forces. Although we would predict Ca^{2+} to be a better choice of cation than Mg^{2+} to remove these trace elements, based on Hard and Soft Acids

Table 7. Cadmium, Cr, Cu, Ni, Pb, and Zn extracted with 1*M* $Mg(NO_3)_2$, 1*M* $MgSO_4$, and 0.5*M* $Ca(NO_3)_2$ and various solution to soil ratios.

Soil	Cd	Cr	Cu	Ni	Pb	Zn
	mg kg^{-1}					
	1.0*M* $Mg(NO_3)_2$ (4 solution / 1 soil ratio)					
Metea 1	<0.2†	0.1±0.0	1.7±0.0	18.0±1.5	0.9±0.1	30±2
Metea 2	<0.2	<0.08	0.1±0.0	0.1±0.0	0.4±0.3	<0.2
Capac	0.3±0.0	0.1±0.0	0.3±0.0	1.2±0.3	0.8±0.3	1.4±0.2
	1.0*M* $MgSO_4$ (4 solution / 1 soil ratio)‡					
Metea 1	<0.2	<0.08	1.6±0.0	17±0	<0.4	29±0
	1.0*M* $Mg(NO_3)_2$ (10 solution / 1 soil ratio)					
Metea 1	<0.5	<0.2	2.8±0.0	26±1	2.4±0.3	44±3
Metea 2	<0.5	<0.2	0.3±0.0	3.4±0.1	1.6±0.3	1.0±0.5
Capac	0.5±0.0	<0.2	0.7±0.0	3.3±0.1	2.5±0.0	4.1±0.1
	0.5*M* $Ca(NO_3)_2$ (10 solution / 1 soil ratio)					
Metea 1	<0.5	<0.2	2.4±0.3	32±1	1.5±0.0	111±2
Metea 2	<0.5	<0.2	<0.2	<0.2	<1.0	<0.6
Capac	1.1±0.0	<0.2	0.4±0.0	0.7±0.1	<1.0	<0.6
	0.5*M* $Ca(NO_3)_2$ (40 solution / 1 soil ratio)					
Metea 1	<2.0	<0.8	1.8±0.0	36±1	<4.0	160±12
Metea 2	<2.0	<0.8	<0.8	<0.8	<4.0	<2.4
Capac	2.1	<0.8	<0.8	1.7	<4.0	<2.4

†The value is less than the analytical detection indicated.
‡Only Metea 1 extracted with this solution.

and Bases (HSAB) concepts (Table 3), differences in molar concentration of the extracting solutions also will be a factor affecting the exchange reactions.

Based only on the amount of the elements extracted, no extracting solution emerged as a best choice. Using 5 g of soil with 10 g of 0.5*M* $Ca(NO_3)_2$ as the extraction technique may be the best option based on a desire to maximize detection limits and minimize matrix problems in salt solutions when using DCP-AES to determine concentration. In addition, HSAB theory holds that Ca has a Misono softness parameter more similar to those of Cd, Cr, Cu, Ni, Pb, and Zn compared to Mg and so should be a better cation for exchange.

The use of NO_3^{2-} appeared to be somewhat more effective than SO_4^{2-} for extracting trace elements, including Cr, at the same solution to soil ratio (Table 7). This agreed with HSAB theory as discussed previously.

Acid-soluble Fraction

The concentrations of trace elements in the acid-soluble fraction of the three soils are listed in Table 6. This fraction probably represents elements associated with carbonates, organic matter, and some mineral forms and can account for a considerable amount of the total Cd, Cu, Ni, and Zn occurring is these soils.

Table 8 lists the amount of Cd, Cr, Cu, Ni, Pb, and Zn extracted by 20 mL of 0.44*M* acetic acid solution and various quantities of the Metea 1 soil. Results indicated that as the solution to soil ratio increased, a greater percentage of the total amount of each element present in the soil was removed. This trend was likely due to the lower

Table 8. Acid-soluble Cd, Cr, Cu, Ni, Pb, and Zn extracted from various amounts of Metea 1 soil to obtain different solution:soil ratios.

	No Prior Fraction Extracted				Exchangeable fraction Extracted		
	solution:soil ratio						
	4:1	10:1	20:1	40:1	10:1	40:1	Total
	mg kg^{-1}						
Cd	1.7	2.1	2.3	2.7	1.7	2.4	4.8
Cr	4.6	10	17	31	11	36	636
Cu	54	99	136	182	92	191	366
Ni	123	151	156	184	99	142	397
Pb	3.6	5.3	9.1	22	2.0	17	180
Zn	896	1101	1088	1220	869	1041	1545

pH of the solution to soil mixture that resulted when smaller quantities of soil were extracted compared to a larger sample size.

Generally, thermodynamic data indicated that the solubility of the mineral forms of these trace elements should increase as solution acidity increases (Table 3). Obviously, factors other than the amount of elements that are "specifically-adsorbed", such as the buffering capacity of the soil and the resulting pH, affect the concentration the elements extracted.

The last two columns in Table 8 show quantities of each metal removed if the exchangeable fraction is extracted prior to the acid-soluble fraction. The differences obtained when comparing acid-soluble quantities without versus the exchangeable fraction removed first could be accounted for by the amount of the trace elements present in the exchangeable fraction alone.

Organic Fraction

In Method 1 trace elements in the soil organic fraction were solubilized using 0.7*M* NaOCl at pH 8.5 following the extraction of the exchangeable fraction using 1*M* $Mg(NO_3)_2$. Removing the trace elements in soil organic matter consisted of multiple extractions using 10 g of fresh extracting solution on the same 5 g soil sample. Table 9 lists the results of this extraction technique for the three soils.

The NaOCl solution generally extracted most of the Cd, Cu, Ni, Pb, and Zn from the soils after the first two extractions. Significant Cr, however, was still being extracted in the fourth and fifth subfractions, especially from the Metea 1 and Capac soils that had high Cr levels. Since two or three separate extractions with NaOCl should be sufficient to remove most of the organic matter from these soils, the data suggested that the NaOCl solution may be dissolving forms of Cr other than strictly organic, contrary to the experimental evidence of other researchers (Anderson, 1963, Bascomb, 1968; Lavkulich and Wiens, 1970). Possibly inorganic, insoluble forms of Cr(III) were being oxidized to the more soluble Cr(VI) form. This oxidation could affect the extraction of Cr in subsequent fractions and may be contraindicative of using NaOCl to fractionate organic forms of Cr.

Multiple extractions with NaOCl to remove elements associated with the organic fraction consistently removed a greater amount of the elements from soils than $Na_4P_2O_7$ used in Method 2 (Tables 5 and 6), except for Pb and Zn. These results differed from those of Shuman (1983). In general, greater amounts of Cu and Zn were extracted using NaOCl than $Na_4P_2O_7$. Acid-soluble and Mn oxide fractions were extracted from soils prior to the organic fraction in Method 2. The addition of

Table 9. Cadmium, Cr, Cu, Ni, Pb, and Zn extracted from the organic fraction of 5-g soil samples by up to five successive 10-g aliquot of 0.7*M* NaOCl.

	10-g Aliquot of 0.7*M* NaOCl					Total Removed by the Five Aliquot
	1st	2nd	3rd	4th	5th	
	------------ mg kg^{-1} ------------					
			Cadmium			
Metea 1	0.7	0.1	<0.1	0.2	<0.1	1.0
Metea 2	<0.1	<0.1	<0.1	0.1	<0.1	0.2
Capac	1.2	0.2	<0.1	0.1	<0.1	1.6
			Chromium			
Metea 1	35	82	84	87	60	377
Metea 2	2.2	4.7	1.3	0.7	0.1	8.9
Capac	15	17	11	6.4	1.8	52
			Copper			
Metea 1	85	17	5.6	2.7	1.7	113
Metea 2	4.1	1.0	0.5	0.5	0.1	6.2
Capac	22	4.1	1.4	1.0	0.4	29
			Nickel			
Metea 1	31	18	6.4	3.2	2.6	65
Metea 2	0.61	0.11	<0.04	0.11	<0.04	0.8
Capac	7.3	1.6	0.39	0.27	0.05	9.6
			Lead			
Metea 1	4.9	2.0	0.5	0.5	0.3	8.3
Metea 2	3.8	0.4	<0.2	0.3	<0.2	4.7
Capac	11	2.1	0.5	0.4	<0.2	14
			Zinc			
Metea 1	303	46	17	3.5	3.3	376
Metea 2	3.6	1.0	0.2	0.7	0.3	6.1
Capac	18	5.2	1.4	1.1	0.3	28

these two fractions to the organic fraction, however, cannot account for the differences in the organic fraction extracted by the two separate methods. NaOCl used in the first method has been shown quite effective in removing organic matter from soils, especially after 2 or 3 subfractions (Anderson, 1963; Lavkulick and Wiens, 1970), whereas $Na_4P_2O_7$ used in Method 2 may only be about 30% effective (Stevenson, 1982). Sodium pyrophosphate also is not as specific for the organic fraction as NaOCl, dissolving Fe and Mn oxides (Bascomb, 1968 and McLaren and Crawford, 1973).

Copper, Pb, and Zn precipitated in the 0.7*M* NaOCl solution at pH 8.5 when making multi-element standard. To keep the trace elements soluble, it was necessary to acidify the solution to a pH less than 2 with concentrated HCl. If this same phenomenon occurred when sequentially extracting soils, then NaOCl may be removing trace elements from the organic matter only to precipitate them in another form (possibly as an oxide) that is insoluble in 0.7*M* NaOCl in the basic pH range. Nevertheless, NaOCl was still generally more effective in extracting trace elements than $Na_4P_2O_7$. Using NaOCl at pH 8.5 rather than 9.5 should reduce precipitation of trace elements (Shuman, 1983).

The difficulty of making analytical standards in NaOCl was not insurmountable. Shuman (1983) did not comment on this problem because he did not measure elemental concentrations in the NaOCl solution. Rather, due to erratic readings upon direct analysis with atomic absorption spectrophotometry, the NaOCl filtrate was evaporated to dryness and then redissolved in 1*M* HNO_3. The problems of erratic readings did not occur with DCP-AES, probably because a peristaltic action was used to pump samples for analysis. In atomic absorption spectrometry, samples are aspirated into the flame using the vacuum created from the flow of the combustion gases.

Multiple extractions using 0.7*M* NaOCl appeared to be more effective for dissolving organic matter and less active at dissolving other soil fractions than $Na_4P_2O_7$. The number of subfractions taken using NaOCl, however, should be limited to three in these soils to reduce the amount of Cr that may solubilize from non-organic forms.

<u>Oxide Fractions</u>

Essentially the same techniques are used in Methods 1 and 2 to determine both Mn and Fe oxides. The major differences included the order of extraction and the composition of the extracting solutions. In Method 1 the Mn oxide fraction followed the organic fraction. The soil was dried, ground, and screened through a 35-mesh sieve after the organic fraction was extracted. This took a considerable amount of time and may have exposed new surfaces to the action of the extractant, resulting in more elements dissolving from previously less soluble fractions.

Comparing the results of the Mn oxide fractions extracted by the two methods in Tables 5 and 6 indicated a large difference in the concentrations of trace elements extracted, probably due to the fractions taken prior to this. Concentrations in the Mn oxide fraction with Method 2 were generally lower than with Method 1. The acid-soluble fraction of Method 2, taken prior to the Mn oxide fraction, probably dissolved elements associated with Mn oxides (and possibly also those associated with organic matter and the Fe oxides).

Method 1 used more concentrated extracting solutions to solubilize the Fe oxide fractions than Method 2. The chemicals in the preparations of these solutions dissolved only with great difficulty. Making multi-element standard solutions also was difficult. Reducing the oxalate concentrations to the level specified in Method 2 may solve these problems without a significant effect on the ability of these solutions to solubilize the desired fractions.

In order to better standardize the technique for removing the crystalline Fe oxide fraction, Shuman (1985) suggested the use of ascorbic acid rather than ultra-violet light. However, multi-element standard solutions were difficult to make using this solution. The elements precipitated and remained in the solid form even after heating, shaking, further diluting, and acidification. Also, results were inconclusive about whether using ascorbic acid was more effective than UV radiation in dissolving elements from the crystalline Fe oxide fraction (Table 5 and 6).

Residual Fraction and Total Analysis

The residual fractions in both methods were determined using the same technique. The differences between methods in trace element concentrations found in this fraction were a result of the preceding extractions. Except for Cu, however, there were only relatively small differences in concentrations measured in the soil residue between the two methods even though large procedural differences were used in extracting preceding fractions (Tables 5 and 6). The most telling differences between the two fractionation techniques was the sum of the individual elements from each fraction (last columns of Tables 5 and 6) compared to the total elemental concentrations determined on intact soils using the wet digestion method (Table 4). The sum of the fractions determined using Method 2, even with all of its theoretical concerns such as determination of an acid-soluble fraction and using $Na_4P_2O_7$ to solubilize the organic fraction, compared well with the total analysis for most of the elements. Method 1 sum totals did not compare as well. The major problems of Method 1, however, were mostly associated with analytical difficulties of trace element analysis in the two Fe oxide fractions. Good analytical standards were difficult to properly make in the Fe oxide solutions of Method 1.

SUMMARY AND CONCLUSIONS

Various soil chemical fractions and techniques that have been used to determine trace element concentrations in them were reviewed. Fractions measured can include water-soluble, exchangeable, carbonate, organic, Mn and Fe oxide, and residual components. Three soils to which municipal sewage sludges were applied were sequentially extracted using two fractionation methods. The methods were chosen based on a review of the literature. Method 1 was modified from the work of Shuman (1985) and Method 2 came from the work of Miller et al. (1986).

For most of the trace elements measured in each of the fractions of the three soils, agreement between the two methods was only fair to poor, whether the comparison was made based on operational criteria (order in which the fractions were measured) or on a theoretical basis (comparing the same expected fractions to one another, regardless of the sequence in which it was measured). Cautions others have voiced when interpreting results become obvious when searching for explanations of the wide disparities between methods. Absolute values obtained from any technique may be of only limited value, as are comparisons made between dissimilar soils. Instead, relative differences using the same extraction technique on similar soils to which different treatments have been imposed (e.g., sludge versus no sludge, changes over time, differences due to cropping, trace element concentration differences) probably have real value and fill a legitimate need few other techniques currently offer.

Another reason for the discrepancies between the two techniques may be attributed to analytical deficiencies when measuring levels of elements in solutions that are near the analytical detection limits of DCP-AES. This may be a good reason to use lower solution to soil ratios when conducting these fractionation studies. However, the sample size will affect the outcome of at least some of the fractionation procedures, even when everything else is held constant. Using a greater solution to soil ratio, for example, can change the relative amount of trace elements extracted by a particular solution, as seen when extracting the acid-soluble fraction. Also, complete reaction or dissolution to obtain a fraction may take longer when greater solution to soil ratios are used.

Of primary importance in choosing any procedure is to insure that the method includes important soil chemical fractions of which the soil is composed. For example, including a carbonate or sulfide extraction when these fractions do not exist in a sample may not only contribute to unnecessary work, but, more importantly, may affect the subsequent extraction of other fractions. Additionally, comparisons with soils on which other extraction methods have been used are made more tenuous.

At this point, further work on justifying the selection of one technique over the other is not warranted. Simply comparing fractionation techniques, whether one is comparing different sample sizes using the same method or two completely different fractionation procedures, did not of itself help to determine which method better measures trace elements in soil fractions, nor their environmental availability. Both methods are adequate for acidic mineral soils with appreciable amounts of Mn and Fe oxides. Method 2 is more appropriate when there is an interest in measuring Cr because of problems when extracting it from the organic fraction using NaOCl. Neither method is necessarily better nor worse than the other, since neither is perfect.

These chemical fractionation techniques are like the simpler soil testing methods that have been used for decades to assess plant uptake of nutrients; they are technique and soil (and, to some extent, time and technician) specific. The opportunity for developing a single method for all soils or sediments and trace elements is low. However, when used properly, they are capable of generating information about differences in trace element bioavailability no other method can give.

LIST OF REFERENCES

LIST OF REFERENCES

Anderson, J.U. 1963. An improved pretreatment for mineralogical analysis of samples containing organic matter. Clays Clay Miner. 10:380-388.

Baker, D.E., and M.C. Amacher. 1982. Nickel, copper, zinc, and cadmium. p. 323-336. *In* A.L. Page (ed.) Method of soil analysis. No. 9 (Part 2). Am. Soc. Agron., Inc., and Soil Sci. Soc. Am., Inc., Madison, WI.

Bartlett, R.J., and J.M. Kimble. 1976a. Behavior of chromium in soils: I. Trivalent forms. J. Environ. Qual. 4.379-383.

Bartlett, R.J., and J.M. Kimble. 1976b. Behavior of chromium in soils: II. Hexavalent forms. J. Environ. Qual. 4.384-386.

Bascomb, C.L. 1968. Distribution of pyrophosphate extractable iron and organic carbon in soils of various groups. J. Soil Sci. 19:251-268.

Bohn, H.L., B.L. McNeal, and G.A. O'Connor. 1979. Soil chemistry. John Wiley & Sons. New York. pp. 329.

Burau, R.G. 1982. Lead. p. 347-365. *In* A.L. Page (ed.) Method of soil analysis. No. 9 (Part 2). Am. Soc. Agron., Inc., and Soil Sci. Soc. Am., Inc., Madison, WI.

Chao, T.T. 1972. Selective dissolution of manganese oxides from soils and sediments with acidified hydroxylamine hydrochloride. Soil Sci. Soc. Am. Proc. 36:764-768.

Chao, T.T. 1984. Use of partial dissolution techniques in geochemical exploration. J. Geochem. Explor., 20:101-135.

Chester, R. and M.J. Hughes. 1967. A chemical technique for the separation of ferro-manganese minerals, carbonate minerals, and absorbed trace elements from pelagic sediments. Chem. Geol. 2: 249-262.

Corey, R.B. 1990. Physical-chemical aspects of nutrient availability. *In* R.L. Westerman (ed.) Soil testing and plant analysis, third edition. SSSA, Madison, WI.

CRC Press. 1987. CRC handbook of chemistry and physics. 67th ed. R.C. Weast (ed.). CRC Press, Inc., Boca Raton, FL.

Doner, H.E. 1978. Chloride as a factor in mobilities of Ni(II), Cu (II), and Cd(II) in soil. Soil Sci. Soc. Am. J. 42:882-885.

Elliott, H.A., B.A. Dempsey, and P.J. Maille. 1990. Content and fractionation of heavy metals in water treatment sludges. J. Environ. Qual. 19:330-334.

Emmerich, W.E., L.J. Lund, A.L. Page, and A.C. Chang. 1982. Movement of heavy metals in sewage sludge-treated soils. J. Environ. Qual. 11:178-181.

Eary, L.E., and D. Rai. 1991. Chromate reduction by subsurface soils under acidic conditions. Soil Sci. Soc. Am. J. 55:676-683.

Florence, T.M. and G.E. Batley. 1977. Determination of the chemical forms of trace metals in natural waters, with special reference to copper, lead, cadmium and zinc. Talanta. 24:151-158.

Grove, J.H., and B.G. Ellis. 1980. Extractable chromium as related to soil pH and applied chromium. Soil Sci. Soc. Am. J. 44:238-242.

Grossman, R.B., and J.C. Millet. 1961. Carbonate removal from soils by a modification of the acetate buffer method. Soil Sci. Soc. Am. Proc. 25:325-326.

Gupta, S.K., and K.Y. Chen. 1975. Partitioning of trace metals in selective chemical fractions of nearshore sediments. Environ. Letters. 10:129-158.

Hirst, D.M., and G.D. Nicholls. 1958. Techniques in sedimentary geochemistry. 1. Separation of detrital and non-detrital fractions of limestones. J. Sediment. Petrol., 28:461-468.

Hodgson, J.F. 1960. Cobalt reactions with montmorillonite. Soil Sci. Soc. Am. Proc., 24:165-168.

Jackson, M.L. 1985. Soil chemical analysis--advanced course. 2nd edition, 11th printing. Published by the author, Madison, WI.

Kotrlý, S., and L. Šůcha. 1985. Handbook of chemical equilibria in analytical chemistry. John Wiley & Sons, New York, NY.

Lake, D.L., P.W.W. Kirk, and J.N. Lester. 1984. Fractionation, characterization, and speciation of heavy metals in sewage sludge and sludge-amended soils: a review. J. Environ. Qual. 13:175-183.

Lavkulich, L.M., and Wiens. 1970. Comparison of organic matter destruction by hydrogen peroxide and sodium hypochlorite and its effect on selected mineral constituents. Soil Sci. Soc. Am. J. 42:421-428.

Lindsay, W.L. 1979. Chemical equilibria in soils. John Wiley & Sons, Inc., New York, NY.

Mattigod, S.V., G. Sposito, and A.L. Page. 1981. Factors affecting the solubilities of trace metals in soils. *In* M. Stelly (ed.) Chemistry in the soil environment. ASA special publication No. 40. ASA, SSSA, Madison, WI.

McLaren, R.G., and D. V. Crawford. 1973. Studies on soil copper I. The fractionation of copper in soils. J. Soil Sci. 24:172-181.

McKeague, J.A., J.E. Brydon, and N.M. Miles. 1971. Differentiation of forms of extractable iron and aluminum in soils. Soil Sci. Soc. Am. Proc. 35:33-38.

McKeague, J.A., and J.H. Day. 1966. Dithionite- and oxalate-extractable Fe and Al as aids in differentiating various classes of soils. Can. J. Soil Sci. 46:13-22.

Merck. 1989. The Merck Index. 11th ed. Merck & Co., Inc., Rahway, NJ

Miller, W.P., D.C. Martins, and L.W. Zelazny. 1986. Effect of sequence in extraction of trace metals from soils. Soil Sci. Soc. Am. J. 50:598-601.

Misono, M., E. Ochiai, Y. Saito, and Y. Yoneda. 1967. A new dual parameter scale for the strength of Lewis acids and bases with the evaluation of their softness. J. Inorg. Nucl. Chem. 29:2685-2691.

NAS. 1974. Chromium. Anna M. Baetjer (Chmn.). National Academy of Sciences. Washington, DC. pp. 155.

Norvell, W.A. 1972. Equilibria of metal chelates in soil solution. *In* J.J. Mortvedt, P.M. Giordano, and W.L. Lindsay (eds.) Micronutrients in agriculture. SSSA, Inc. Madison, WI. pp. 115-138.

Pearson, R.G. 1963. Hard and soft acids and bases. J. Am. Chem. Soc. 22:3533-3539.

Pickering, W.F. 1981. Selective chemical extraction of soil components and bound metal species. CRC Critical reviews in analytical chemistry. 12:233-266.

Ray, S., H.R. Gault, and C.G. Dodd. 1957. The separation of clay minerals from carbonate rocks. Am. Mineralogist, 42:681-686.

Reisenauer, 1982. Chromium. p.337-346. *In* A.L. Page (ed.) Methods of soil analysis, Number 9 (Part 2) 2nd ed. American Society of Agronomy, Inc., and Soil Science Society of America, Inc., Madison, WI.

Shuman, L.M. 1979. Zinc, manganese, and copper in soil fractions. Soil Sci. 127:10-17.

Shuman, L.M. 1982. Separating soil iron- and manganese-oxide fractions for microelement analysis. Soil Sci. Soc. Am. J. 46:1099-1102.

Shuman, L.M. 1983. Sodium hypochlorite methods for extracting microelements associated with soil organic matter. Soil Sci. Soc. Am. J. 47:656-660.

Shuman, L.M. 1985. Fractionation method for soil microelements. Soil Sci. 140:11-22.

Shuman, L.M. 1988. Effect of phosphorus level on extractable micronutients and their distribution among soil fractions. Soil Sci. Soc. Am, J. 52:136-141.

Sims, J.T., and J.S. Kline. 1991. Chemical fractionation and plot uptake of heavy metals in soils amended with co-composted sewage sludge. J. Environ. Qual. 20:387-395.

Sposito,. G. 1981. The thermodynamics of soil solutions. Oxford Univ. Press, New York, NY.

Stevenson, F.J. 1982. Humus chemistry. John Wiley & Sons, New York, NY.

Stover, R.C., L.E. Sommers, and D.J. Silviera. 1976. Evaluation of metals in wastewater sludge. J. Water Poll. Contr. Fed. 48:2165-2175.

Tessier, A., and P.G.C. Campbell. 1988. Partitioning of trace metals in sediments. *In* J.R. Kramer and H.E. Allen (ed.) Metal speciation: Theory, analysis, and application. Lewis Publishers, Inc., Chelsea, MI.

Tessier, A., P.G.C. Campbell, and M. Bisson. 1979. Sequential extraction procedure for the speciation of particulate trace metals. Anal. Chem. 51:884-851.

CHAPTER TWO:

MOVEMENT OF Cd, Cr, Cu, Ni, Pb, AND Zn FROM MUNICIPAL SEWAGE SLUDGES IN A SANDY LOAM SOIL

ABSTRACT

The protection of our soil resource requires a complete understanding of the movement of trace elements. Municipal sludges containing Cd, Cr, Cu, Ni, Pb, and Zn were applied to research plots beginning in 1977 and continuing through 1986. Total elemental analysis of soils collected in 1989 and 1990 indicated that some lateral movement of trace elements, associated with physically moving soil particles with agronomic operations, had occurred. These elements, however, have not moved below the 15 to 30 cm sample depth. Mass balance calculations resulted in average recoveries of trace elements from 45 to 114% of the total applied. These calculations were highly variable, indicative of the highly variable nature of sewage sludge composition, lack of total uniform sludge applications, soil movement due to tillage, and sampling methods.

INTRODUCTION

Municipal sewage sludges, which are high in organic matter and nutrient content, can have a beneficial effect on plant growth and crop yields when applied to soils. However, municipal sludges may have high concentrations of trace elements, e.g., Cd, Cr, Cu, Ni, Pb, and Zn, that may be of environmental concern, especially when applied to soil at relatively high rates.

Many research projects have addressed the agronomic and environmental effects of applying municipal sewage sludges to cropland. This was seen as at least one answer to the reuse and recycling of this potentially useful material. Research efforts have emphasized the fate of trace elements from sludges applied to soil and their uptake by plants. The benefits of sewage sludge application to land in terms of the nutrition, growth, and yields of crops have been documented.

Scientists also have dealt with environmental quality concerns resulting from the application of sludges and the concurrent loading of trace elements onto soils. Several studies have addressed the potential for trace elements to leach from the plow layer into subsoil horizons (Chang et al., 1982; Chang et al., 1984; Emmerich et al., 1982; Kelling et al., 1977; McGrath, 1987; Williams et al., 1980; and Williams et al., 1984). Most of these studies concluded that downward movement of trace elements either did not occur (Chang et al., 1982; Emmerich et al., 1982; Kelling et al., 1977; and McGrath, 1987) or was limited to a depth of 10 cm or less below the zone of incorporation (Chang et al., 1984; McGrath and Lane, 1989; Williams et al., 1980; and Williams et al., 1984).

Many of these experiments were of relatively short duration, reporting data that were taken while sludge applications were still being made or less than ten years after the last sludge application (McGrath, 1987). Results indicated that the trace elements were strongly bound in the topsoil to which they were applied. However, few studies have been able to account for more than 90% of the estimated quantities of trace elements applied to the soil. Most studies could only account for 70% or less of the elements applied. This has been attributed to errors in sampling, chemical analysis, the conversion of concentrations of metals in soil to loadings (which are based on measurements of soil bulk density), and depth of sludge incorporation (Chang et al., 1984; McGrath, 1984). Plant uptake and leaching have not been found to significantly affect trace element concentrations in the soil. Soil movement as a result of tillage operations and soil erosion also may contribute to the low apparent recoveries of trace elements, especially in field experiments (McGrath, 1987).

The objective of this study was to examine the extent to which Cd, Cr, Cu, Ni, Pb, and Zn, applied via sewage sludges to an experimental plot area between 1977 and 1986, have moved laterally in the plow layer and vertically in the soil profile. Additionally, the extent to which their lateral distribution in the soil can affect their apparent recovery will be examined.

MATERIALS AND METHODS

Sample Collection

Municipal sewage sludges from different sources were applied from 1977 to 1986 to plots located on the Michigan State University Soil Science Farm, at the corner of Mt. Hope and Hagadorn Roads. The soil was mapped as a Metea sandy loam (loamy, mixed, mesic Typic Hapludalf).

The experimental field design was established as a randomized complete block having an untreated control and three treatments, each replicated four times. Each of the 16 plots was 6.1 x 30.5 m (20 x 100 ft) in size. Control plots have had no sludges applied to them throughout the duration of the experiment. The sources and composition of sludges used in the study are summarized in Table 10. The years in which sewage sludges were applied to the three treatments (Treatment 1, 2, and 3), the amount of sludge applied, and approximate total trace element loading rates are summarized in Table 11.

Municipal sludges were applied to the treated plots using a manure spreader. Moist weight of the sludges applied was calculated by subtracting the empty weight of the spreader from the weight of the spreader and sludge. Subsamples of the sludge were taken at the time of application to determine the average dry weight and total elemental concentration of the sludge.

Surface soil samples were collected in the spring of 1989 along three transects that were perpendicular to the direction in which the sludges were applied. The transects were parallel to the direction of the dominant slope (<2%) and the plowing, disking, and planting

Table 10. Source and composition of municipal wastewater sludges applied from 1977 to 1986.

Year	Source	Cd	Cr	Cu	Ni	Pb	Zn
		- - - - - - - -	-	mg kg^{-1} sludge	-	- - - - - - -	
1977	Grand Rapids, MI	38	5990	3440	1800	650	5200
1978	Grand Rapids, MI	44	6020	3000	2510	1360	12000
1979	Grand Rapids, MI	30	3200	2200	1400	700	4200
1980	Grand Rapids, MI	30	2090	1700	630	190	4120
1981	Saline, MI	11	8800	5700	5000	260	1020
1982	Saline, MI	6	4310	2910	4170	390	360
1982	Tawas City, MI	8	3630	750	12980	680	21500
1983	Saline, MI	16	4280	3600	3550	490	990
1983	Sandusky, MI	15	110	310	50	360	24000
1984	Jackson, MI	240	4200	625	540	250	7050
1986	Saline, MI	11	4300	3250	3860	440	680

operations. The transects were located about 8, 15, and 23 m north and parallel to the southern boundary of the experimental plot area (Figure 1). Soil samples were collected from the 0 to 15 cm depth at 30 cm increments along each transect for a total of 161 samples per transect (16 plots $\times$ 10 samples per plot + 1 sample at the boundary of the experimental area and first plot). Plastic-lined paper bags were used to store the samples, which were air-dried and sieved through a 2-mm mesh metal screen prior to elemental analysis. A dry-ashing method (Ritter et al., 1978) was used on these samples to determine total element concentrations.

In the spring of 1990, soil samples were collected at 0-15, 15-30, 30-46, 46-76, and >76 cm depths (0-6, 6-12, 12-18, 18-30, and >30 in.) by using a Giddings soil probe to sample sixteen 6 x 33 m (20 x 100 ft) plots in the spring of 1990. Samples from each plot were composites of

Table 11. Sludge application data for Treatments 1, 2, and 3 from 1977 to 1986.

Sludge Source	Year	Sludge† Applied	Cd	Cr	Cu	Ni	Pb	Zn
		$Mg\ ha^{-1}$	- - - - - - - - - -	kg ha^{-1}	- - - - - - - -			
			Treatment 1					
Grand Rapids, MI	1977	10	0.4	70	40	20	7	60
Grand Rapids, MI	1980	20	0.6	40	30	10	4	80
Saline, MI	1981	30	0.3	260	170	150	8	30
Jackson, MI	1984	180	43	750	110	100	45	1260
Total		240	44	1120	350	280	60	1430
			Treatment 2					
Grand Rapids, MI	1977	110	4.3	670	390	200	73	580
Grand Rapids, MI	1978	110	4.8	650	330	270	150	1300
Grand Rapids, MI	1979	82	2.5	260	180	115	57	340
Grand Rapids, MI	1980	110	3.4	235	190	71	21	460
Saline, MI	1981	94	1.0	830	540	470	24	100
Tawas City, MI	1982	73	0.6	265	2	950	50	1570
Sandusky, MI	1983	265	3.9	30	83	13	100	6400
Sandusky, MI	1984	22	0.3	2	7	1	8	530
Total		870	21	3000	1800	2100	480	11300
			Treatment 3					
Grand Rapids, MI	1978	110	4.8	650	330	270	150	1300
Grand Rapids, MI	1979	165	4.9	530	360	230	120	690
Grand Rapids, MI	1980	110	3.4	240	190	71	21	460
Saline, MI	1982	105	0.6	450	310	440	41	37
Saline, MI	1983	130	2.1	550	460	450	62	130
Saline, MI	1986	70	0.8	300	230	270	31	47
Total		690	17	2700	1870	1730	420	2670

†Dry weight basis

5 subsamples taken from about a 1.5 x 17 m (5 x 50 ft) area in the middle of each plot. The five subsamples were mixed in plastic buckets

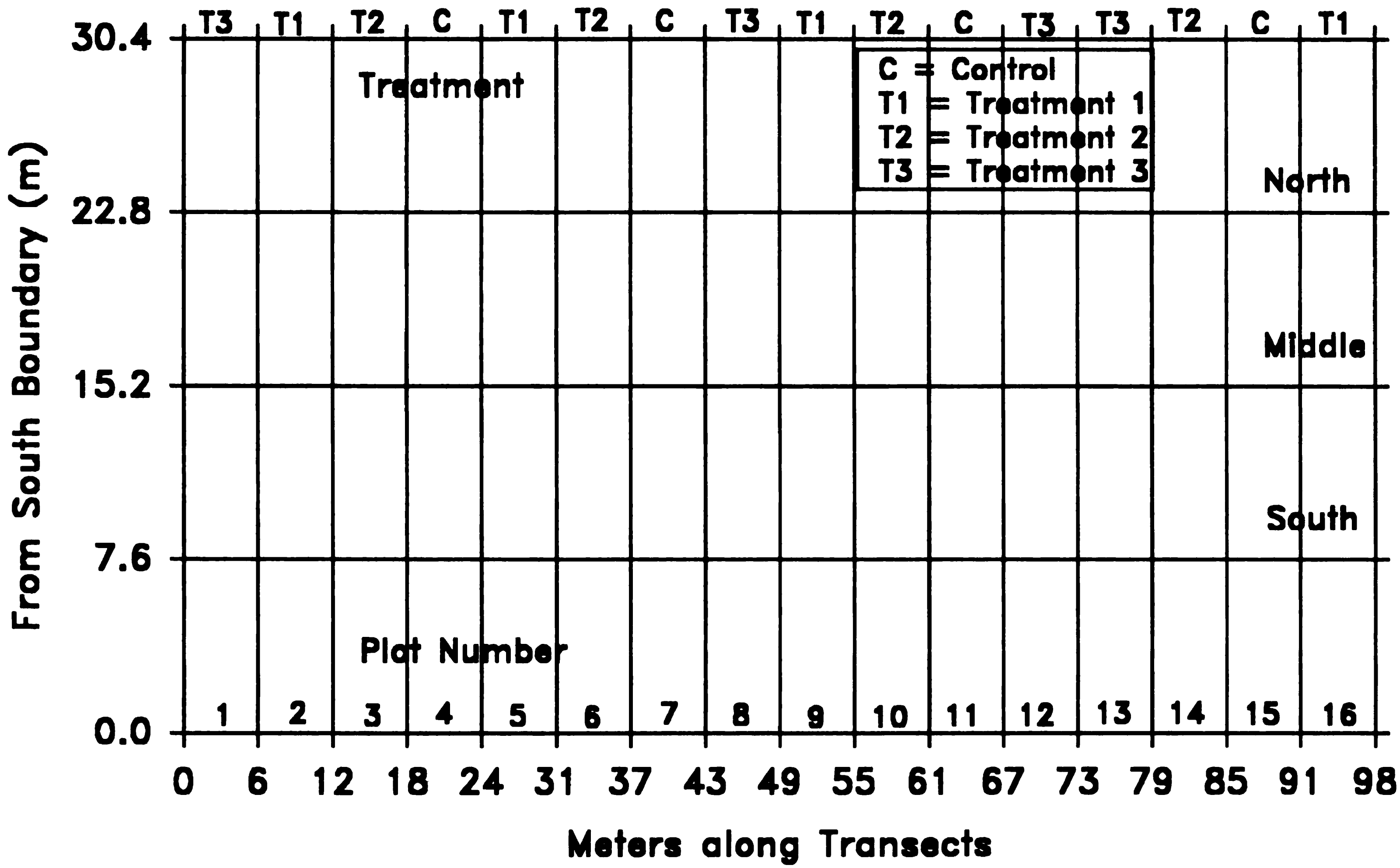

Figure 1. Plot and transect layout for sludge application experiment.

in the field; then a portion of this soil was air-dried, passed through a 2-mm sieve, and stored in plastic bags at room temperature until analyzed.

Laboratory Analyses

Total elemental concentration of the sludges were determined on ground samples using a wet digestion technique (Pierzynski, 1985). Total trace element concentrations in the soil samples were determined using the dry ashing method of Ritter et al. (1978) and a wet digestion technique from Shuman (1979). Analytical concentrations of Cd, Cr, Cu, Ni, Pb, and Zn in filtrates and digest solutions were determined using an Applied Research Laboratory SpectraSpan VB Direct Current Plasma-Atomic Emission Spectrometer (DCP-AES). Statistical analyses were performed using procedures supplied by SAS Institute Inc. (SAS, 1985).

Total Elements in Sewage Sludge (Pierzynski, 1985):

Samples were ground in Coors AD-99 aluminum oxide grinding vials. Sludge digestion was done by placing 1-g samples in Teflon beakers covered with Teflon watch covers and refluxing on a sand bath at 120°C with 20 mL of concentrated HNO_3 overnight. The covers were then removed and the volume was reduced to about 3 mL. Fifteen mL of concentrated HF and 2 mL of concentrated $HClO_4$ were then added, the watch covers were returned, and the beakers were allowed to reflux on a sandbath at 120°C overnight. The covers were removed and the samples were allowed to come to dryness. The dried material was dissolved in 12.5 mL of 6*M* HNO_3 and brought to 25 mL with 2000 mg Li^{+} L^{-1}.

Total Elements in Soil by Dry Ashing (Ritter et al., 1978):

A 2-g sample of air-dried 2-mm soil was weighed into a porcelain crucible and ignited at 550°C for 2.5 h in a muffle furnace. The sample was transferred using about 25 mL 3*M* HCl to a 50-mL Folin-Wu tube and mixed. The mixture was then heated at 120°C for 2 h on a Technicon block heater. The sample was vortexed immediately prior to placing it on the heating block, after 1 h on the block, and once again at the end of the two h heating period. Then it was diluted to 50 mL with deionized water and mixed thoroughly. The sample was then allowed to stand overnight to permit the solids to settle before decanting into a plastic vial. The solution was subsequently analyzed for Cd, Cr, Cu, Ni, Pb, and Zn using DCP-AES.

Total Elements by Wet Digestion (Shuman, 1979):

A half-gram air-dry soil sample was finely ground to pass through a 35-mesh sieve and weighed into a 50-mL Teflon beaker. One mL of aqua regia (1 part concentrated HNO_3 to three parts concentrated HCl) was added to wet the sample. Eight mL of concentrated HF were then added and the sample digested in a sand bath on a hot plate for 3 h at 80°C. The temperature was subsequently raised to 120°C and the sample evaporated to dryness. Five mL of concentrated HNO_3 were added, the sample left overnight at room temperature, and then evaporated to dryness at 100°C. Five mL of concentrated HCl were added and the above procedure repeated. Residual salts were dissolved by warming in 1*M* HNO_3, transferred into a 25-mL volumetric flask with rinsing and taken to volume in 1*M* HNO_3. The digestion solution was analyzed for Cd, Cr, Cu, Ni, Pb and Zn by DCP-AES.

RESULTS AND DISCUSSION

Horizontal Movement of Trace Elements

Figures 2 to 7 graphically display the average trace element concentrations found in the surface soil samples collected along the three transects. Examination of these graphs indicate some lateral movement of the trace elements, but the greatest concentrations remained in the center of the sludge treated plots while control plots (plots 4, 7, 11, and 15) had the lowest concentrations. Soils collected near plot boundaries usually exhibited trace element concentrations that might result as a consequence of mixing soils from the two plots. Trace element concentrations near the plot borders tended to be either greater or less than those found in the middle, depending on the relative differences in trace element loadings between the bordering plots. The lateral movement of trace elements appeared to be related to the physical movement of soil particles during tillage, which also was observed by McGrath and Lane (1989). It was not possible, however, to determine whether factors other than tillage may have influenced this movement (e.g., mass flow or diffusion) or if some elements have moved more than others.

McGrath and Lane (1989) concluded that the dispersion of elements with soil due to tillage can be enough to reduce the apparent concentration of the elements over a period of time. This would be apart from any loss due to leaching or plant uptake. The elements in a treated plot would be diluted as the soil was plowed, tilled, or eroded

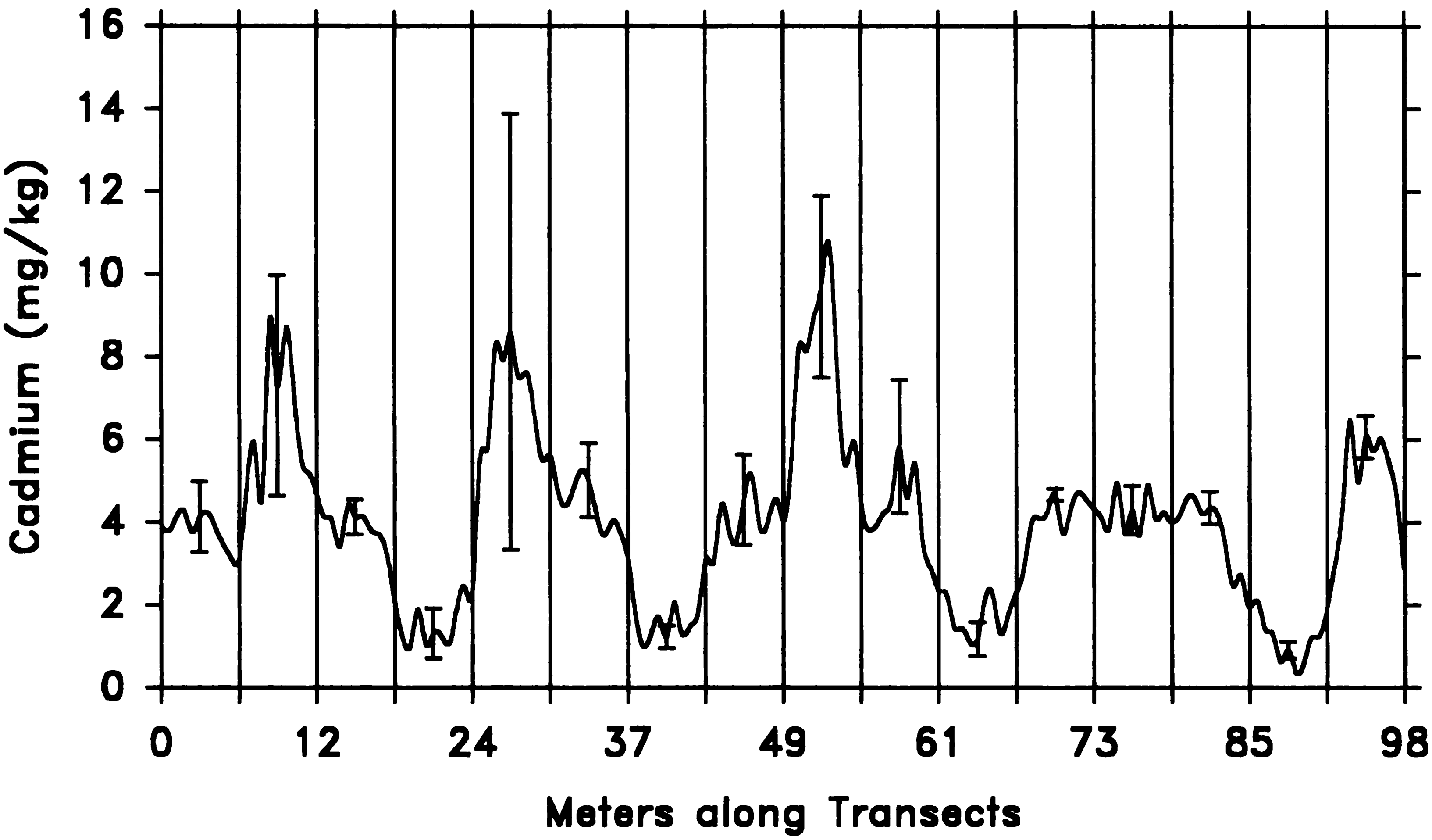

Figure 2. Average total Cd concentrations (with error bars representing standard deviations) found in surface samples collected along three transects across sludge-treated plots.

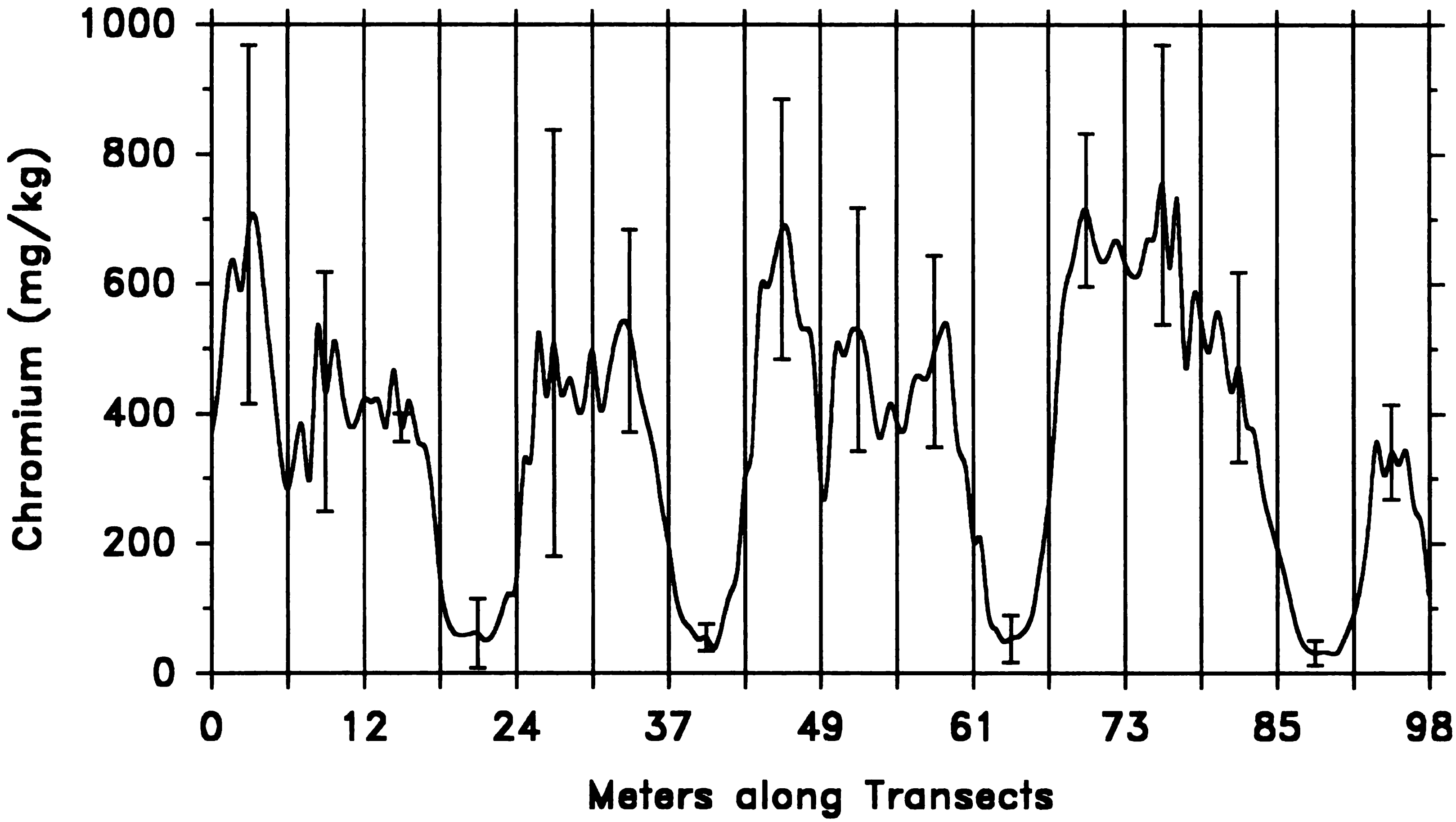

Figure 3. Average total Cr concentrations (with error bars representing standard deviations) found in surface samples collected along three transects across sludge-treated plots.

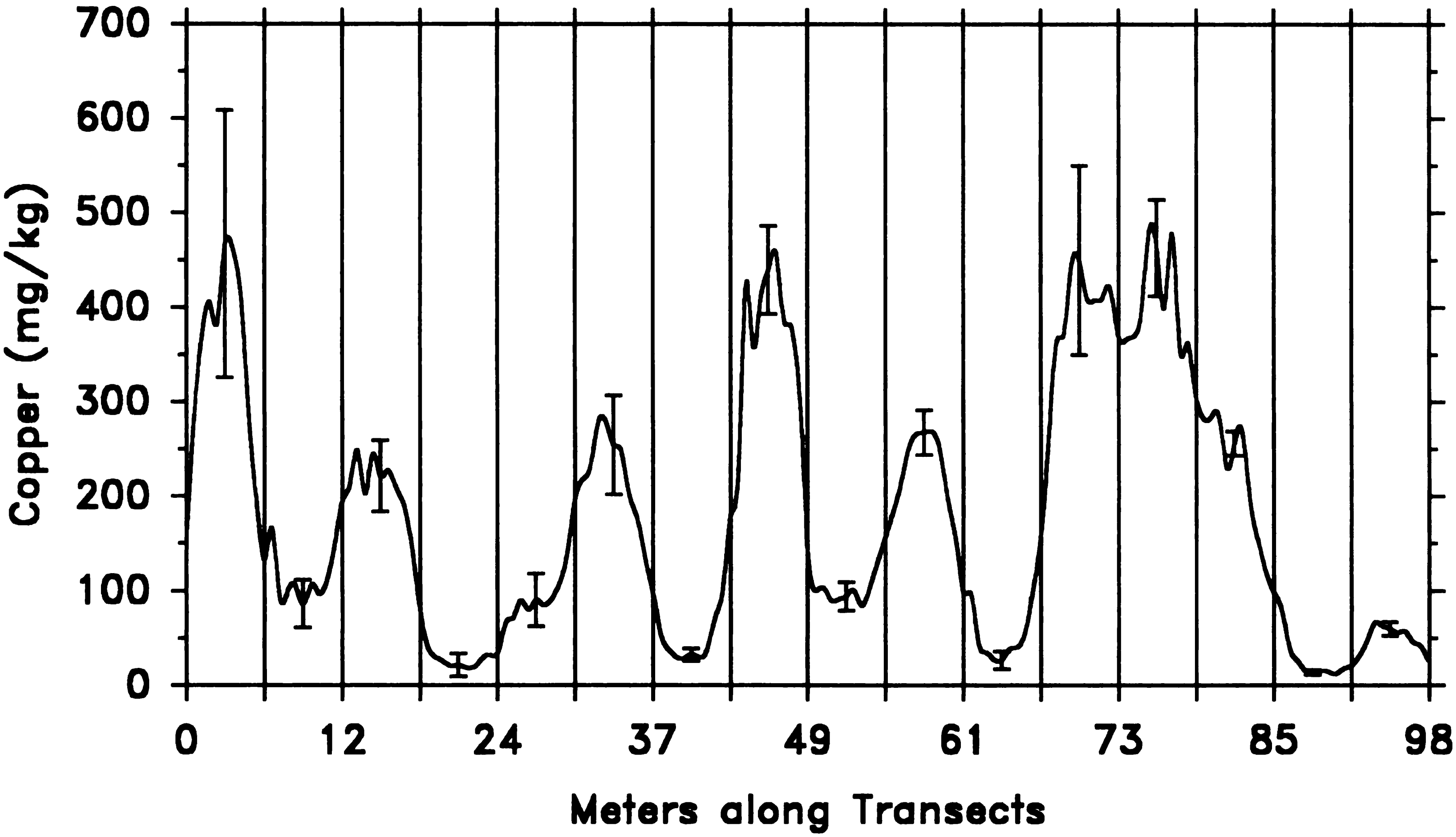

Figure 4. Average total Cu concentrations (with error bars representing standard deviations) found in surface samples collected along three transects across sludge-treated plots.

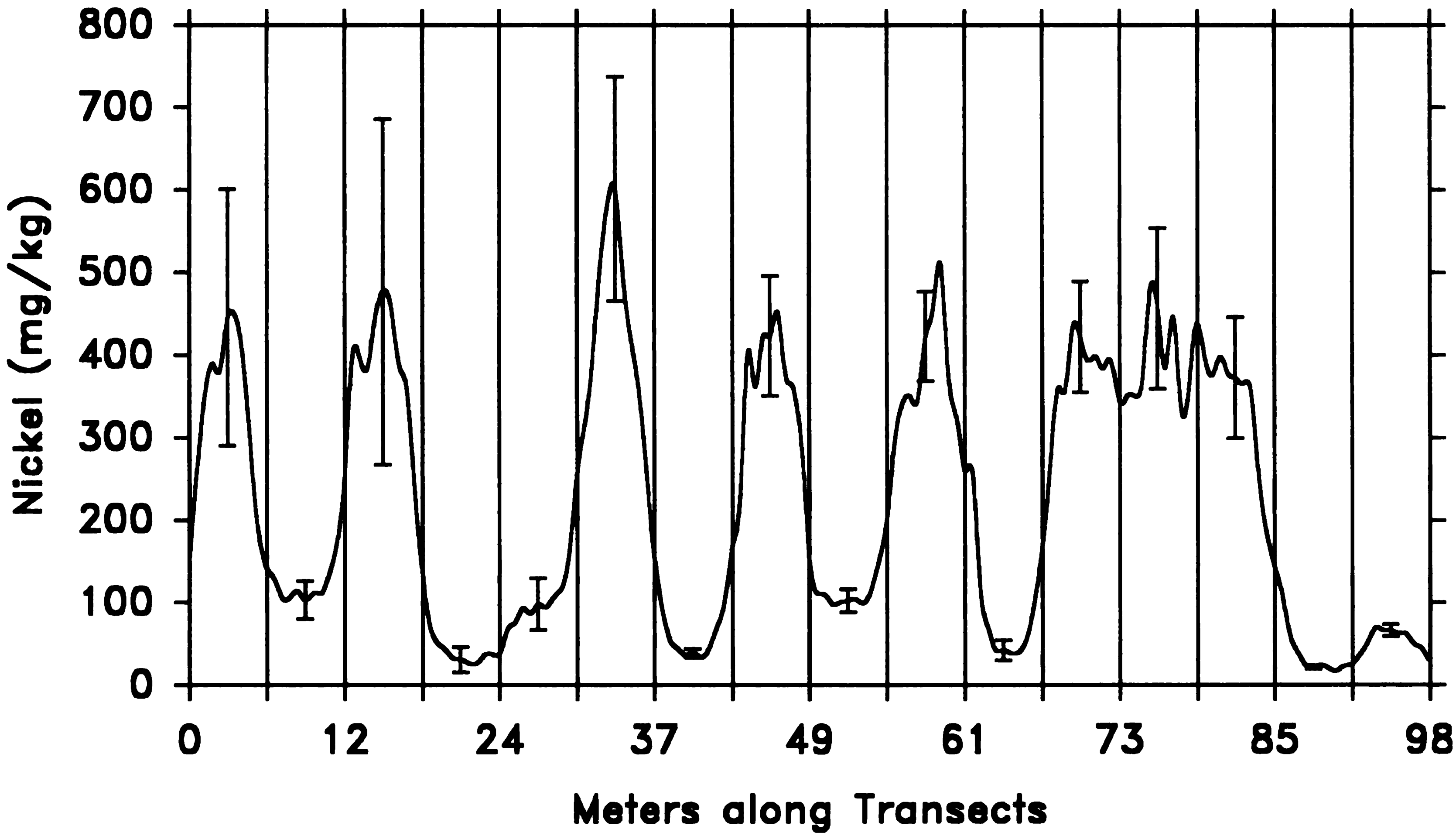

Figure 5. Average total Ni concentrations (with error bars representing standard deviations) found in surface samples collected along three transects across sludge-treated plots.

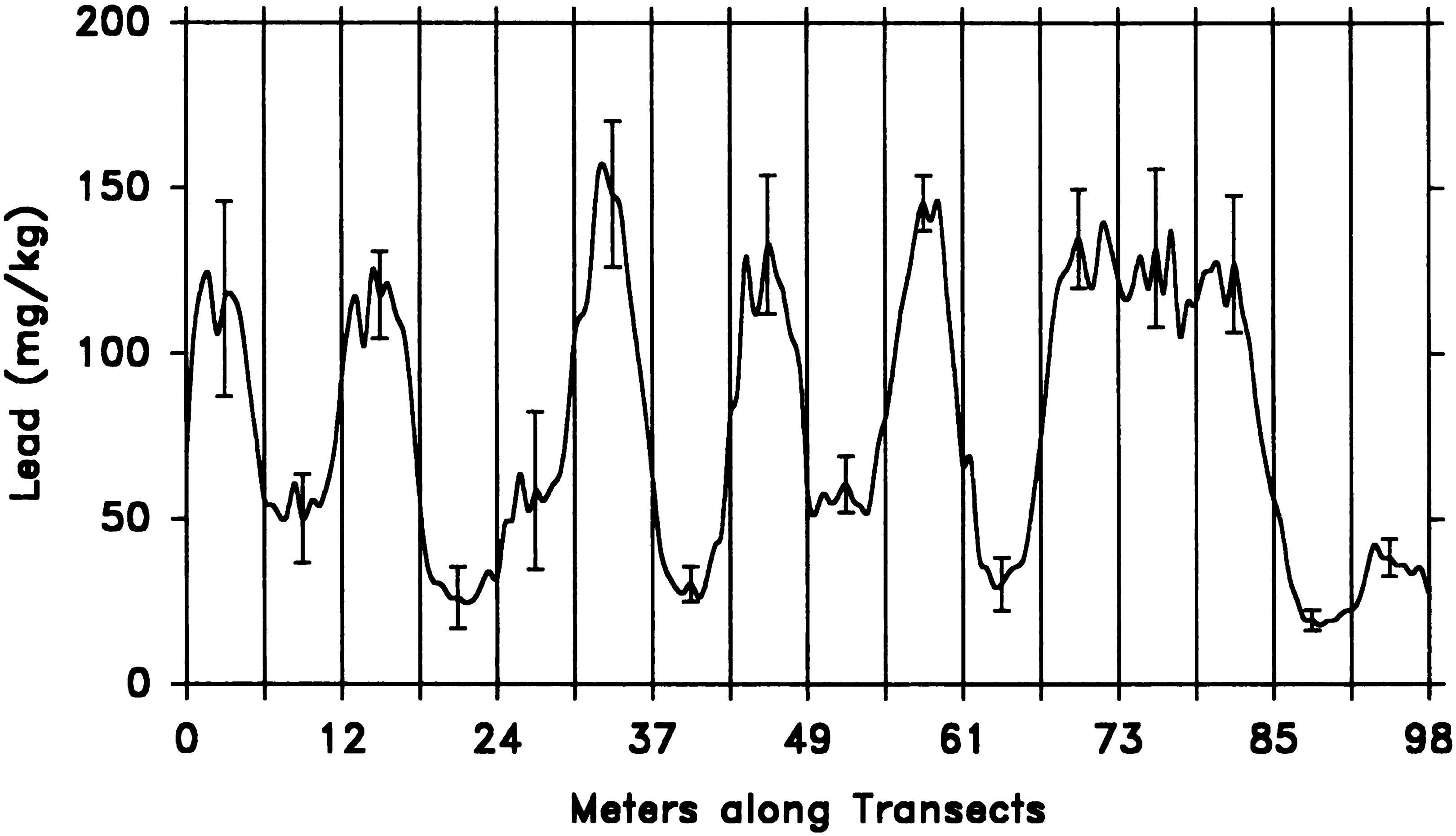

Figure 6. Average total Pb concentrations (with error bars representing standard deviations) found in surface samples collected along three transects across sludge-treated plots.

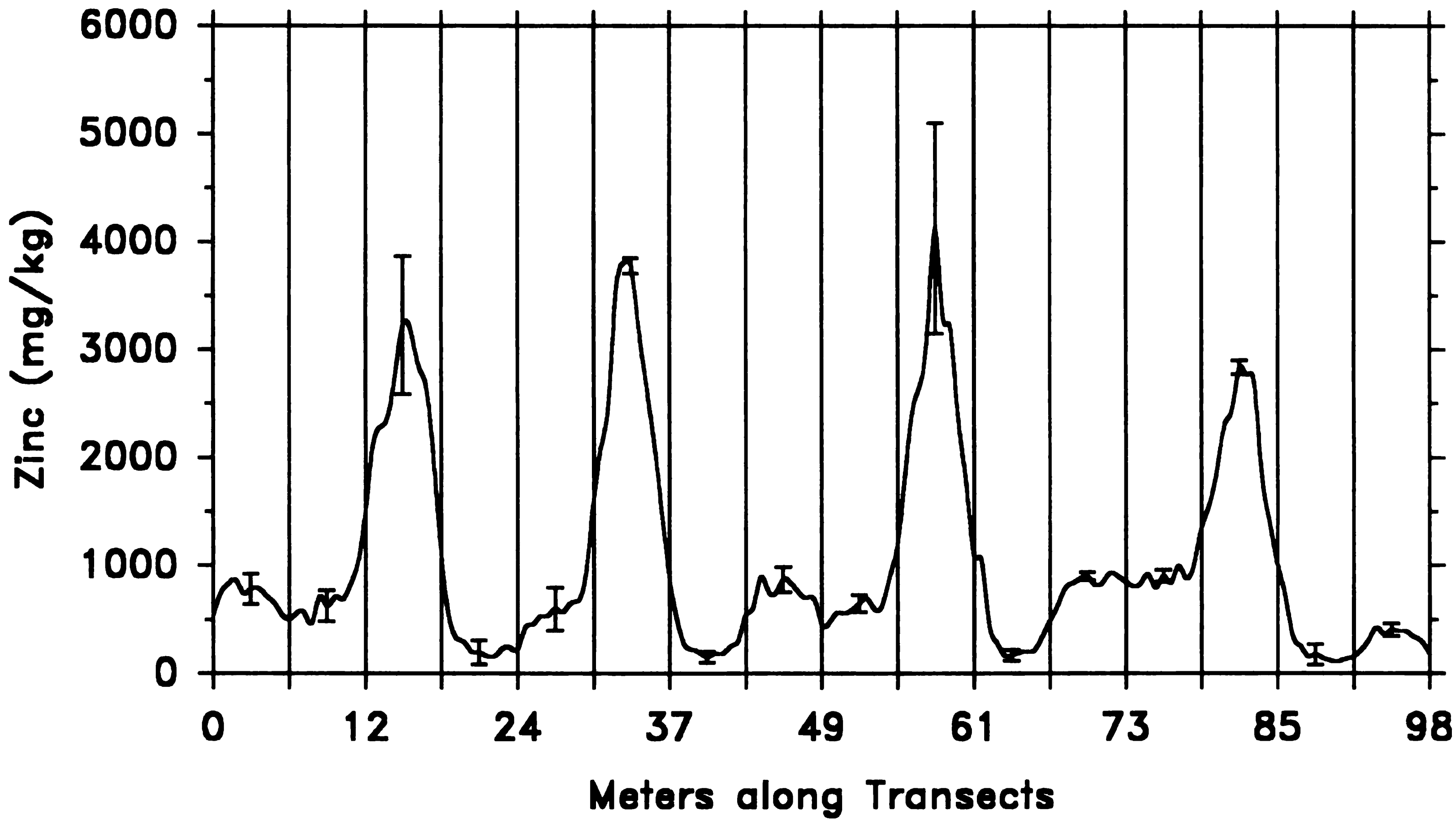

Figure 7. Average total Zn concentrations (with error bars representing standard deviations) found in surface samples collected along three transects across sludge-treated plots.

out of the plot area and as untreated soil came into the plot from neighboring plots and boundaries. McGrath and Lane (1989) were able to account for as much as 80% of the elements applied in their own experiment by modeling this dispersion using a technique described by Sibbesen et al. (1985) and Sibbesen and Anderson (1985).

Vertical Movement of Trace Elements

The middle of each plot was chosen as the most appropriate location to sample the soil profile since the 1989 sampling along the transects indicated that the trace element concentrations in the middle of each plot were least influenced by bordering treatments. Table 12 lists total elemental concentrations found in soil samples at different depths. Figures 8 to 13 depict the results graphically for Cd, Cr, Cu, Pb, Ni, and Zn, respectively. Included in both the table and the figures are the metal concentrations found in a Metea sandy loam soil that was collected in the spring of 1991 from an offsite area proximal to the study plots. Because the control plots may have been contaminated by soil from adjacent treated plots due to soil movement, we assumed that this offsite sample would represent the treated area prior to any sludge applications.

Little or no leaching of the trace elements occurred below the depth of tillage. Equal or lower concentrations of the elements occurred in the 15 to 30 cm layer compared with the 0 to 15 cm surface horizon. This likely was due to the incorporation of the sludges and subsequent plowing and tillage that may have been as deep as 20 to 25 cm (Pierzynski, 1985) but not as deep as the 30-cm sampling depth. Soil samples taken below the 15 to 30 cm horizon had trace element

Table 12. Concentrations of Cd, Cr, Cu, Ni, Pb, and Zn in soil profile under sludge experiment.

Depth	Cd†	Cr	Cu	Ni	Pb	Zn
- cm -			mg kg^{-1}			
			Control			
0-15	1.6±0.2	120± 10	51± 7	56± 8	59± 6	230± 20
15-30	1.7±0.1	67± 7	35± 7	34± 4	50± 3	130± 20
30-46	1.4±0.2	49± 11	16± 5	23± 6	50± 7	47± 8
46-72	1.8±0.2	56± 10	19± 3	27± 6	63±10	47± 6
>72	1.9±0.2	55± 8	19± 2	27± 3	76± 6	48± 3
			Treatment 1			
0-15	8.0±1.0	540± 80	110±20	97±19	82±10	480±100
15-30	6.0±1.3	360±120	110±60	95±54	73±15	390±150
30-46	1.8±0.5	81± 43	22± 8	26± 6	51± 3	69± 32
46-72	1.9±0.2	60± 8	19± 3	27± 4	59± 9	50± 2
>72	1.9±0.6	63± 5	21± 2	29± 3	77±19	48± 4
			Treatment 2			
0-15	4.5±0.3	590± 30	290±10	430±50	170±10	2500±200
15-30	4.6±0.4	490± 40	270±20	390±40	140±20	1970± 50
30-46	1.7±0.1	58± 8	21± 3	33± 3	54± 5	72± 14
46-72	2.2±0.2	64± 4	20± 3	30± 3	69±14	63± 2
>72	2.1±0.3	62± 3	23± 2	32± 3	78±13	67± 12
			Treatment 3			
0-15	4.8±0.6	850±120	520±60	440±40	180±20	820±130
15-30	5.0±0.2	680± 70	430±80	375±70	140±10	760±120
30-46	2.1±0.4	160±111	85±66	71±45	71±18	170±110
46-72	1.5±0.1	51± 2	18± 1	24± 2	52± 4	43± 5
>72	1.9±0.2	59± 8	25± 5	31± 4	80±10	49± 5
lsd‡	0.6	62	38	37	13	100
			Offsite§			
0-15	0.5±0.3	26± 1	7± 0	11± 0	40± 1	45± 6
15-30	0.8±0.5	27± 1	7± 0	11± 1	42± 1	41± 12
30-46	0.8±0.0	24± 0	5± 0	10± 0	34± 1	31± 8
46-72	0.9±0.0	31± 2	8± 0	14± 1	41± 0	31± 5
>72	1.7±0.0	67± 10	18± 0	29± 5	69± 1	59± 9

†Cd concentrations less than 2.5 mg kg^{-1} were below the linear analytical detection limits of DCP-AES.
‡Fisher's (protected) least significant difference at p = 0.05.
§Included for general comparisons but not in statistical analysis.

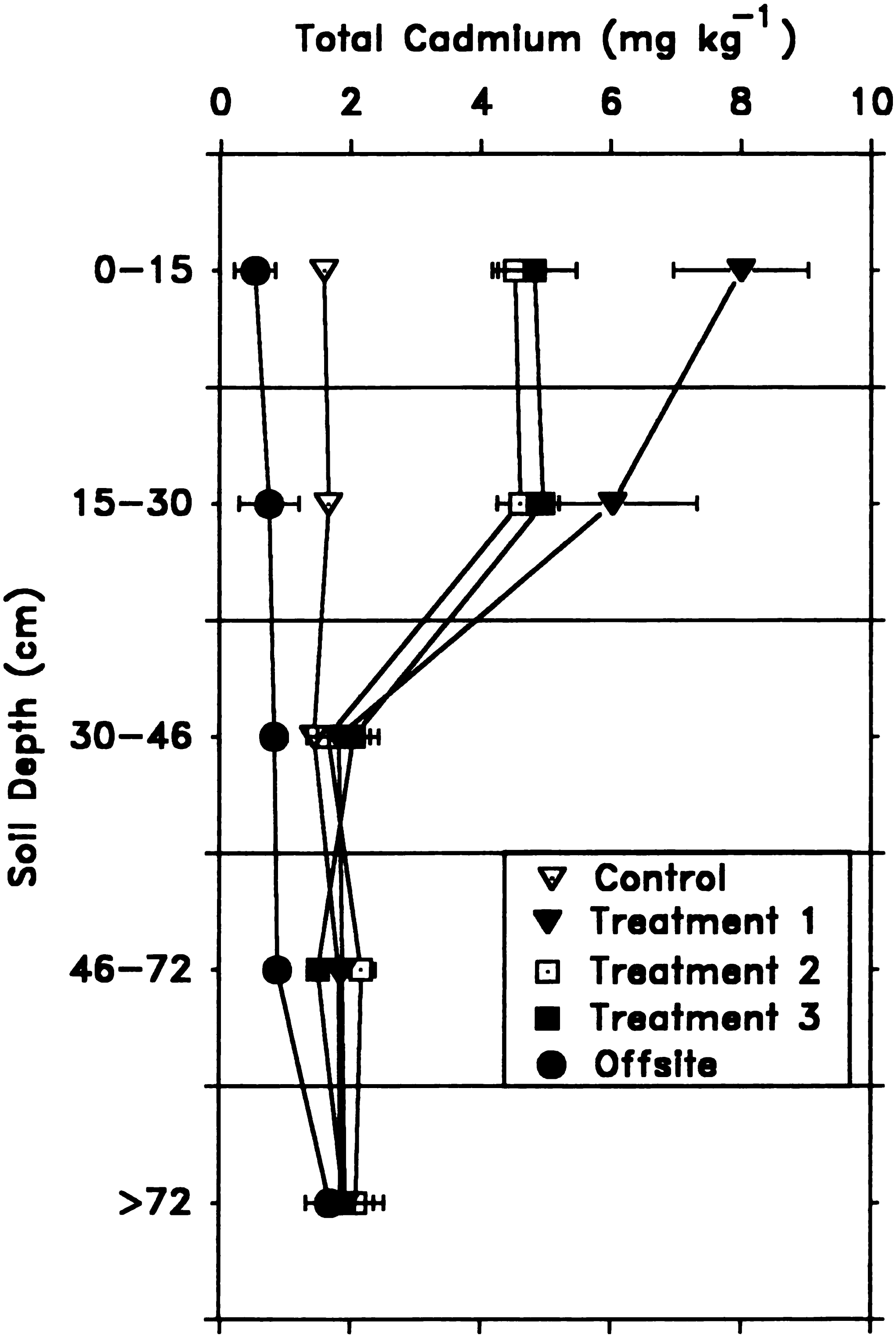

Figure 8. Total soil Cd in profile under four sludge application treatments.

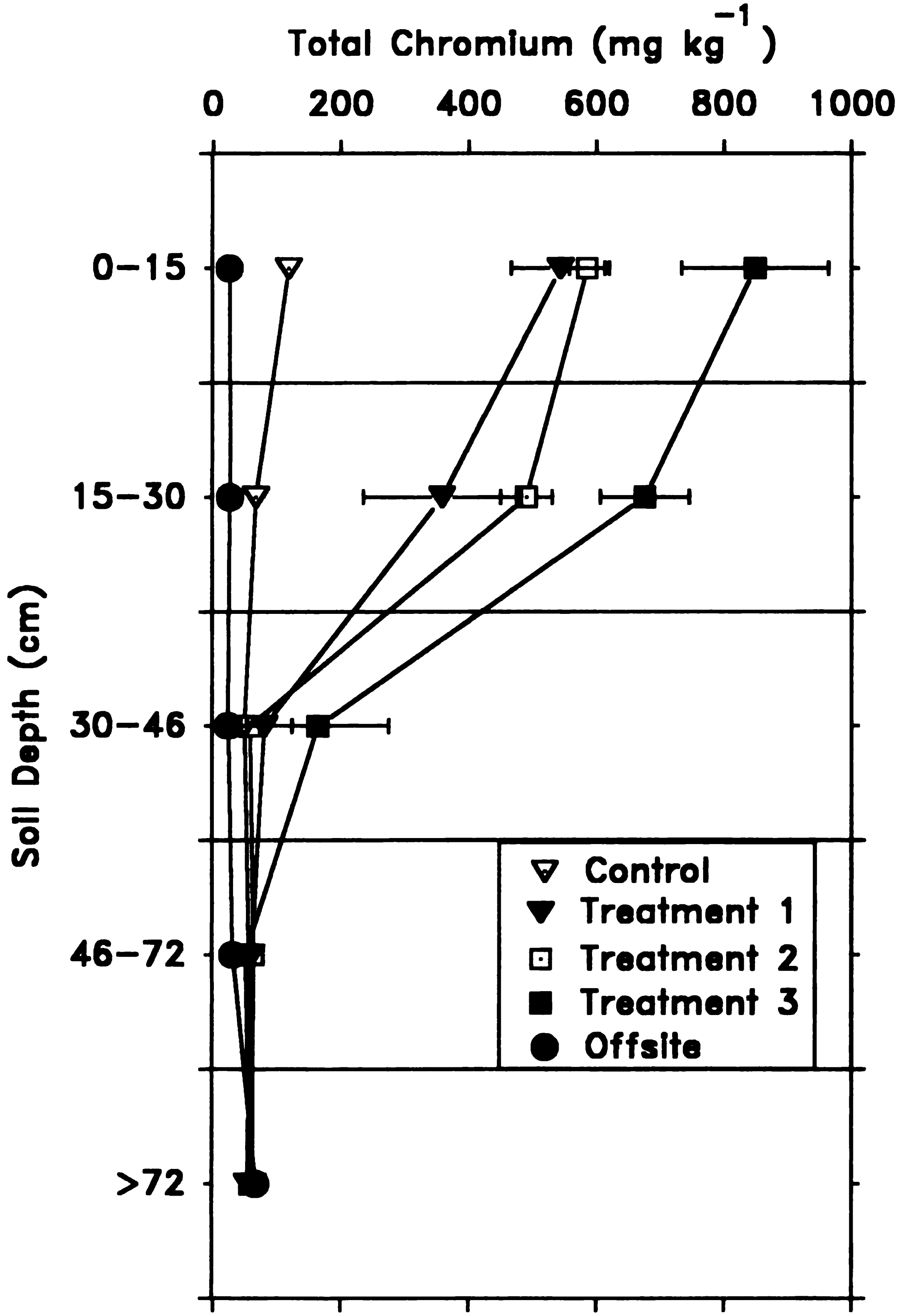

Figure 9. Total soil Cr in profile under four sludge application treatments.

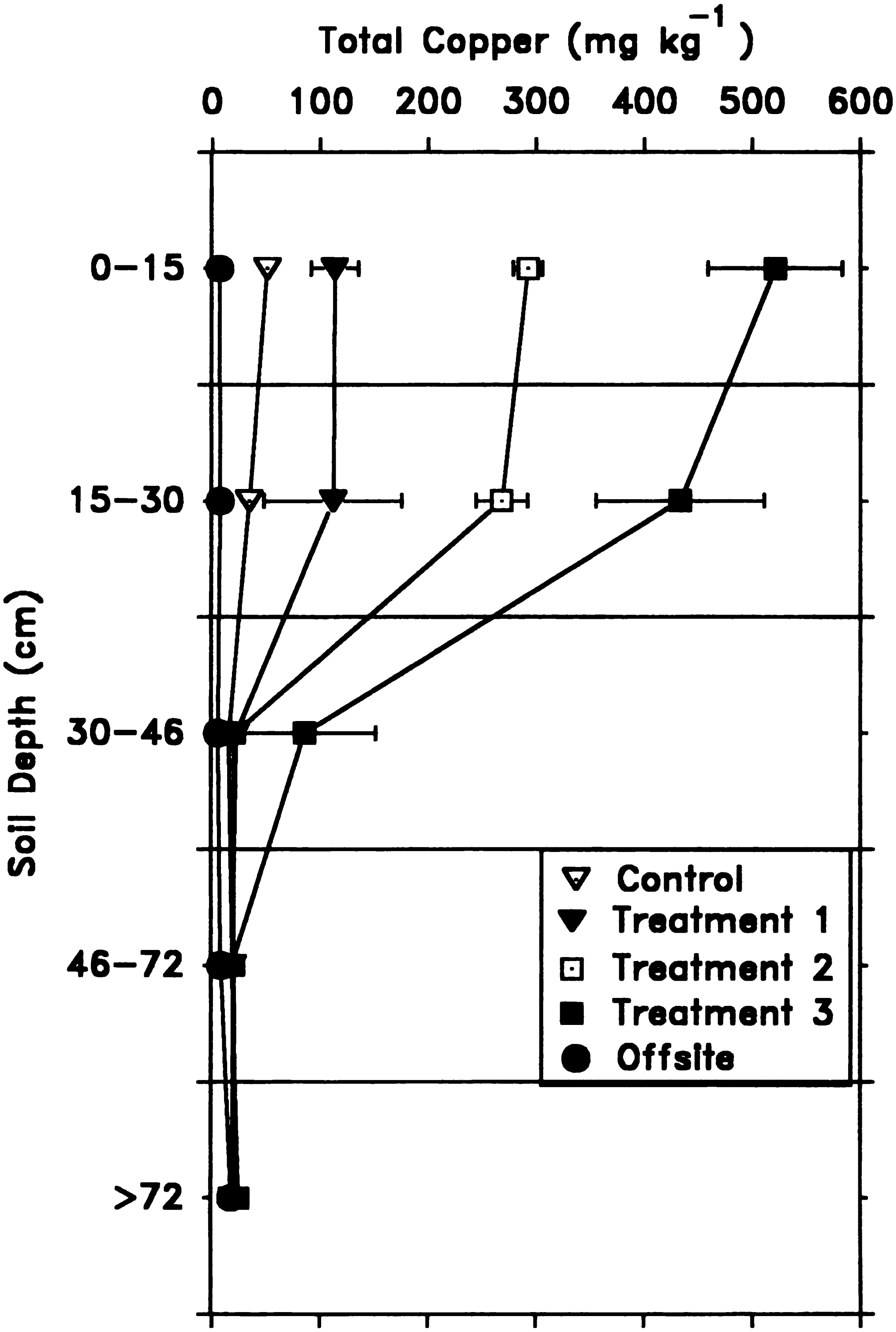

Figure 10. Total soil Cu in profile under four sludge application treatments.

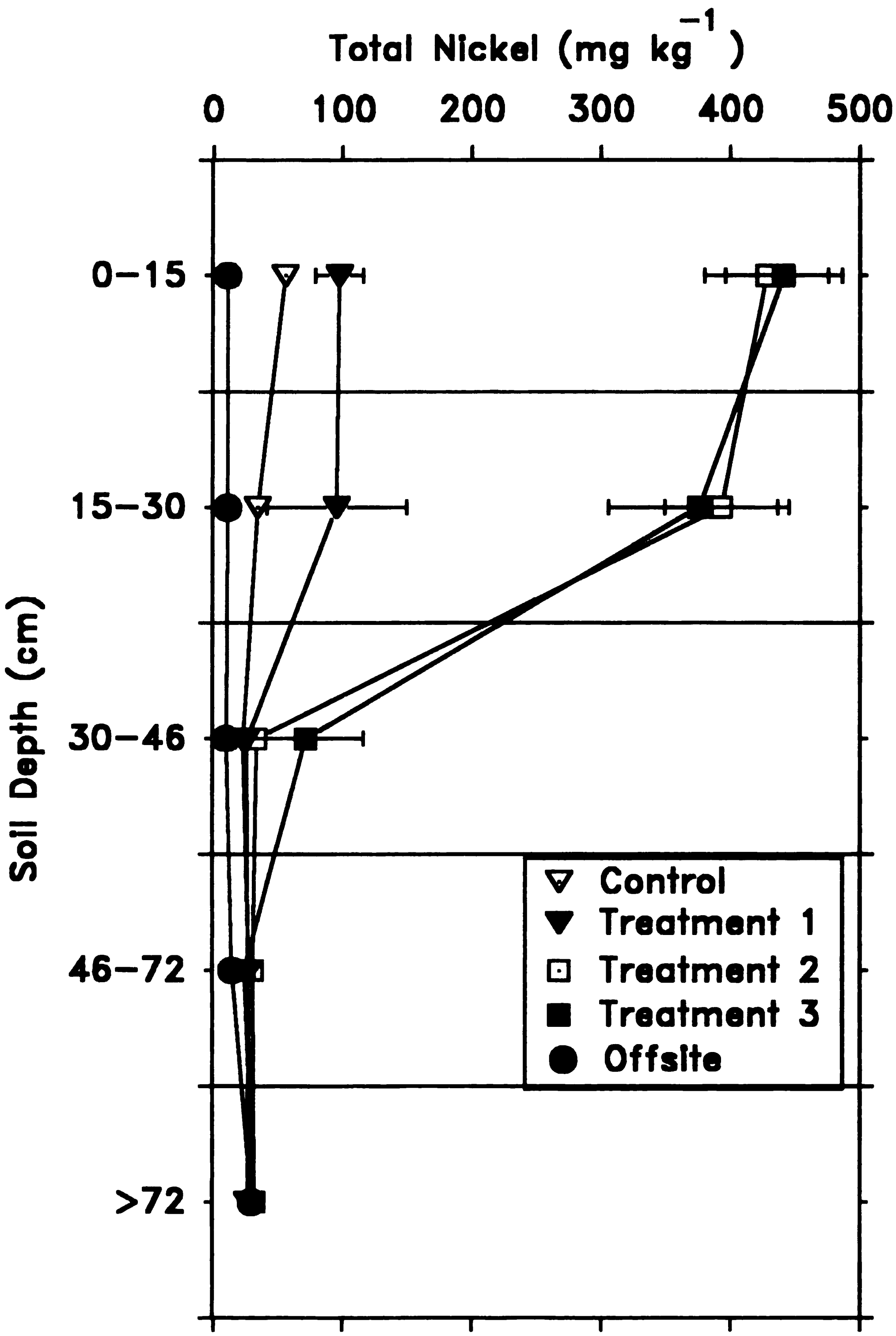

Figure 11. Total soil Ni in profile under four sludge application treatments.

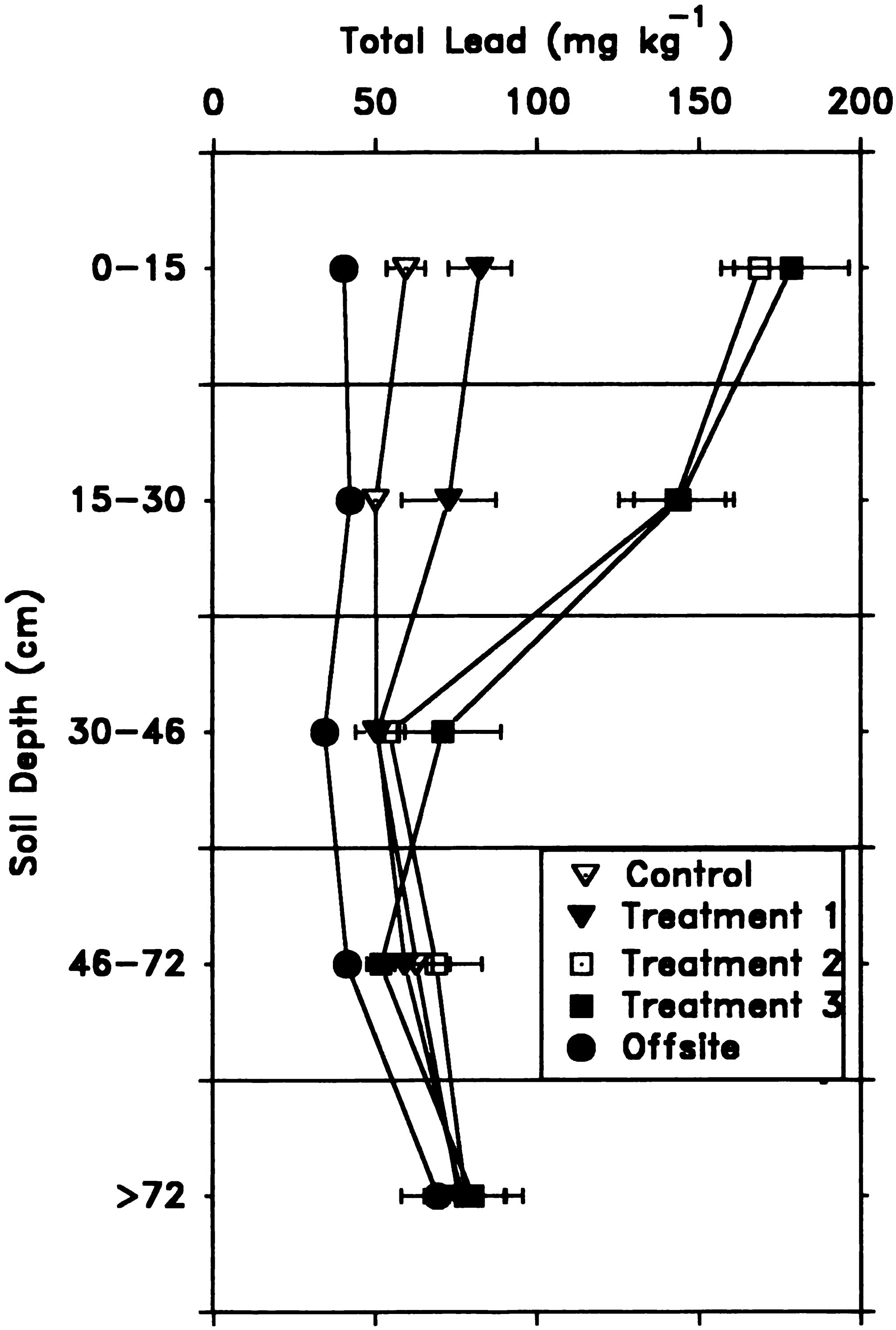

Figure 12. Total soil Pb in profile under four sludge application treatments.

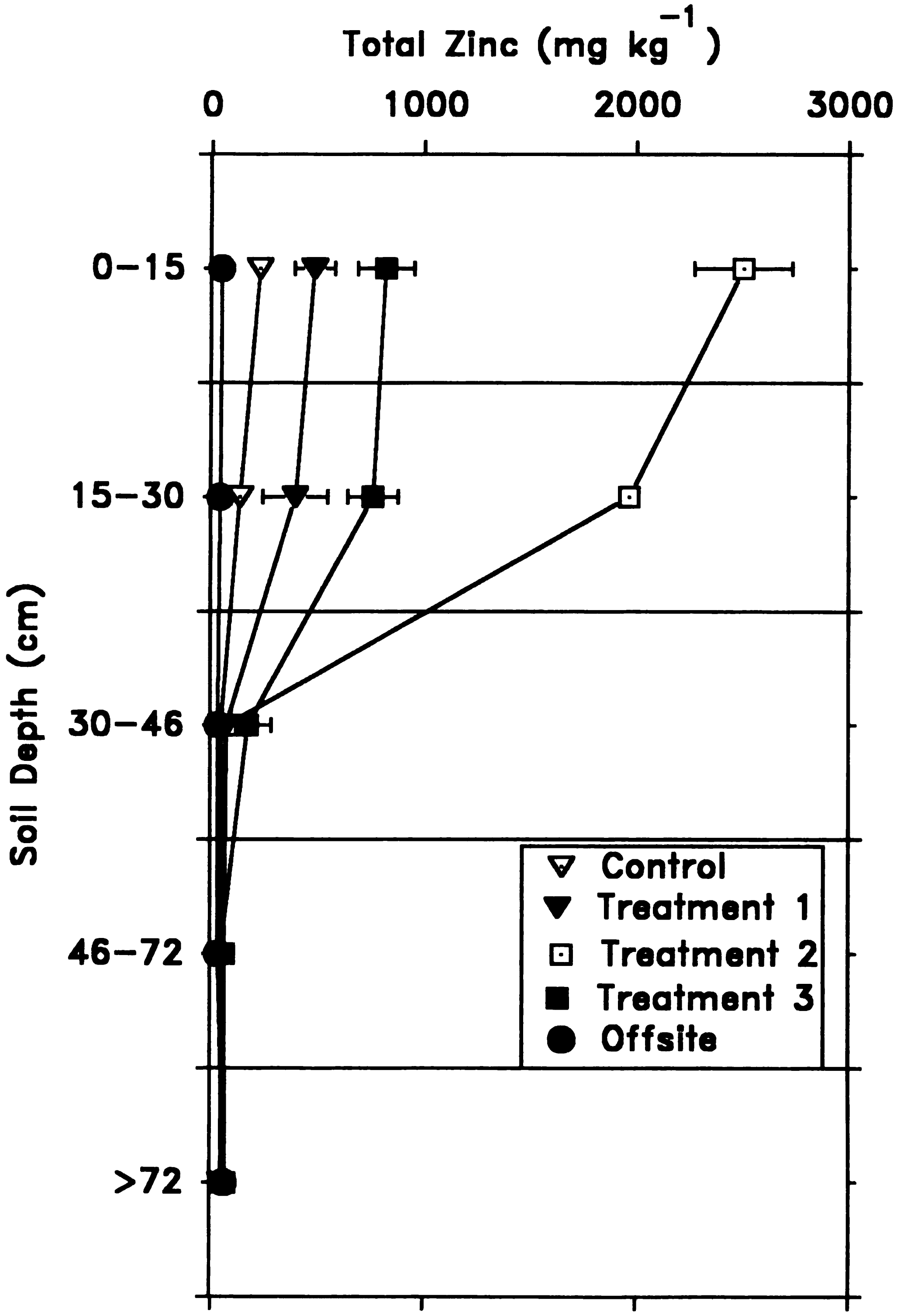

Figure 13. Total soil Zn in profile under four sludge application treatments.

concentrations not significantly different from those of the control plots or in the offsite sample. One exception was the apparently lower Pb concentrations found in the depths deeper than 30 cm in the offsite soil compared to the Pb concentrations found at similar depths below the sludge-treated soils or even below the control soils. Concentrations of Pb in subsoils of the control plots were similar to sludge-treated soils.

The background levels of Pb in the subsoil under the experimental plots must be naturally greater than in the subsoil at the offsite area. If Pb were leaching, higher concentrations of Pb should be found in subsurface horizons for soils receiving high metal loadings compared to that found in control subsoils. Instead, no differences were found in the depths deeper than the 30-cm horizons between the treated plots and the control.

Mass Balance Calculations of Trace Elements

In order to calculate sludge loading rates on the experimental plots, we assumed that all sludge applications were incorporated to 23 cm and that the bulk density of the soils throughout the years of application was 1.4 g cm^{-1}. These values were those determined in a study by Pierzynski (1985) in which he calculated the mass balance of Mo applied in sewage sludge to a Metea sandy loam on an experimental area proximal to this study. Depth of tillage measurements were made by visual observation of sludge particles in soil cores. This resulted in a hectare furrow slice with a weight of 3.2×10^6 kg.

Examining the amount of each element remaining in surface soils as a percentage of the amount applied (correcting for background levels of each element using data from the offsite soil samples) may give a measure of the levels of trace elements unrecovered. Table 13 lists mass balance calculations as recovery percentages based on surface soil samples collected in 1989 from the middle 120 cm of each plot along the three transects. This table also lists mass balance calculations as percent recoveries using the surface soils collected from the middle of each plot in 1990.

Mass balance calculations resulted in average recoveries of trace elements ranging from 45 to 120% for samples collected in 1989. In 1990, average recoveries ranged from 49 to 150%. Calculated recoveries of applied Cd, Cr, Cu, Ni, Pb, and Zn averaged over the three treatments were 69, 82, 63, 80, 61, and 100% in 1989 and 77, 110, 69, 86, 86, and 110% in 1990, respectively. These calculations were highly variable among the six trace elements. Sewage sludge applications resulted in a heterogenous mixture in the soil for reasons that included the highly variable nature of sludge composition, difficulty of getting a uniform application over large plots, and soil movement due to tillage. The lack of total accuracy when sampling both sludges and soils in order to make these kinds of calculations must not be underestimated.

The extent to which mass balance calculations differed between 1989 and 1990, however, also was significant, especially in soils of Treatment 3. This showed the effect that sampling alone can have when making these types of calculations, regardless of the accuracy of application and application data of the trace elements. This variability was reduced by restricting the sampling to the middle of each plot. However, sampling time and method also will affect the outcome of results.

Table 13. Mass balance calculations of percentage of applied trace elements recovered in 1989 and 1990 from surface soils.

	Cd	Cr	Cu	Ni	Pb	Zn
	Percent of Total Applied					
	1989 Soil Sampling†					
Treatment 1	57±13	121±25	71±16	94±22	57±47	120±30
Treatment 2	69± 6	48± 5	45± 3	67±13	62±10	90±12
Treatment 3	80± 4	77± 2	74± 1	78± 2	63± 5	94± 5
Average	69±12	82±34	63±16	80±18	61±26	100±20
	1990 Soil Sampling‡					
Treatment 1	61±9	150±30	83±19	110±27	100±50	120±30
Treatment 2	76±5	63± 2	49± 5	69± 9	69± 7	85± 8
Treatment 3	100±5	100± 3	79± 3	84± 3	88± 3	110± 7
Average	77±18	110±40	69±20	86±23	86±32	110±20

†Collected from the middle 120 cm of each plot along each of three transects. Each sample was individually extracted.
‡Composite samples from the middle of each plot.

Because of the potential errors associated in mass balance calculations, percent recoveries that have been calculated in this and other studies that deviate substantially from 100 should reasonably be expected. Examining the relatively few ways in which trace elements are lost from a soil (e.g., plant uptake, soil movement via tillage, water and wind erosion, and deep leaching) and in what components they accumulate may be better approaches than to rely on mass balance calculations.

SUMMARY AND CONCLUSIONS

Municipal sludges containing Cd, Cr, Cu, Ni, Pb, and Zn were applied to research plots beginning in 1977 and continuing through 1986. Treatments included three sets of plots to which different amounts of municipal sewage sludges from various locations in Michigan were applied. Total elemental analysis of soils collected in 1989 and 1990 indicated some lateral movement of trace elements associated with physically moving soil particles with agronomic operations. These elements, however, have not moved below the 15 to 30 cm sample depth. An accounting of elements applied in the sewage sludges indicated that greater than 90% of the trace elements applied can be recovered in surface soil samples.

On average, only 50% or more of the applied trace elements from plots of Treatment 2 and greater than 77% from plots of Treatment 3 could be recovered. This indicated the difficulty of accounting for the elements applied. Movement of trace elements from the plots due mostly to soil movement and occurred primarily at plot boundaries. Concentration of metals unaccounted for resulted from inherent inaccuracies of techniques and records, and the movement of soil into or out of the plots, not from leaching.

LIST OF REFERENCES

LIST OF REFERENCES

Chang, A.C., A.L. Page, and F.T. Bingham. 1982. Heavy metal absorption by winter wheat following termination if cropland sludge applications. J. Environ. Qual. 11:705-708.

Chang, A.C., J.E. Warneke, A.L. Page, and L.J. Lund. 1984. Accumulation of heavy metals in sewage sludge-treated soils. J. Environ. Qual. 13:87-91.

Emmerich, W.E., L.J. Lund, A.L. Page, and A.C. Chang. 1982. Movement of heavy metals in sewage sludge-treated soils. J. Environ. Qual. 11:174-178.

Kelling, K.A., D.R. Keeney, L.M. Walsh, and J.A. Ryan. 1977. A field study of the agricultural use of sewage sludge: III. Effect on uptake and extractability of sludge-borne metals. J. Environ. Qual. 6:352-358.

McGrath, S.P. 1984. Metal concentrations in sludges and soil from a long-term field trial. J. Agric. Sci. 103:25-35.

McGrath, S.P. 1987. Long-term studies of metal transfers following application of sewage sludge. *In* P.J. Coughtrey, M.H. Martin, and M.H. Unsworth (eds.) Pollutant transport and fate in ecosystems. Special Pub. No. 6 British Ecological Society. Blackwell Scientific Pubs, London, UK.

McGrath, S.P., and P.W. Lane. 1989. An explanation for the apparent losses of metals in a long-term field experiment with sewage sludge. Environ. Pollu. 60:235-256.

Pierzynski, G.M. 1985. Agronomic considerations for the application of a molybdenum-rich sewage sludge to an agricultural soil. Master thesis. Michigan State University, East Lansing, MI.

Ritter, C.J., S.C. Bergman, C.R. Cothern, and E.E. Zamierowski. 1978. Comparison of sample preparation techniques for atomic absorption analysis of sewage sludge and soil. Atomic Absorption Newsletter. 17(4):70-72.

SAS. 1985. SAS user's guide: Statistics, version 5 ed. SAS Institute Inc., Cary, NC.

Shuman, L.M. 1979. Zinc, manganese, and copper in soil fractions. Soil Sci. 127:10-17.

Sibbesen, E., and C.E. Anderson. 1985. Soil movement in long-term field experiments as a result of cultivations. II. How to estimate the two-dimensional movement of substances accumulating in the soil. Experimental Agriculture. 21: 109-117.

Sibbesen, E., C.E. Anderson, S. Anderson, and M. Flenstde-Jensen. 1985. Soil movement in a long-term field experiments as a result of soil cultivations. I. A model for approximating soil movement in one horizontal dimension by repeated tillage. Experimental Agriculture. 21: 101-107.

Williams, D.E., J. Vlamis, A.H. Pukite, and J.E. Corey. 1980. Trace element accumulation, movement, and distribution in the soil profile from massive applications of sewage sludge. Soil Sci. 129:119-132.

Williams, D.E., J. Vlamis, A.H. Pukite, and J.E. Corey. 1984. Metal movement in sludge-treated soils after six years of sludge addition: 1. Cadmium, copper, lead, and zinc. Soil Sci. 137:351-359.

CHAPTER THREE:

CHEMICAL FRACTIONATION AND PLANT UPTAKE OF Cd, Cr, Cu, Ni, Pb, and Zn IN A SANDY LOAM SOIL FROM THE APPLICATION OF MUNICIPAL SEWAGE SLUDGES

ABSTRACT

Surface soils to which municipal wastewater sludges were applied from 1977 to 1986 were sequentially extracted and trace elements were measured in each of eight fractions. Cadmium, Cu, and Zn resided primarily in the exchangeable and acid-soluble fractions, Cr in the organic and Fe oxide fractions, Ni in the acid-soluble and Fe oxide fractions, and Pb in the residual fraction. Overall, two of the three sludge treatments had significantly greater yields of corn grain (*Zea mays* L.) and sorghum-sudangrass (*Sorghum bicolor* L. Moench X *S. Sudanese* P. Stapf.), whereas soybean (*Glycine max* L.) grain yields on treated plots were equal to or less than those of controls, due to phytotoxic concentrations of one or more trace elements in the soil. Plant uptake of trace elements was variable from year to year, plant part, and crop. Plant samples of sludge treated plots collected between 1985 and 1990 had greater concentrations of Cu, Ni, and Zn in corn diagnostic tissue; Cu and Ni in sorghum-sudangrass; Ni and Zn in corn grain; and Ni in corn stover and soybean grain when compared with controls. Results of the sequential extraction and plant analysis suggested that Cd, Ni, and Zn

continued to be environmentally available, whereas Cr and Cu were relatively less available, and Pb was not environmentally available. Soil test methods for trace elements that were plant available generally correlated well with the most labile soil fractions (i.e., water-soluble, exchangeable, and acid-soluble). Only AB-DTPA and HCl correlated well with acid-soluble Cr. None correlated well with Pb, an indication of its limited environmental availability in soils. Toxicity Characteristic Leaching Procedure (TCLP) was inappropriate as a soil test method to access concentrations of trace elements that were toxic to plants.

INTRODUCTION

Chemical fractionation techniques have been used to sequentially extract different forms of trace elements in soils, sediments, sludges, and dissolved solids in natural waters. Several research efforts (Emmerich et al., 1982a; Sims and Kline, 1991; Sposito et al., 1982) have used the fractionation method developed by Stover et al. (1976) in which the forms of metals in wastewater sludge were evaluated. Sposito et al. (1982) found the application of sewage sludge to two arid-zone soils tended to reduce the residual fraction (determined using concentrated HNO_3) and to increase the organic (NaOH) and carbonate (EDTA) fractions of Cd, Cu, Ni, Pb, and Zn. The application of sludge resulted in forms of trace elements in soils that were more chemically labile compared to those occurring prior to sludge application. This change to more labile forms may in turn make these trace elements more readily available to plants. At the highest rate of sludge application, Zn, Cd, and Pb were predominately in carbonate forms; Cu in organic; and Ni in residual (Sposito et al., 1982). The results indicated that the chemical behavior of Zn, Cd, and Pb in the arid soils may be similar, whereas that of Ni and Cu differed from the other three and from each other. The percentage of total metal content in the two most labile chemical forms [e.g., exchangeable (extracted in 0.5*M* KNO_3) and sorbed (extracted using water)] was low, averaging from 1 to 4% for all of the metals regardless of the type of soil or sludge applied or sludge application rate.

Sims and Kline (1991) used the same fractionation technique to determine the soil components and plant availability of Cd, Cr, Cu, Ni, Pb, and Zn in three acidic Atlantic coastal plain soils amended with composted sewage sludge. With the exception of Cd, amending the soils significantly altered the distribution of the elements among the various soil fractions. The percentages of Cr and Ni in the organic and carbonate fractions increased, while that in the residual fraction decreased, although actual concentrations did increase. The majority of soil Pb was found in the carbonate fraction. Application of the sludge increased soil Cu in all fractions with the greatest increases occurring in the organic fraction. Zinc was primarily found in the carbonate and residual fractions. Sludge application generally resulted in a greater concentrations of soil Zn in organic, carbonate, and residual fractions, similar to that observed with Cr and Pb.

The results reported by Sims and Kline (1991) differed somewhat from those of Sposito et al. (1982) in that they found a greater percentage of soil Cd in the exchangeable and adsorbed fractions. Sposito et al. (1982) found that less than 1% of the total soil Cd was in these two fractions. Sims and Kline (1991) attributed this difference to the lower pH of the soils they used compared to those used by Sposito et al. (1982). Results were consistent between the two studies for Cr, Ni, Pb, Cu, and Zn.

The sludge had little effect on concentrations of Cd, Cr, or Pb in either wheat (*Triticum aestivum* L.) and soybean (*Glycine max* L.) that Sims and Kline (1991) grew in the greenhouse, but consistently increased

Cu, Ni, and Zn in vegetative tissues of wheat and soybean, and Ni and Zn in soybean grain. With the exception of Zn, consistent correlations between total soil metal content or individual metal fractions and plant metal concentrations or uptake were not observed. However, significant multiple regression models between soil metal fractions and pH and metal concentrations in the two crops were obtained for Ni, Cu, and Zn.

Emmerich et al. (1982a) also used the fractionation technique developed by Stover et al. (1976). Their work, however, was a leaching study performed on soils probably more similar to those used by Sposito et al. (1982) than by Sims and Kline (1991), i.e., neutral to alkaline pH rather than acidic. The results reported by Emmerich et al. (1982a) were similar to those reported by Sposito et al. (1982). Most of the elements were found in the organically bound, carbonate, or residual forms. Except for Cd, the soils contained more than 65% of each metal in the residual form. Cadmium, Ni, and Zn appeared to be shifting to the residual component from more labile forms found in the sewage sludge. The chemical forms of Cu had not changed significantly during the study from being primarily organically bound. Nickel was the only metal that showed any appreciable percentage in the exchangeable form. The occurrence of metals in the stable organically bound, carbonate, and residual forms in the sludge, coupled with a shift toward the more stable form after sludge incorporation, contributed to the lack of metal movement in the soil profile. Chromium and Pb were not included in their study.

Chang et al. (1984a) extended some of the work of Sposito et al. (1982) using arid soils and found that without sludge, most of the trace elements were either in the carbonate (Cd and Pb) or the residual (Cr, Cu, Ni, and Zn) forms. After seven years of sludge application, the carbonate and organic forms in the soil became the most prevalent solid-phases for Cu, Ni, and Zn. The distribution patterns of Cd, Cr, and Pb, however, were not significantly affected by the amounts of sludge added. The accumulation pattern of solid-phase elements in sludge-treated soils did not appear to change during the growing season following sludge application. Three years after termination of sludge application, the distribution pattern of heavy metals in sludge-affected soil remained the same.

Chang et al. (1984a) also addressed the uptake of Cd and Zn into barley (*Hordeum vulgare* L.) at the same time they were sampling the soil. They found that the concentrations of these two elements were consistently higher in sludge-treated soils than those in the non-sludge control. Concentrations of Cd and Zn in each extracted fraction did not change appreciably in soil samples taken during the growing season, so that the changes seen in plant concentrations of Cd and Zn were related exclusively to plant growth and development. Total uptake of trace elements by the barley amounted to less than 1% of that applied in the sludge (Chang et al., 1984b)

Hickey and Kittrick (1984) sequentially extracted Cd, Cu, Ni, and Zn in three dissimilar soils and a harbor sediment (pH 6.0, 7.0, 4.6, and 6.9) using a scheme developed by Tessier et al. (1979). The greatest amount of Cd and an appreciable amount of the Zn were found in the exchangeable fraction (1.0*M* $MgCl_2$). Fe and Mn oxides [0.04*M* $NH_2OH \cdot HCl$ in 25% (v/v) acetic acid] and residual (HF and $HClO_3$) fractions were the most important for the soils and sediment examined and contained high levels of Cd, Cu, Ni, and Zn. The carbonate (1.0*M* NaOAc) fraction, however, was of about equal importance as the oxide fraction in binding metals for samples containing appreciable quantities of inorganic carbon. Cu was the only element significantly associated with the organic (H_2O_2) fraction.

Chang et al. (1982), working with a fine-textured, calcarious soil in California, found higher yields of wheat (*Triticum* spp. var. Anza) on plots 3 and 4 years following the termination of sludge applications compared to control plots. They attributed the yield increase to the residual N, P, micronutrients, and organic matter from the sludge treatment. Although the treated soils received substantial quantities of Cu, Ni, and Pb, elevated plant-tissue concentrations of these elements were not found in the wheat grain and straw. The plants did, however, accumulate greater amounts of Cd and Zn from the sludge-treated than from non-treated soils.

Dowdy et al. (1978) also saw the benefits of the land application of sewage sludge on the yields of edible snap bean (*Phaseolus vulgaris* L. var. Tendergreen) in their study on a sandy soil. Yields increased as rates of sludge application increased, and often exceeded those of a well-managed, fertilized control. Both Zn and Cu contents of edible tissue increased as rates of sludge application increased, and reached an apparent maximum value from which they did not decrease once sludge applications ended. Cd levels in edible tissue did not respond directly to sludge applications and never exceeded 0.1 mg Cd kg^{-1} tissue.

Work by Kelling et al. (1977) found that the application of sewage sludge on a sandy loam and a silt loam in Wisconsin increased the concentrations of Cu, Zn, Cd, and Ni in the vegetative tissue of rye (*Secale cereale* L.), sorghum-sudangrass (*Sorghum bicolor* L. Moench X *S.*

sudanese P. Stapf.), and corn (*Zea mays* L.). Except for Zn, however, the additions had relatively little effect on the elemental content of corn grain. Chromium did not accumulate in the tissue or grain.

Soil testing methods have been proposed and used to help assess the plant availability of trace elements, including "heavy metals" such as Cd, Ni, and Pb. These efforts have been concentrated in the use of five different general reagents; salt solutions, dilute weak acids or water, strong acids, dilute strong acids, and chelating agents. Measuring an exchangeable fraction using a salt solution, as has been done with macronutrients (Ca, Mg, K), has not been found to be a reliable index of plant uptake. The amounts extracted are too small to be of much predictive value. Dilute weak acids or water have been used to imitate the plant root in its release of organic acids to facilitate micronutrient uptake. Unfortunately, these methods also do not extract large enough amounts of the elements to mimic plant uptake or approach the soil's capacity to replenish the nutrient supply of the soil solution (capacity factor). Strong acids have also been used but they often extract too much of the trace elements and do not necessarily correlate well with plant uptake or the capacity factor of soils. Using dilute solutions of strong acids has often improved results. Chelating agents offer another alternative for assessing a soil's ability to supply nutrients to a crop. Of these different types of reagents, dilute strong acids and chelating agents have proven to be the most effective for evaluating the micronutrient status of a wide range of soils.

Baker and Amacher (1982) listed three methods that may be of value to determine the availability of Cd, Cu, Ni, and Zn: the DTPA method

(Lindsay and Norvell, 1978), 0.1*M* HCl (Nelson et al., 1959), and the double-acid (0.05*M* HCl + 0.25*M* H_2SO_4) extraction method (Sabbe, 1980). Risser and Baker (1990) included another testing procedure to measure readily available Cr, which used 0.01*M* KH_2PO_4 (Bartlett and James, 1979, 1988; James and Bartlett, 1983). A variety of extractants have been tried for their ability to predict or describe Pb uptake by plants. The most popular reagents were 1*M* NH_4OAc, three different concentrations of EDTA, and 2.5% acetic acid (Burau, 1982). Other extractants for Pb include various concentrations of HNO_3 (John, 1972), 0.1*M* HCl (Misra and Pandey, 1976), and soil P tests (Miller et al., 1975).

Although not a soil testing method, the Toxicity Characteristic Leaching Procedure (TCLP), a U.S. Environmental Protection Agency testing protocol (Federal Register, 1990a), may be used to determine if contaminated soils should be classified as "hazardous." The method was promulgated to identify those "wastes" that are hazardous because they may leach significant concentrations of specific toxic constituents to groundwater when placed in a landfill. Materials so classified are subject to regulation under subtitle C of the Resource Conservation and Recovery Act (RCRA). A soil that has become contaminated, which results in it being classified as a "hazard" based on TCLP, may be subject to the same regulatory control as hazardous wastes.

Previously (Chapter One), the techniques that scientists have used to determine chemical fractions in which trace elements reside in soils were reviewed, examining the theoretical and experimental evidence for method selection. One objective of this chapter is to examine the changes in the soil chemical fractionation of Cd, Cr, Cu, Ni, Pb, and Zn that resulted from the application of municipal wastewater sludges to experimental field plots applied over ten years, four years after cessation of sludge application. Also, the uptake of these elements into the tissue of corn and sorghum-sudangrass and the grain of corn and soybean for a period of up to four years after sludge applications ended was studied. Finally, the results of four soil testing methods [AB-DTPA, EDTA-$Ca(NO_3)_2$, DTPA-TEA, and 0.1*M* HCl] and the TCLP were studied and compared with the sequential extraction of trace elements.

MATERIALS AND METHODS

Sample Collection

Municipal sewage sludges from different sources were applied from 1977 to 1986 to plots located on the Michigan State University Soil Science Farm, at the corner of Mt. Hope and Hagadorn Roads. Chapter Two summarizes the sludge and trace element loading rates and the experimental design used in the treatments and plot layout.

During the course of the experiment, several different crops were grown on the experimental plots, including corn (*Zea mays* L.), soybean (*Glycine max* L.), sorghum-sudangrass (*Sorghum bicolor* L. Moench X *Sorghum sudanense* P. Stapf.), and alfalfa (*Medicago sativa* L.). This paper reports on the grain yield and uptake of trace elements in corn grain, corn diagnostic leaf tissue sampled between tasseling and silking, and the whole plants of corn at harvest collected between 1985 and 1990. Plant material was hand harvested from the center of each plot, except for soybean grain and sorghum-sudan grass in 1988, which were harvested using machines. Corn diagnostic tissue samples collected at tasseling were comprised of a single leaf opposite and below the corn ear, All plant material was then dried at 104°C, ground using a Wiley mill, and stored in plastic bags.

Soil samples were collected annually from each of the 16 6.1 x 30.5 m (20 x 100 ft) from 1986 to 1991. At this time samples were collected from the 0 to 15 cm depth. The soil samples were collected and mixed in plastic buckets in the field, air-dried, passed through a 2-mm sieve, and stored at room temperature until analyzed. Unless otherwise noted, soil analyses were performed on air-dried, 2-mm size samples.

Laboratory Analyses

Cadmium, Cr, Cu, Ni, Pb, and Zn were determined in the plant material using a dry ashing method. These trace elements were sequentially extracted using a technique modified from Miller et al. (1986) and outlined in Chapter One. Other laboratory techniques for soil and plant analyses are outlined below. Unless otherwise noted, elements were determined in plant and soil solutions using a direct current plasma-atomic emission spectrometer (DCP-AES). Statistical analyses were performed using procedures supplied by SAS Institute, Inc. (SAS, 1985).

Soil pH (Eckert, 1988)

To 10.0 g of soil, 10-mL distilled water were added. The mixture was stirred and allowed to stand 15 min. The pH reading was taken immediately after stirring again, using an electrode that had been standardized at pH 4.0 and 7.0.

Extractable P (Knudsen and Beegle, 1988)

Two g of soil and 20 mL of 0.03*M* NH_4F - 0.025*M* HCl at pH 2.6 were shaken in a 50 mL Erlenmeyer flask for 5 min at 180 oscillations per min (opm) and filtered through Whatman #1 filter paper. Forty mL of a 2 L solution containing 125.0 g ammonium molybdate $[(NH_4)_6Mo_7O_{24} \cdot 4H_2O]$, 2.9 g potassium antimony tartrate $[K(SbO)C_4H_4O_6 \cdot \frac{1}{2}H_2O]$, and 1500 mL concentrated H_2SO_4 were mixed to a final volume of 2 L with 20 mL of a 1 L solution containing 105.6 g ℓ-ascorbic acid ($C_6H_8O_6$). Eighteen mL of this solution were mixed with 2 mL of the filtrate. Color development was measured after a minimum of 5 min at 660 nm using a Brinkmann PC800 Fiberoptic Probe Colorimeter.

Extractable Ca, K, Mg (Brown and Warncke, 1988)

To a 50 mL Erlenmeyer flask containing 2.5 g of soil were added 20 mL of 1*M* (ammonium acetate) NH_4OAc at pH 7. The mixture was shaken for 5 min at 180 opm and then filtered through Whatman #1 filter paper. Calcium and K were measured in the filtrate photometrically and Mg was measured colormetrically using an Auto-analyzer.

Cation Exchange Capacity (Rhoades, 1982)

A 2.0-g soil sample (corrected to oven-dry moisture content as determined using a separate subsample) plus 20 mL of 0.1*M* $BaCl_2$ (24.426-g $BaCl_2 \cdot 2H_2O\ L^{-1}$) saturating solution were added to a preweighed centrifuge tube, which was then stoppered and shaken for 2 h. The tube was centrifuged at 10,000 rpm for 15 min and solution decanted. The soil was equilibrated with three successive 20-mL increments of 0.002*M* $BaCl_2$ (at pH 7.0 using $Ba(OH)_2$ or HCl) each time by "sonifying" the solution for 10 to 30 s to disperse sediment, shaking for 1 h between centrifugations, and then discarding the supernatant after centrifugation. The centrifuge tube plus soil and entrained 0.002*M* $BaCl_2$ solution was weighed following the last decantation of supernatant. Ten-mL 0.005*M* $MgSO_4$ reactant solution was added and the solution shaken gently for 1 h. Electroconductivity (EC) of the reactant suspension was adjusted to that of the 0.0015*M* $MgSO_4$ ionic strength reference solution at the ambient laboratory conditions by the addition of 0.005*M* $MgSO_4$ reactant solution or distilled water. The reactant suspension conductivity, if necessary, was readjusted after allowing to gently shake overnight. Centrifuge tubes plus contents were weighed to determine the volume of $MgSO_4$ or water added. If both 0.005*M* $MgSO_4$ and water were added to the sample, the quantity of $MgSO_4$ solution was recorded. The volume of water added was calculated by difference. The supernatant was centrifuged, decanted, and analyzed for pH and Mg concentration.

If only distilled water was added:

$$CEC\text{-}cmol_c kg^{-1} = \frac{5-10(C_1 V_2)}{ovendry\ weight\ soil\ sample\text{-}g}$$

If more than 10 mL of the $MgSO_4$ reactant solution were added:

$$CEC\text{-}cmol_c kg^{-1} = \frac{0.5V_1 - 10(C_1V_2)}{ovendry\ weight\ soil\ sample\text{-}g}$$

where V_1 and V_2 are volumes (mL) of added $MgSO_4$ reactant solution and final supernatant solution, respectively, and C_1 is the concentration of Mg^{2+} in the supernatant in $cmol_c\ mL^{-1}$.

Particle Size Analysis (Day, 1965)

Fifty g of oven-dry soil (100 g for coarse textured soils) was weighed into a 600-mL or larger beaker. The organic matter was removed by adding 200-mL water to the soil followed by 20 mL of 30% hydrogen peroxide (H_2O_2) added slowly in 5-mL increments. The suspension was slowly heated until about 100 mL of water remained, taking care that the foam produced from the oxidizing organic matter did not overflow the beaker. This process should destroy most of the organic matter.

The soil suspension was quantitatively transferred to the dispersing cup, making sure not to overfill (one-third to one-half full). Fifty mL of dispersion solution (35.7 g sodium hexametaphosphate, $Na(PO_3)_6$ and 7.94 g sodium carbonate, $Na_2CO_3\ L^{-1}$) were added. The dispersing cup was attached to a malt mixer and mixed for 5 min.

After mixing, the suspension was poured into the hydrometer cylinder (a 1000-mL cylinder) and filled to mark with distilled water.

If necessary, 10 drops of a non-ionic anti-foaming agent was added. The suspension was stirred with a special plunger for 60 s to ensure good turbulent mixing throughout the cylinder. The plunger was removed on the last up-stroke and timing started immediately. The hydrometer was slowly lowered into the slurry to minimize turbulence. At 40 s the hydrometer reading and suspension temperature were recorded. Another hydrometer reading and temperature measurement were taken 2 h after the first.

A correction for density was made (R_d) by preparing and treating a blank that was similar in all respects to a sample except that no soil was used. The 40-s and 2-h blank hydrometer readings were subtracted from the 40-s and 2-h sample hydrometer readings, respectively.

To correct for changes in liquid viscosity due to temperatures that vary from 20°C (R_z), the following equation was used:

$$R_z = (\text{Temperature, °C} - 20) * 0.36$$

The amount of dispersed material remaining in suspension at a given reading was calculated as follows:

$$P = \frac{R - R_d + R_z}{W} \times 100$$

where R was the original hydrometer reading and W was the weight in grams of sample. P should be rounded to a whole percent.

At 40 s the assumption was that all the sand has settled. Therefore, the calculations are as follows:

$$\text{Corrected 40-s reading } (P_{40s}) = \text{percent silt and clay}$$

$$\text{Percent sand} = 100 - P_{40s}$$

$$\text{Corrected 2-hr reading } (P_{2h}) = \text{percent clay}$$

$$\text{Percent silt} = P_{40s} - P_{2h}$$

<u>Organic Matter by Wet Digestion</u> (Schulte, 1988)

To 1 g of soil in a 50-mL Erlenmeyer flask, 10 mL of 0.5*M* $Na_2Cr_2O_7$ and 10 mL of concentrated sulfuric acid were added and allowed to react for 30 min. The solution was diluted with 15 mL of water, mixed, and allowed to stand overnight (minimum of least 3 h). Ten mL was transferred into a colorimetric tube, taking care not to disturb the sediment on the bottom of the flask. The blue color intensity of the supernatant was determined on a colorimeter at 645 nm, with the reagent blank set to give 100% transmittance. The instrument was calibrated to read percent organic matter from a standard curve prepared from soils of known organic matter content.

<u>Plant Tissue Dry Ashing</u>

One g of plant material dried at 104°C was weighed into a ceramic crucible and placed in a furnace for 6 h at 500°C. Five mL of 6*M* HNO_3 were added to the ash in the crucible and the mixture allowed to stand for at least 2 h. The mixture was then transferred to a 10-mL volumetric flask and brought to a final volume using a LiCl solution having 2000 mg Li^+ kg^{-1}.

Total Elemental Analysis (Shuman 1979)

One-half g of air-dry soil, finely ground to pass through a 35-mesh sieve, was weighed into a 50-mL Teflon beaker and 1 mL of aqua regia (1 part concentrated HNO_3 to three parts concentrated HCl) was added to wet the sample. Eight mL of concentrated HF were added and the sample was digested in a sand bath/hot plate for 3 h at 80°C. Then the temperature was raised to 120°C and the sample was evaporated to dryness. Five mL of concentrated HNO_3 were added, the sample left overnight at room temperature, and then heated to dryness at 100°C. Five mL of concentrated HCl were added, the sample left overnight at room temperature, and heated again to dryness at 100°C. Residual salts were dissolved by warming in 1*M* HNO_3 (64.94-mL 69% HNO_3 L^{-1}), transferred with rinsing into a 25-mL volumetric flask, and taken to volume in 1*M* HNO_3.

0.1*M* HCl Soil Extraction (Whitney, 1988)

Five grams of soil plus 20 mL of 0.1*M* HCl were shaken for 30 min on a reciprocating shaker at 180 rpm. The solution was filtered through Whatman #5 paper.

AB-DTPA Soil Extraction (Soltanpour et al., 1982)

Ten g of air dried soil were weighed into an Erlenmeyer flask and 20.0 mL 1.0*M* NH_4HCO_3 (ammonium bicarbonate-AB) + 0.005*M* DTPA (diethylenetriaminepentaacetic acid) at pH 7.6 added. The mixture was shaken 15 min at 180 rpm on a reciprocating shaker and filtered through Whatman #42 filter paper. One mL of concentrated HNO_3 was added to 8.0 mL of the filtrate and shaken for about 10 min to remove any carbonate-bicarbonate remaining.

DTPA-TEA Soil Extraction (Lindsay and Norvell, 1978)

Twenty g of air-dry soil were weighed into an Erlenmeyer flask and 40 mL of 0.005*M* DTPA + 0.1M TEA (triethanolamine) + 0.01*M* $CaCl_2$ added. The mixture was shaken for 2 h, filtered through Whatman #42 filter paper, and then refrigerated until analyzed.

EDTA-$Ca(NO_3)_2$ Soil Extraction (Fujii and Corey, 1986)

Ten g of air-dry soil were weighed into an Erlenmeyer flask and 25 mL of 0.005*M* EDTA (ethylenediaminetetraacetic acid) + 0.01*M* $Ca(NO_3)_2$ added, the mixture shaken for 16 h, and then filtered through a 0.45-μm filter. The filtrate was mixed with an equal volume of 0.2*M* HNO_3 before analysis.

TCLP (Federal Register, 1990b)

The type of extraction fluid to use was determined by weighing a 5.0 $\pm$ 0.1 g subsample of soil with a particle-size of 1-cm or less in diameter into a 500-mL beaker or Erlenmeyer flask. Ninety-five mL of reagent water (ASTM Type II) were added, the solution covered with a watchglass, and stirred vigorously for 5 min using a magnetic stirrer. The pH was then measured. For mixtures with pH <5.0, extraction fluid #1 should be used. For pH >5.0, 3.5-mL 1.0*M* HCl was added and the slurry was mixed briefly, covered with a watchglass, and heated to 50°C for 10 min. The solution was cooled to room temperature and the pH determined. If the pH was <5.0 extraction fluid #1 was used. If the pH was >5.0, extraction fluid #2 was used.

Extraction fluid #1: Made by adding 5.7-mL glacial HOAc to 500-mL reagent water, adding 64.3-mL 1.0*N* NaOH, and diluting to 1 L. The pH of this fluid is 4.93±0.05 when correctly prepared.

Extraction fluid #2: Made by diluting 5.7-mL glacial HOAc with reagent water to a volume of 1 L. When correctly prepared, the pH of this fluid is 2.88±0.05.

A 100-g (minimum) representative sample was weighed into a suitable extractor vessel made out of plastic and 2000 g of extraction fluid was added. The vessel was closed tightly using Teflon tape to ensure a tight seal. Once secured in a rotary agitation device, the vessel was rotated at 30±2 rpm for 18±2 h at 23±2°C. To relieve excess pressure that may build up, the extractor bottle may be periodically opened and vented into a hood. The material in the extractor vessel was vacuumed filtered through a new, acid-washed glass fiber filter (Whatman filter model GFF, nominal pore size 0.7, Whatman Laboratory Products, Inc.). Filters were acid washed with 1*M* HNO_3 followed by three consecutive rinses with deionized distilled water using 1 L per rinse. The filtered liquid material obtained is defined as the TCLP extract. The pH of this solution was measured and the solution preserved until analyzed by acidifying an aliquot with nitric acid to pH <2.

RESULTS AND DISCUSSION

Sequential Extraction of Surface Soils

Chemical and physical measurements performed on the surface soils from Control plots and Treatments 1, 2, and 3 are listed in Table 14. The pH values of soils from Treatment 3 were significantly higher than those of Treatments 1 and 2 and the Control. Extractable Mg values were higher in soils from the Control and Treatment 1 compared to Treatments 2 and 3. No treatment differences occurred for exchangeable K and cation exchange capacity (CEC). Extractable P and Ca and percent organic matter were higher in soils from sludge treated areas compared to controls as a result of sludge applications. These differences were because of sludge application since lime and fertilizers (except N on corn) were not applied during the course of the study. Differences between treatments for soil pH, extractable nutrients, and organic matter may have been large enough to have had an effect on crop growth and yields. Extractable nutrients, except for K, were at levels at which no additional lime or fertilizer applications were recommended.

The results of the sequential extraction performed on these samples, which were collected in the spring of 1990, are tabulated in Table 15 and shown graphically in Figures 14 to 19. Total values listed in this table were determined using a wet digestion technique (Shuman, 1979) and are not a summation of the eight other fractions. For this reason, values for total analysis did not equal the sum of the individual fractions.

Table 14. Average soil characterization values for surface samples†.

Soil Parameter‡	Control	Treatment 1	Treatment 2	Treatment 3
pH	7.0±0.4b§	6.9±0.2b	6.9±0.1b	7.5±0.1a
Extractable Nutrients (mg kg^{-1})				
P	170±10c	327±40b	640±30a	360±20b
Ca	840±190c	1070±200b	1230±110b	1710±50a
Mg	130±10a	130±30a	100±10b	90±10b
K	70±10a	80±20a	100±10a	90±10a
CEC	5.8±1.1c	6.9±1.1b	7.0±0.6b	8.6±0.5a
Texture	Sandy Loam	Sandy Loam	Sandy Loam	Sandy Loam
Sand (%)	69±4	70±3	68±3	66±2
Silt (%)	21±3	20±1	21±2	23±1
Clay (%)	11±2	11±2	11±1	11±0
% O.M.	2.1±0.2c	3.64±0.3b	5.0±0.5a	5.1±0.3a

†Values for pH, extractable P, Ca, Mg, and K, and Organic Matter (%) from samples collected fall 1991. Values for CEC and texture from samples collected spring 1990.
‡CEC is Cation Exchange Capacity in $cmol_c kg^{-1}$. % O.M. is percent Organic Matter.
§Values in rows followed by the same letter were not significantly different at $p = 0.05$.

Cadmium

Concentrations of Cd in the chemical fractions of the four control plots were below analytical detection limits (2 mg Cd kg^{-1}). The Cd measured resided primarily in the acid-soluble fraction of treated soils (Table 15 and Figure 14). The soil of Treatment 1, which had the greatest amount of total Cd in these surface soils, also had measurable

Table 15. Sequential fractionation of surface soil samples collected spring 1990.

Fraction†	Control	Treatment 1	Treatment 2	Treatment 3
	-------------	mg kg^{-1}	-------------	
		Cadmium		
Soluble	nd‡	nd	nd	nd
Exchange	nd	2.5±0.5	nd	nd
Acid Sol.	nd	4.8±0.8a§	3.0±0.3b	3.1±0.2b
MnO	nd	nd	nd	nd
Organic	nd	nd	nd	nd
Am.FeO	nd	nd	nd	nd
Cr.FeO	nd	nd	nd	nd
Residual	nd	nd	nd	nd
Total	nd	7.7±0.9a	4.4±0.1b	4.8±0.6b
		Chromium		
Soluble	nd	nd	nd	nd
Exchange	nd	nd	nd	nd
Acid Sol	8± 1b	39± 3a	38± 4a	38± 3a
MnO	nd	11± 1b	11± 1b	13± 1a
Organic	25± 7c	288±44a	202±20b	243± 13b
Am.FeO	15± 5d	56±12c	114±21b	286± 40a
Cr.FeO	29± 8b	74±16b	208±40a	224± 29a
Residual	29± 5c	35± 3bc	48±11ab	52± 15a
Total	110±12c	517±68b	565±28a	822±118a
		Copper		
Soluble	0.8±0.1d	1.3±0.3c	2.4±0.3b	4.0±0.3a
Exchange	nd	1.0±0.4c	1.5±0.4b	2.5±0.4a
Acid Sol	22±5d	57±12 c	142±14b	261±15a
MnO	3±1c	9± 2 c	32± 4b	48± 5a
Organic	7±2c	17± 4 c	56± 8b	86± 9a
Am.FeO	7±3c	14± 3 c	39± 6b	71±10a
Cr.FeO	6±1c	9± 2 c	25± 2b	35± 2a
Residual	5±1b	6± 1 b	11± 3a	12± 4a
Total	50±8c	110±21 c	287±13b	513±64a

Table 15 (cont'd)

Fraction	Control	Treatment 1	Treatment 2	Treatment 3
	mg kg^{-1}			
		Nickel		
Soluble	nd	1.1±0.4c	4±0.4a	2± 1b
Exchange	5±2d	16± 5c	58± 6a	28± 6b
Acid Sol	17±4c	29± 7c	173±25a	126± 3b
MnO	2±1c	5± 2c	33± 6a	23± 2b
Organic	2±1c	6± 1c	31± 6b	39± 4a
Am.FeO	9±2c	20± 5c	91±17b	169±20a
Cr.FeO	10±2b	17± 5c	56± 6a	61± 4a
Residual	14±3c	20± 9bc	24± 3ab	28± 5a
Total	52±7b	94±17b	416±49a	432±49a
		Lead		
Soluble	nd	nd	nd	nd
Exchange	nd	nd	nd	nd
Acid Sol	10± 1b	8± 1b	19± 1a	18± 3a
MnO	6± 3b	5± 3b	19± 2a	17± 3a
Organic	nd	13± 3b	35± 4a	30± 4a
Am.FeO	nd	nd	nd	4± 2
Cr.FeO	nd	7± 4b	9± 2ab	12± 2a
Residual	64±19a	105±79a	99±25a	110±10a
Total	59± 7b	80±10b	166±12a	176±20a
		Zinc		
Soluble	nd	3± 5b	10± 4a	nd
Exchange	31±18c	71±21b	511± 39a	32± 13c
Acid Sol	107±35d	309±71c	1940±120a	559± 53b
MnO	12± 9c	17±10c	80± 15a	33± 8b
Organic	14± 3c	22± 6c	89± 7a	48± 11b
Am.FeO	12± 3c	22± 5c	119± 5a	75± 10b
Cr.FeO	44± 6c	71±11c	172± 8a	127± 9b
Residual	41±12a	35±17a	56± 10a	73± 46a
Total	217±27c	461±88c	2450±240a	801±137b

†Fractions: Water Soluble; Exchangeable; Acid Soluble; Mn oxides; Organic; Amorphous Fe oxides; Crystalline Fe oxides; Residual; and Total concentration found by analysis, respectively.
‡nd = not detectable. Concentrations of Cd, Cr, Cu, Ni, Pb, and Zn less than 2.5, 0.8, 0.8, 0.8, 4, and 2 mg kg^{-1}, respectively, were below analytical detection limits.
§Values in rows followed by the same letter were not significantly different at p = 0.05.

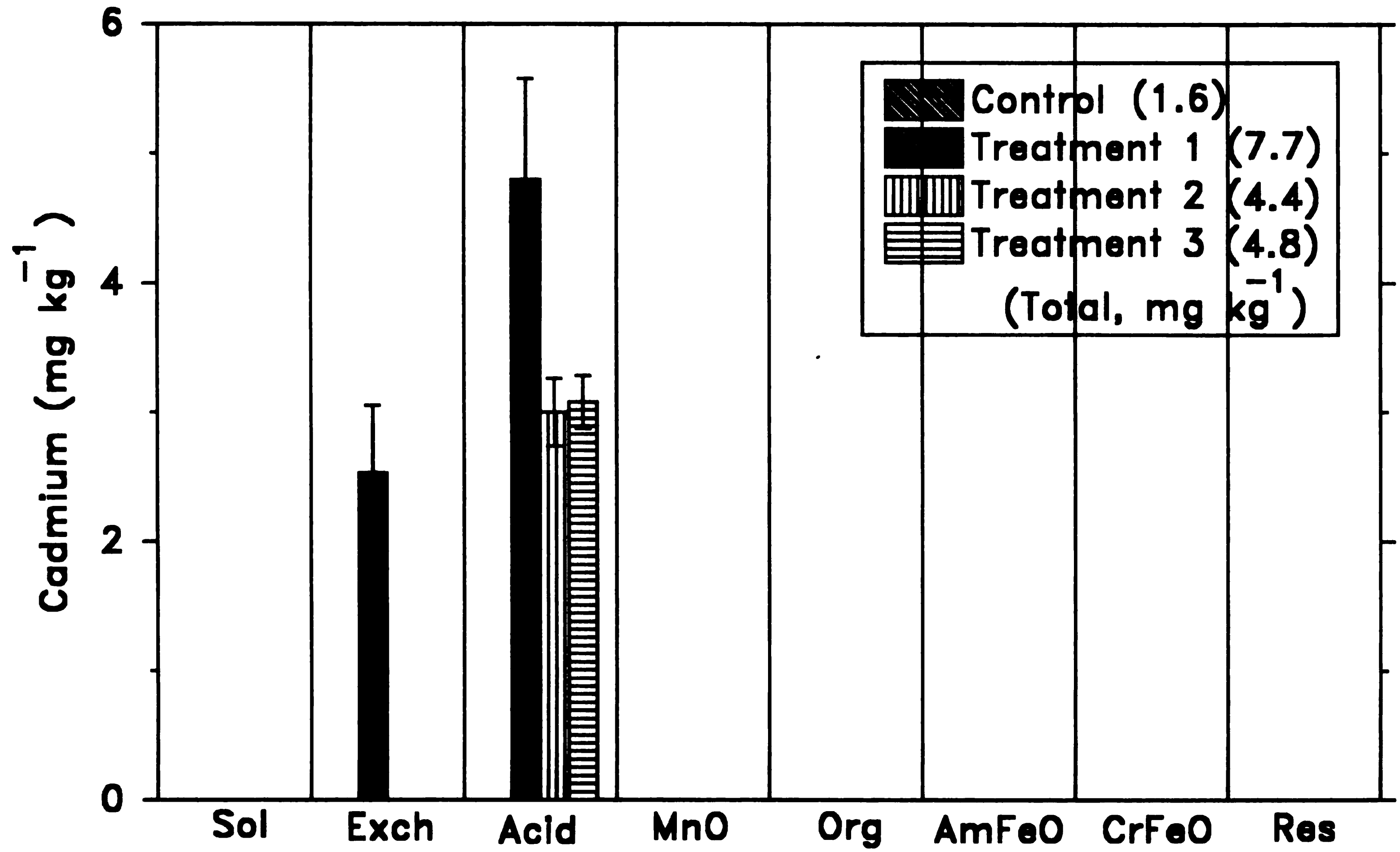

Figure 14. Chemical fractionation of Cd in surface soils to which wastewater sludges were applied.

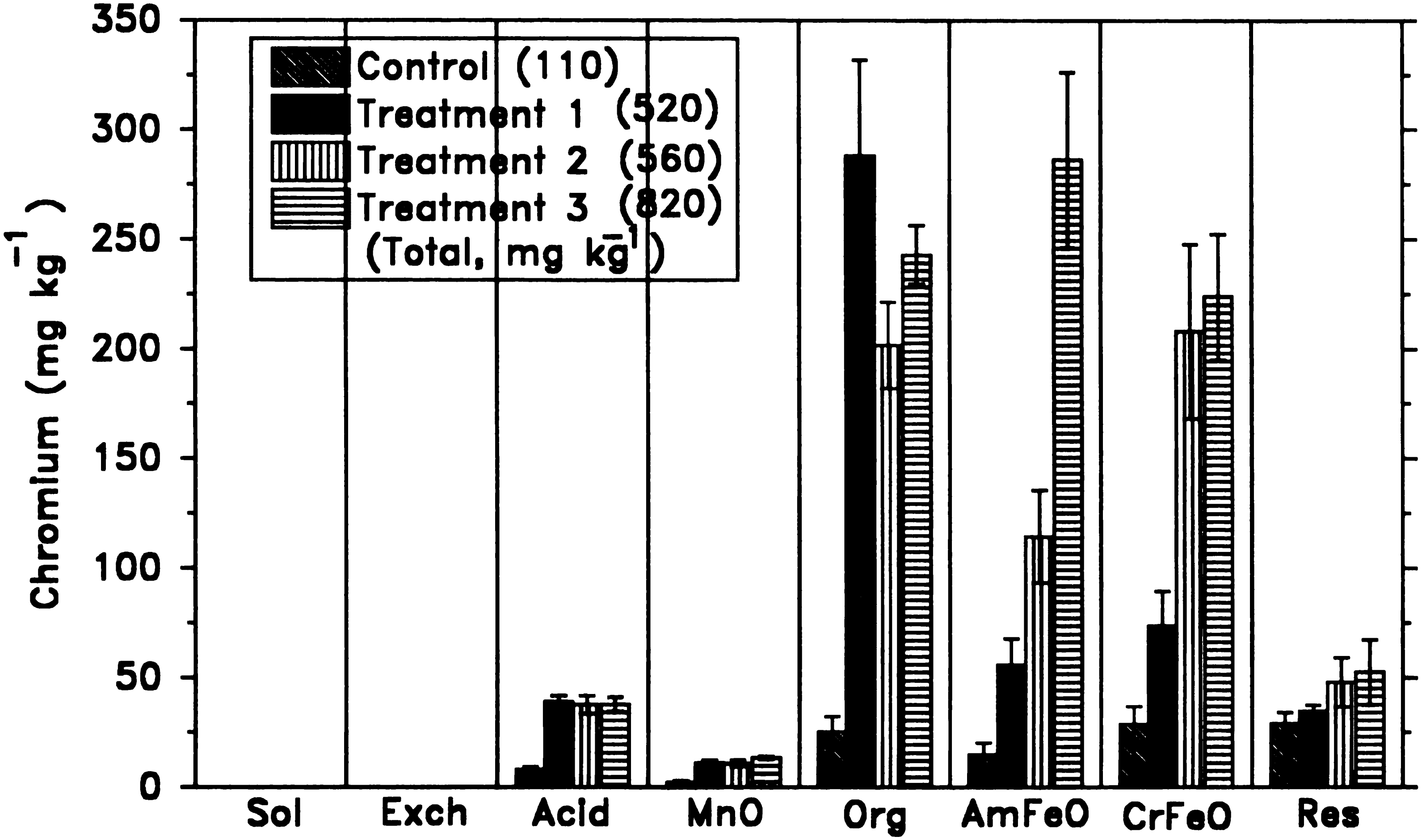

Figure 15. Chemical fractionation of Cr in surface soils to which wastewater sludges were applied.

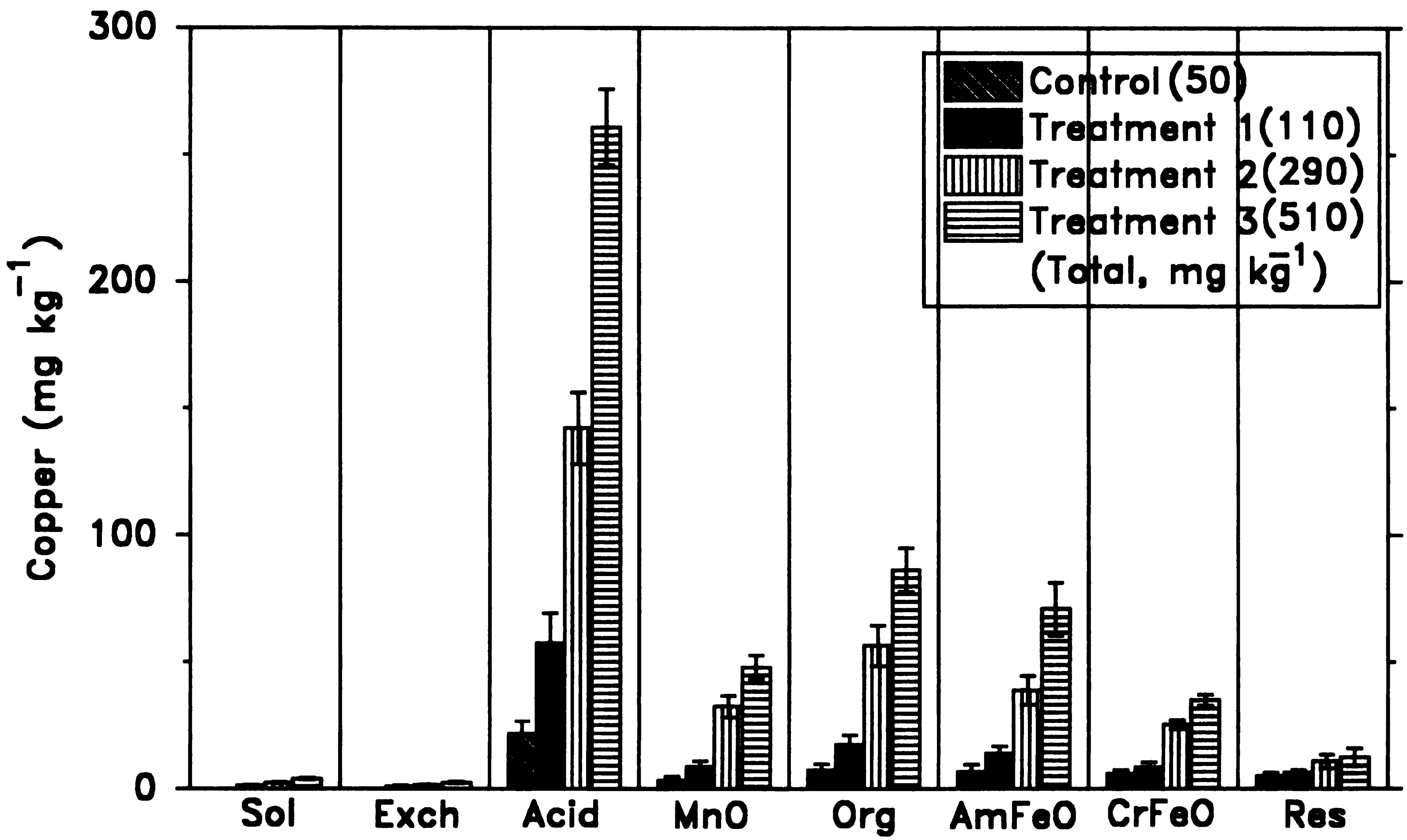

Figure 16. Chemical fractionation of Cu in surface soils to which waterwater sludges were applied.

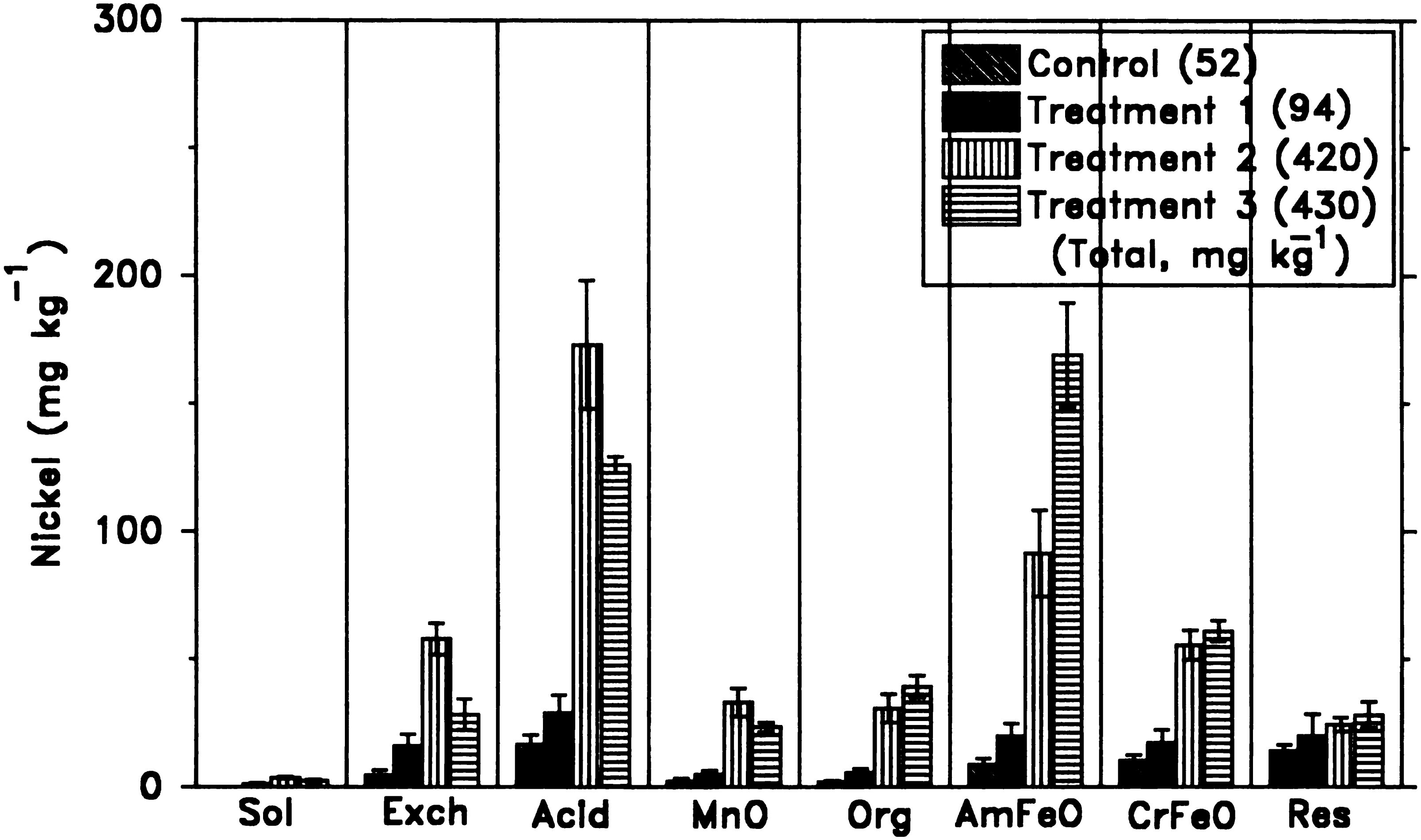

Figure 17. Chemical fractionation of Ni in surface soils to which wastewater sludges were applied.

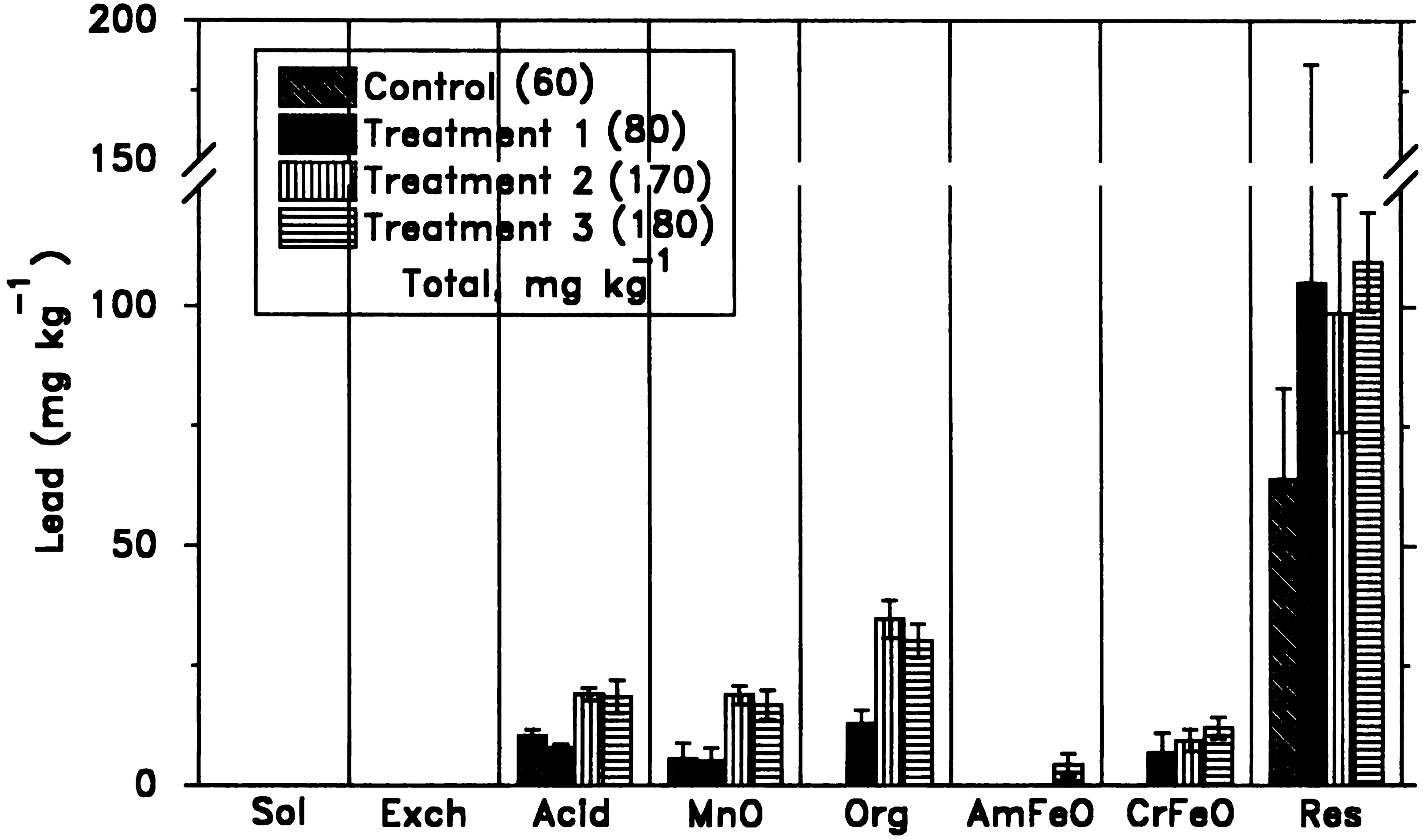

Figure 18. Chemical fractionation of Pb in surface soils to which wastewater sludges were applied.

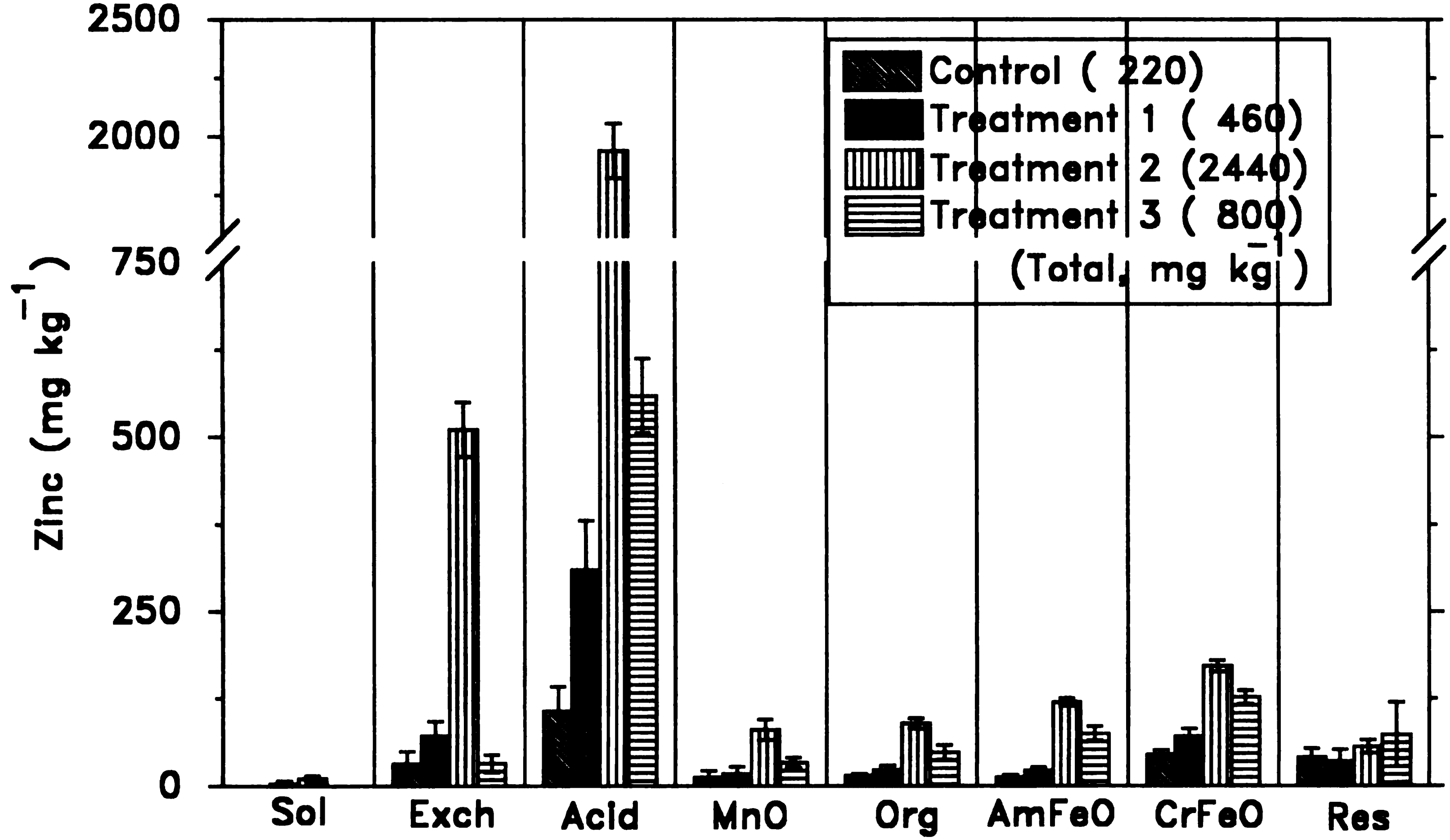

Figure 19. Chemical fractionation of Zn in surface soils to which wastewater sludges were applied.

quantities of Cd in the exchangeable fraction. Although sludge containing Cd was last applied in 1986 on Treatment 1 and in 1984 on Treatments 2 and 3, 65% or more of the total sludge-applied Cd occupied the exchangeable and acid-soluble fractions, indicating its potential environmental availability. Cadmium did not appear to be transformed into less available soil fractions even with a soil pH of about neutral (Table 14). Rather, at highest Cd loading rates (Treatment 1), measurable amounts of Cd begin to reside in the more environmentally available exchangeable fraction rather than other fractions.

Cadmium fractionation data of this study contrasted with the observations of other researchers, which indicated greatest Cd concentrations in the exchangeable (Hickey and Kittrick, 1984) and free ionic form (Emmerich et al., 1982b), forms of Cd considered to be more environmentally available than the acid-soluble fraction. A study by Elliott et al. (1990) examining the composition and distribution of Cd in water treatment sludges found that less than 6% of the total Cd was in an exchangeable form (1*M* $MgCl_2$ @ pH 7), 19% in an acid-soluble form, 38% organically bound, and 38% in the residual fraction. Although their sludges had lower total concentrations of Cd than did the treated soils of this study, comparison seemed to indicate that Cd in our sludge-treated soils were in more readily available fractions compared to the sludges, indicating that application of Cd in sludge to soils may become more bioavailable because of the differences in chemistry between sludge and soil Cd.

Chromium

The average concentrations of Cr in water-soluble and exchangeable fractions were below analytical detection limits (0.8 mg kg^{-1}) for soils of all treatments and the control (Table 15 and Figure 15). In contrast to Cd, soils with greater concentrations of total Cr had increasing amounts of the added Cr in the less available fractions, including the organic, the two Fe oxide fractions, and residual forms rather than water-soluble, exchangeable, and acid-soluble. Fractionation of wastewater sludges indicated that much of the Cr already resided in the oxide component (Elliott et al., 1990). Rather than changing from more available to less available forms when the sludge was applied to soils, the Cr in the sludge likely remained in oxide and residual components. As more Cr was applied to the soil, a smaller percentage of the total resided in the acid-soluble and Mn oxide fractions.

Copper

Copper resided primarily in the acid-soluble fraction (Table 15 and Figure 16). Greater concentrations of total Cu in this soil resulted in greater concentrations in all fractions. The percentage residing in the first three fractions, however, did not significantly change as total soil Cu content increased from 110 to 510 mg kg^{-1}. The percent of the total Cu found in the residual fraction decreased from soils with low total Cu (Control; 10% of total) to those with high total Cu (Treatment 3: 2% of total). Less than 20% of the total Cu resided in the organic component, and the percent of total Cu in this fraction did not change as total soil Cu increased. This differed from the results reported by others (Emmerich et al., 1982a; Sposito et al., 1982) in which Cu was primarily in the organic fraction.

<u>Nickel</u>

Fractionation data for Ni (Table 15 and Figure 17) indicated that as total soil Ni increased in the surface soils of the treated plots, the concentrations of Ni in the individual fractions also increased. This increase was significant in the water-soluble and exchangeable fractions, indicating that additions of Ni to soil become increasingly environmentally available. Nickel, however, predominated in the acid-soluble and Fe oxide fractions, regardless of the total concentration in the soil.

The sequential extraction of Ni from soils of Treatments 2 and 3 provided an interesting comparison. Although the total concentration of Ni measured in soils from both treatments was about equal, greater concentrations of Ni resided in the water-soluble, exchangeable, acid-soluble, and Mn oxide components of Treatment 2 soils, whereas greater concentrations of Ni in Treatment 3 soils resided in the organic and amorphous Fe oxide fractions. Thus, Ni in Treatment 2 soils was present in chemical components considered more environmentally available compared to those of Treatment 3.

An examination of the sludge application records for these two treatments (Table 11) indicated that greater amounts of Ni were applied to Treatment 2 soils (2100 and 1730 kg Ni kg^{-1} for Treatments 2 and 3, respectively). Also, the sludges came from different municipalities. Furthermore, Ni was applied to the Treatment 3 plots more recently than it was to Treatment 2 plots (Table 11). More than 65% of the total Ni was applied to Treatment 3 since 1982, and the most recent application was in 1986. Treatment 2 plots, however, received only about 12% of their total Ni since 1982 and the last application was in 1984.

This may indicate that Ni became more available with time in the soil or that there were differences in Ni in the applied sludges that are reflected in the fractionation of soils in the two treatments. Fractionation of wastewater treatment sludges indicated that Ni was primarily (>80%) in oxide and residual forms, which are considered unavailable (Elliott et al., 1990).

Lead

As the total Pb concentration increased from 60 mg kg^{-1} in control plots to about 170 mg kg^{-1} in the plots of Treatments 2 and 3, Pb in the acid-soluble, Mn oxide, organic, and Fe oxide fractions increased (Table 15 and Figure 18). Concentrations of Pb in water-soluble and exchangeable fractions, however, remained below the levels of analytical detection (4 mg kg^{-1}), even though total Pb increased about three-fold. Most of the Pb was in the residual component. Results indicated that Pb continued to reside in soil chemical fractions that were not environmentally available.

Zinc

Additional loadings of Zn applied to the treated plots increased the quantities of this element in all but the residual fraction, again indicating increased plant availability with increasing total Zn concentrations in the soil (Table 15 and Figure 19). More than 70% of the total Zn resided in the exchangeable and acid-soluble fractions of sludge-treated soils.

Complete yield loss of the legume crops grown on plots of Treatment 2, i.e., soybean (*Glycine max* L.) and alfalfa (*Medicago sativa* L.) was due to the toxic effect of Zn on these legumes. The plants did not grow much after emergence from the soil. The few plants that emerged and grew were stunted and chlorotic, symptoms indicative of general Zn toxicity (Jones, 1991).

Crop Yields and Uptake of Elements

Corn

Corn grain harvest data for 1985 to 1988 and 1990 are listed in Table 16. Corn grain yields in control plots were equal to or less than yields of the sludge treated plots. The average yield (corrected to 15.5% moisture) in control plots and Treatment 2 for these years was about 6.1 Mg ha^{-1}. Treatments 1 and 3 had average grain yields of 7.6 and 7.7 Mg ha^{-1}, respectively, which were significantly higher ($p = 0.05$) compared to the Control and Treatment 2.

Elemental concentrations in corn diagnostic tissue samples were used as guidelines to assess corn nutrition. Concentrations of the macronutrients, i.e., Ca, K, Mg, and P (Table 17), and the micronutrients, i.e., B, Fe, Mn, Mo, Cu, and Zn (Tables 18 and 19), were greater or the same in the diagnostic tissue of corn plants from Treatments 1, 2, and 3 compared to the Control. Sludge-treated soils had greater amounts of organic matter than the control soils (Table 14). The yield increase of treated plots may be due to the residual N and organic matter from sludge application (Chang et al., 1982).

Table 16. Corn grain harvest data for 1985 to 1988 and 1990.

Year	Control	Treatment 1	Treatment 2	Treatment 3
	- - - - - - - - - - Mg ha^{-1} @ 15.5% Moisture - - - - - - - - -			
1985	7.2±1.0a†	8.1±0.7a	6.7±0.5a	7.9±0.9a
1986	10.0±0.7a	11.3±1.0a	6.8±0.7b	11.1±1.8a
1987	5.7±1.5b	8.5±1.7a	6.4±1.4b	8.0±2.0a
1988	2.1±0.2a	2.5±0.3a	2.4±0.3a	2.7±0.5a
1990	5.6±0.9b	7.6±1.2a	8.3±0.8a	8.6±1.5a
Average	6.1±2.8b	7.6±3.1a	6.1±2.2b	7.7±3.1a

lsd‡ (Year*Treatment) = 1.1

†Values in a row or column followed by the same letter are not significantly different at the p = 0.05 level.
‡Fisher's (unprotected) least significant difference at p = 0.05.

Corn grain yield in Treatment 2 plots likely was reduced compared to the other two sludge treatments because of the high levels of Ni and Zn in the soil that is reflected in diagnostic tissue concentrations (Table 19). Plants in Treatment 2 plots also had equal or lower uptake of B, Fe, Mn, K, Mg, and P compared to plants from control plots. Nutrient sufficiency ranges listed by Jones et al. (1990) for corn diagnostic tissue (corn ear leaf) taken at silking were similar to those found in our study, with the exception of Zn in Treatment 2. Excessive amounts of available Zn have been shown to effect the uptake and metabolism of other elements, including P and Fe, and Mn (Murphy and Walsh, 1972; Jones, 1991).

Trace elements in corn grain and in the whole plant at the end of the growing season probably have the greatest chance of entering the human food chain because of their use as animal feeds and in human food products. The data for the trace elemental content of corn grain

Table 17. Concentrations of Ca, Mg, K, and P in corn diagnostic tissue samples taken between 1985 and 1990.

Year	Control	Treatment 1	Treatment 2	Treatment 3
	- - - - - - - -	- - - - - - - - g kg^{-1}	- - - - - - - -	- - - - - - - -
		Calcium		
1985	0.63±0.09c†	0.91±0.08b	1.01±0.04a	1.02±0.03a
1987	0.84±0.09c	1.05±0.05bc	1.32±0.18a	1.11±0.11ab
1988	0.81±0.05b	0.94±0.07a	0.99±0.04a	1.00±0.07a
1990	0.73±0.06c	0.87±0.08b	1.06±0.06a	0.94±0.02b
Average	0.75±0.11d	0.94±0.09c	1.09±0.16a	1.02±0.09b
lsd‡ (Year*Treatment) = 0.11				
		Potassium		
1985	1.29±0.18ab	1.40±0.16a	1.26±0.14ab	1.18±0.13b
1987	1.90±0.31a	1.64±0.35ab	1.55±0.28b	1.50±0.16b
1988	1.11±0.09a	1.08±0.14a	1.14±0.20a	0.98±0.15a
1990	1.63±0.15a	1.52±0.11a	1.68±0.10a	1.58±0.06a
Average	1.48±0.36a	1.41±0.29a	1.41±0.28a	1.31±0.28b
lsd (Year*Treatment) = 0.17				
		Magnesium		
1985	0.30±0.09b	0.42±0.08b	0.37±0.09ab	0.42±0.06b
1987	0.41±0.10a	0.48±0.03a	0.46±0.05a	0.50±0.10a
1988	0.42±0.06bc	0.55±0.11a	0.35±0.11c	0.47±0.04ab
1990	0.30±0.06b	0.37±0.04a	0.22±0.05c	0.27±0.01b
Average	0.36±0.09c	0.45±0.09a	0.35±0.11c	0.42±0.11b
lsd (Year*Treatment) = 0.07				
		Phosphorus		
1985	0.23±0.02a	0.25±0.01a	0.22±0.01a	0.23±0.02a
1987	0.29±0.02a	0.28±0.05a	0.26±0.03a	0.28±0.02a
1988	0.24±0.01a	0.24±0.01a	0.21±0.01b	0.24±0.02a
1990	0.23±0.01a	0.24±0.01a	0.22±0.02a	0.24±0.01a
Average	0.24±0.03a	0.25±0.03a	0.23±0.03b	0.25±0.03a
lsd (Year*Treatment)was not significant at p = 0.05.				

†Values in rows followed by the same letter were not significantly different at p = 0.05.
‡Fisher's (protected) least significant difference at p = 0.05.

Table 18. Concentrations of B, Fe, Mn, and Mo in corn diagnostic tissue collected in 1985 to 1988 and 1990.

Year	Control	Treatment 1	Treatment 2	Treatment 3
	mg kg^{-1}			
		Boron		
1985	10.7±1.2c†	15.6±2.8b	11.1±1.8c	19.4±1.5a
1986	5.5±0.7b	6.0±0.5b	6.3±0.5b	8.1±1.5a
1987	11.7±2.1a	10.0±2.3a	11.1±2.2a	15.3±5.5a
1988	9.0±1.4b	8.8±0.4b	10.0±2.1ab	12.1±1.7a
1990	6.1±0.5a	7.2±0.7a	7.2±0.8a	7.1±0.3a
Average	8.6±2.8b	9.5±3.7b	9.1±2.5b	12.4±5.3a
lsd‡ (Year*Treatment) = 2.3				
		Iron		
1985	87±12a	93± 7a	83± 5a	84±10a
1986	89± 4b	97± 4a	81± 4c	89± 6b
1987	102±13a	113±10a	108± 4a	106± 9a
1988	96± 3a	97± 2a	97± 5a	100±10a
1990	109± 8b	120±10ab	129±14a	115± 3b
Average	97±12b	104±13a	100±19ab	99±13b
lsd (Year*Treatment) = 10				
		Manganese		
1985	52±16a	56± 5a	35±4b	52±10a
1986	68±21a	54± 9a	34±6b	51± 4ab
1987	46±12a	42± 6a	33±4a	47± 4a
1988	44± 7a	37± 4bc	35±3c	41± 4ab
1990	28± 2c	32± 2bc	37±4ab	38± 4a
Average	48±18a	44±11a	35±4b	46± 7a
lsd (Year*Treatment) = 10				
		Molybdenum		
1985	2.2±0.7d	3.1±0.2c	4.0±0.4b	5.1±0.4a
1986	0.9±0.5c	1.9±0.2b	2.0±0.6b	2.8±0.4a
1987	0.4±0.7c	2.3±0.3b	3.1±0.6a	3.0±0.9a
1988	1.5±0.6b	3.1±1.1a	4.0±0.9a	4.3±0.3a
1990	3.1±1.1c	4.9±1.3bc	8.3±2.0a	6.7±0.5ab
Average	1.6±1.2c	3.1±1.3b	4.3±2.4a	4.4±1.6a
lsd (Year*Treatment) = 1.2				

†Values in rows followed by the same letter were not significantly different at p = 0.05.
‡Fisher's (protected) least significant difference at p = 0.05.

Table 19. Concentrations of Cd, Cr, Cu, Ni, Pb, and Zn in corn diagnostic tissue for years 1985 to 1988 and 1990.

Year	Control	Treatment 1	Treatment 2	Treatment 3
	- - - - - - - - - - - - -	mg kg^{-1}	- - - - - - - - - - - - -	
		Cadmium		
1985	nd†	1.41±0.31	nd	nd
1986	nd	0.99±0.13	nd	nd
1987	nd	1.16±0.05	nd	nd
1988	nd	0.52±0.23	nd	nd
1990	nd	0.69±0.08	nd	nd
Average	nd	0.95±0.37	nd	nd
		Chromium		
1985	nd	0.20±0.03	0.31±0.09	0.30±0.04
1986	nd	nd	0.28±0.06	nd
1987	nd	nd	0.27±0.05	nd
1988	0.37±0.03a‡	0.45±0.05a	0.50±0.04a	0.94±0.86a
1990	nd	nd	nd	nd
Average	nd	nd	0.30±0.15	0.30±0.49
		Copper		
1985	9.6±0.7c	16.2±2.3ab	15.6±1.5b	18.7±1.5a
1986	13.5±0.6a	13.7±0.9a	10.8±1.5b	14.7±1.8a
1987	11.8±1.0c	16.6±0.6b	15.7±1.5b	21.4±1.8a
1988	13.9±2.4c	18.4±2.0b	17.8±0.5b	22.1±1.8a
1990	9.8±0.8c	12.8±1.7b	16.8±1.7a	15.3±1.4a
Average	11.7±2.2c	15.5±2.5b	15.4±2.8b	18.5±3.5a
lsd§ (Year*Treatment) = 2.1				
		Nickel		
1985	0.41±0.03b	1.37±0.48b	6.87±1.15a	0.90±0.11b
1986	0.40±0.28c	1.20±0.52bc	6.01±1.13a	1.45±0.22b
1987	0.24±0.12b	0.48±0.14b	3.57±1.18a	0.75±0.19b
1988	0.26±0.12b	0.51±0.21b	2.12±0.30a	0.43±0.07b
1990	nd	0.44±0.27b	4.53±0.43a	0.25±0.10b
Average	0.29±0.19c	0.80±0.52b	4.62±1.92a	0.76±0.45b
lsd (Year*Treatment) = 0.75				

Table 19 (cont'd)

Year	Control	Treatment 1	Treatment 2	Treatment 3
	- - - - - - - - - - - - mg kg^{-1} - - - - - - - - - - - -			
		Lead		
1985	nd	1.05±0.11	1.08±0.31	1.23±0.25
1986	nd	nd	nd	nd
1987	nd	nd	1.62±0.43	nd
1988	nd	nd	nd	nd
1990	nd	nd	1.17±0.50	nd
Average	nd	nd	nd	nd
		Zinc		
1985	45± 8c	174±51b	314± 54a	117±15b
1986	101±42c	167±31b	264± 55a	143±18bc
1987	80±28c	146± 7b	265± 51a	138±10b
1988	96±24c	174±33b	289± 28a	150±19b
1990	83±31b	172±74b	646±167a	144±21b
Average	81±33c	167±42b	356±169a	138±19b

lsd (Year*Treatment) = 74

†nd = not detectable. Concentrations of Cd, Cr, Ni, and Pb less than 0.5, 0.2, 0.2, and 1.0 mg kg^{-1}, respectively, were below analytical detection limits.
‡Values in rows followed by the same letter were not significantly different at p = 0.05.
§Fisher's (protected) least significant difference at p = 0.05.

harvested in 1985, 1987, 1988, and 1990 are listed in Table 20. Values for Cr and Pb were below the analytical detection capabilities of the DCP-AES for all samples. Values of Cd also were below detection limits for many of the samples so statistical analysis was not possible.

There were no treatment differences for Cu concentrations ($p = 0.05$). Corn grain concentrations of Ni and Zn, however, were significantly higher in plants grown on sludge-treated soils compared to the control. Corn grain from plants grown on Treatment 2 exhibited the greatest concentrations of Ni and Zn, corresponding to the highest levels in the soils (Table 15).

Concentrations of trace elements in whole corn plant tissue at the end of the 1990 growing season (corn stover) are listed in Table 21. This was the only year in which corn was sampled at this stage of growth. Average concentrations of Cd and Pb were at or below detection limits (0.5 mg Cd kg^{-1} and 1.0 mg Pb kg^{-1}) for all treatments even though soils of some treatments contained greater than twice the concentrations of total Cd and Pb compared to Control (Table 15). Tissue from plants grown on control plots were below detection (0.2 mg kg^{-1}) for the average value of Ni. There were no significant differences in Cr concentrations among treatments. Concentrations of Cu, Ni, and Zn were significantly higher in corn stover collected from one or more of the treated plots compared with the controls (Table 21). Except for Cr and Pb, significant differences existed among treatments between concentrations of trace elements in the corn stover and in surface soils of the same treatments (Table 15).

Chromium and Pb concentrations in corn diagnostic tissue, grain, and stover were at or below the analytical detection limits of DCP-AES.

Table 20. Concentrations of Cd, Cr, Cu, Ni, Pb, and Zn in corn grain harvested in 1985, 1987, 1988, and 1990.

Year	Control	Treatment 1	Treatment 2	Treatment 3
	- - - - - - - - - - - - - -	mg kg^{-1}	- - - - - - - - - - - - -	
		Cadmium		
1985	0.56±0.16	0.54±0.10	nd	0.58±0.08
1987	0.65±0.12a†	0.71±0.12a	0.68±0.05a	0.75±0.10a
1988	nd‡	nd	nd	nd
1990	nd	nd	nd	nd
Average	nd	0.50±0.18	nd	0.51±0.20
		Copper		
1985	1.64±0.10a	1.70±0.47a	1.85±0.20a	2.23±0.32a
1987	2.54±0.22b	2.37±0.18b	2.49±0.56b	4.94±2.20a
1988	1.82±1.61a	1.15±0.41a	2.69±2.07a	1.42±1.03a
1990	3.57±2.10a	2.54±0.25a	2.65±0.60a	2.74±1.00a
Average	2.39±1.42a	1.94±0.65a	2.42±1.06a	2.83±1.79a
lsd§ (Year*Treatment) was not significant at p = 0.05.				
		Nickel		
1985	0.35±0.09c	1.89±0.80b	5.74±0.65a	1.95±0.54b
1987	0.29±0.34d	1.14±0.32c	4.04±0.66a	2.18±0.55b
1988	0.75±0.40b	1.65±1.01ab	2.89±0.77a	1.77±0.61ab
1990	0.32±0.15b	0.70±0.50b	4.70±0.28a	0.74±0.20b
Average	0.42±0.31c	1.34±0.79b	4.34±1.20a	1.66±0.73b
lsd (Year*Treatment) = 0.74				
		Zinc		
1985	15.2±2.1c	22.1±2.1b	26.9±1.7a	19.7±3.0b
1987	29.3±4.7a	30.4±2.7a	35.9±2.9a	31.5±2.6a
1988	21.1±0.3c	25.3±3.7ab	28.9±2.5a	23.8±2.7bc
1990	23.2±1.8b	24.7±3.1b	35.8±2.5a	24.2±1.6b
Average	22.2±5.8c	25.6±4.1b	31.9±4.7a	24.8±5.0b
lsd (Year*Treatment) = 3.6				

†Values in columns followed by the same letter were not significantly different at p = 0.05.
‡nd = not detectable. Concentrations of Cd, Cr, and Pb below 0.5, 0.2, and 1.0 mg kg^{-1}, respectively, were below analytical detection limits.
§Fisher's (protected) least significant difference at p = 0.05.

Table 21. Concentrations† of Cd, Cr, Cu, Ni, Pb, and Zn in corn stover collected fall 1990.

Treatment	Cd	Cr	Cu	Ni	Zn
			mg kg^{-1}		
Control	nd‡	0.28±0.66a§	9.9±0.9b	nd	74±31b
1	0.59±0.24	0.50±0.41a	9.6±1.3b	0.46±0.40b	94±26b
2	nd	2.72±1.88a	14.6±2.0a	7.05±4.17a	342±85a
3	nd	1.85±1.36a	15.1±1.0a	1.58±0.78b	89±14b

†Values of Pb below detection limits of 1.0 mg kg^{-1}
‡nd = not detectable for concentrations of Cd below 0.5 mg kg^{-1} and Ni below 0.2 mg kg^{-1}.
§Values in columns followed by the same letter were not significantly different at p = 0.05.

This was an indication of the unavailable forms of these two elements occurring in the soils, as seen in the soil chemical fractionation results (Table 15 and Figures 15 and 18). No Cr or Pb were found in the most available soil fractions: water-soluble and exchangeable.

Copper uptake by corn was substantially greater than Cr and Pb. There were not, however, large differences in Cu uptake between sludge-treated plots and controls. Copper was not accumulating into the plants as soil loadings increased. Most of the Cu added to the soils occupied unavailable soil fractions, although there were increases in water-soluble and exchangeable fractions at the highest loadings to the control plots (Table 15 and Figure 16). The increases in these two soil fractions corresponded to small increases of Cu uptake into corn diagnostic tissue and stover from sludge-treated plots compared to controls. The increases in plant uptake of Cu from soils of sludge-treated plots compared to controls, however, were less than a factor of two, while the differences in total soil Cu was as much as a factor of 10 (Table 15).

Soil loadings of Cd, Ni, and Zn may be of greater concern than that of Cr, Cu, and Pb. Cadmium and Ni appeared to be accumulating in the tissue of plants grown on sludge-treated plots. It may be difficult to draw definitive conclusions of Cd accumulation in plants because Cd concentrations of soil and plant samples from control plots were below analytical detection limits. Nickel, because of the available forms that it occupied, was 10 to 35 times or more concentrated in plant tissue from Treatment 2 plots compared to controls, whereas there was only an eight-fold difference between total soil Ni of Treatment 2 and control soil samples (Table 15). Plant uptake responses to Zn applications to soil were not as pronounced as that of Ni. It was, however, as much as four times the concentration in plants from control plots compared to those grown on Treatment 2 plots.

Cadmium is a known animal carcinogen. Any Cd applied to soil may result in its uptake by plants, which must be limited when plants are used by animals as food sources and in human food products. Potential plant toxicities resulting from high plant Ni and Zn uptake would be the primary concern of loading these two elements onto soil.

Soybean

Soybean grain yield data for 1985 through 1989 is in Table 22. Planted soybeans failed to grow on Treatment 2 plots during these years. Average soybean grain yields in plots of Treatments 1 and 3 were

Table 22. Soybean grain yield at 13% moisture for 1985 to 1989.

Year	Control	Treatment 1	Treatment 3
	- - - - - - - - - Mg ha^{-1} @ 13% Moisture - - - - - - - - -		
1985	1.9±0.3a†	1.4±0.3b	1.4±0.3b
1986	1.4±0.2a	1.0±0.4b	0.8±0.2b
1987	1.5±0.1a	1.7±0.2a	1.5±0.4a
1988	3.9±0.1a	4.3±0.3a	3.8±0.7a
1989	2.5±0.2a	2.5±0.3a	2.3±0.5a
Average	2.3±1.0a	2.2±1.2a	2.0±1.12b
lsd‡ (Year*Treatment) = 0.5			

†Values in the rows followed by the same letter were not significantly different at p = 0.05.
‡Fisher's (protected) least significant difference at p = 0.05.

significantly lower than those of control plots in 1985 and 1986. However, in 1987 to 1989 there were no differences between treatments (Table 22).

Trace element concentrations in soybean grain are listed in Table 23. All values of Cd, Cr, and Pb were below analytical detection limits of 0.5, 0.2, and 1.0 mg kg^{-1}, respectively. Overall, there were no differences between the concentrations of Cu and Zn in soybean grain of the control plots versus plots of Treatments 1 and 3. Concentrations of Ni, however, were significantly lower in soybean grain from control plots compared to that from plots of Treatments 1 and 3. This reflected the differences also found in the total Ni concentrations of the soils and the soil chemical fractionation data (Table 15).

Although Cd was not measured in detectable levels in the soybean grain, this only may mean that detection limits must be lowered to see differences among treatments. A concern not addressed by this study is

Table 23. Concentrations† of Cu, Ni, and Zn in soybean grain for 1985 and 1987 to 1989.

Year	Control	Treatment 1	Treatment 3
	- - - - - - - - - - - - - - - - mg kg^{-1}	- - - - - - - - - - - - - - - -	
		Copper	
1985	10.9±0.3a‡	11.3±0.6a	10.7±0.8a
1987	10.5±0.3a	9.4±0.5b	10.5±0.6a
1988	14.9±2.6a	14.5±5.3a	13.5±3.8a
1989	10.3±0.6a	11.0±1.6a	10.0±0.8a
Average	11.6±2.3a	11.5±3.1a	11.2±1.3a
lsd§ (Year*Treatment) = not significant.			
		Nickel	
1985	15.0±5.9b	30.6±6.9a	27.6±1.5a
1987	14.3±1.7a	17.2±7.7a	22.1±1.7a
1988	13.8±2.8b	16.4±1.6b	20.8±3.4a
1989	10.1±3.9b	14.6±3.3b	18.8±0.8a
Average	13.3±4.0b	19.7±8.2a	22.3±3.8a
lsd (Year*Treatment) = not signficant.			
		Zinc	
1985	48.8±4.1a	51.8±1.7a	45.8±2.9a
1987	56.3±5.4a	51.7±4.6a	48.4±2.3a
1988	59.5±3.9a	60.2±7.9a	55.0±8.6a
1989	55.8±5.8a	57.0±1.7a	56.4±1.7a
Average	55.1±5.9a	55.2±5.6a	51.4±6.2a
lsd§ (Year*Treatment) = not signficant.			

†Cd, Cr, and Pb were below their analytical detection limits of 0.5, 0.2, and 1.0 mg kg^{-1}, respectively. Plants did not grow on plots of Treatment 2.

‡Values in rows followed by same letter were not significantly different at $p = 0.05$.

§Fisher's (protected) least significant difference at $p = 0.05$.

Cd uptake into other plant parts, especially leaves and stems. Although not normally harvested for animal or human consumption, these plant parts are food for indigenous animals. This may represent a mode of entry into the human food chain when the animals are hunted and consumed.

As pointed out above, soybean did not grow on plots of Treatment 2, probably due to phytotoxic Zn (and possibly Cr and Ni) concentrations in the soils of these plots. Zinc concentration in the surface soil of Treatment 2 averaged more than 2400 mg kg^{-1}, 10 times greater than that found in soils of control plots (Table 15). Total soil Ni and Cr also were significantly higher (8 and 5 times greater, respectively) in Treatment 2 compared to the Control. For Ni, concentrations in soils of Treatments 2 and 3 were about the same. The chemical forms of Ni extracted from soils of Treatment 2, however, were those considered more bioavailable than those of Treatment 3 (Table 15 and Figure 15). On the other hand, chemical forms of Cr in soils of Treatment 2 were those residing in fractions with low bioavailable (Table 15 and Figure 15).

Risser and Baker (1990) concluded from work by other researchers that both Ni and Cr can be highly phytotoxic compared to other elements (i.e., Cd and Pb). The results of this study showed that even when Ni and Cr have high loading rates, phytotoxicity may not be apparent. Phytotoxicity of an element will depend more on the soil chemical fractions in which it resides than total soil loadings. Nevertheless, soil loadings of trace elements have an upper limit, which depends on soil and crop type, beyond which phytotoxicity and animal toxicity and carcinogenicity will be of concern.

Sorghum-Sudangrass

Sorghum-sudangrass biomass yields for 1985 through 1988 are shown in Table 24. The sludge amended plots had plant biomass yields as high or higher than the control plots, again evidence that sludge applications can help increase crop yields years after the last application. It should be stress, however, that too much sludge, especially when loadings of trace elements also are high, can depress yields. Plots of Treatment 2 had significantly lower yields than the other two treatments and were no better than control plots. This stress was probably from the high bioavailability of Zn and Ni, resulting in a reduction of yield compared to the other two treatments.

Trace elemental uptake by the sorghum-sudangrass biomass is listed in Table 25. The uptake of trace elements into this plant depended not only on the total concentration in the soil but also on the fractions in which they were found (Table 15). Only Pb was below analytical detection limits for all but one Treatment in one year. Cadmium, Cr, Cu, Ni, and Zn were greater in one or more sludge treatments compared to the control. For five of the six trace elements measured, loadings of trace elements to a soil resulted in greater uptake into the plants, which can be of concern to animals, in human nutrition, and to the plants growing on the soil. However, plant uptake also will depend on the soil fractions in which the elements reside and on the plant.

Plant uptake of Cd, Cr, Cu, Ni, and Zn elements reflected the concentrations found in the soils (Tables 15). The four fold or greater increase of Cd in sorghum-sudangrass tissue in plants from Treatment 1 compared to controls (assuming concentrations of Cd in plants from control plots were at the analytical detection limit of 0.5 mg Cd kg^{-1})

Table 24. Dry weight of sorghum-sudangrass harvested from 1985 to 1988.

	Control	Treatment 1	Treatment 2	Treatment 3
	- - - - - - - - - - - - - - - - Mg ha^{-1}	- - - - - - - - - - - - - - - -		
1985	7.2±0.8b†	9.6±1.6a	6.5±0.3b	8.3±1.9ab
1986	12.0±1.3b	13.5±1.2ab	11.8±1.2b	15.2±1.8a
1987	7.7±0.8b	9.7±0.9a	7.2±0.6b	10.1±1.6a
1988	9.6±1.8a	11.3±3.5a	8.2±2.4a	11.9±1.7a
Average	9.1±2.2b	11.0±2.5a	8.4±2.4b	11.4±3.1a

lsd‡ (Year*Treatment) was not significant at $p = 0.05$.

†Values in rows followed by the same letter were not significantly different at $p = 0.05$.
‡Fisher's (protected) least significant difference.

should be of special concern and highlighted the need to limit soil loadings of this element. Nickel and Zn did not appear to present as much of an uptake problem with this crop as Ni did in corn grain and stover and Zn did in corn stover. Sorghum-sudangrass grown on Treatment 2 plots had about three times higher Ni and Zn concentrations compared to plants from control plots.

Trace Element Soil Testing Extraction Methods

Concentrations of Cd, Cr, Cu, Ni, Pb, and Zn in surface soils from control and sludge-treated plots collected from 1986 to 1990 and extracted with 0.1*M* HCl are listed in Table 26. In general, concentrations of elements extracted did not vary greatly among years within the same treatment, although significant differences did exist. These differences may have had more to do with how the plots were sampled from one year to another, however, (depth of sampling and points from which subsamples were collected) rather than real differences in

Table 25. Concentration of Cd, Cr, Cu, Ni, Pb, and Zn in sorghum-sudangrass tissue collected in 1985, 1987, and 1988.

Year	Control	Treatment 1	Treatment 2	Treatment 3
	- - - - - - - - - -	- - - - mg kg^{-1}	- - - - - - - - - -	- - - - - - - - - -
		Cadmium		
1985	nd†	3.5±0.8a‡	1.2±0.4b	0.8±0.0b
1987	nd	2.1±0.4a	0.6±0.1b	0.6±0.1b
1988	0.57±0.19b	1.2±0.1a	0.6±0.1b	nd
Average	nd	2.2±1.0a	0.7±0.3b	0.6±0.2b
lsd§ (Year*Treatment) = 0.4				
		Chromium		
1985	nd	0.2±0.1b	1.0±0.6a	0.5±0.2ab
1987	0.4±0.2b	1.0±0.8a	0.9±0.5ab	1.1±0.5a
1988	5.0±1.4b	6.9±1.7b	11.5±6.2a	5.1±3.1b
Average	1.9±2.4b	2.3±2.9b	3.6±5.5a	1.9±2.4b
lsd (Year*Treatment) = 1.9				
		Copper		
1985	17±4b	18±4b	27± 7a	19±1b
1987	11±1c	14±1b	15± 0b	16±1a
1988	13±2b	16±2ab	20± 7a	14±5b
Average	13±3c	16±3b	19±7a	16±3b
lsd (Year*Treatment) = 3				
		Nickel		
1985	3.3±1.4c	8.0±1.4b	16.0±2.5a	6.6±1.2b
1987	1.4±0.2c	2.2±0.7bc	6.5±1.8a	2.7±0.4b
1988	5.5±1.1b	5.9±0.8b	15.2±5.3a	5.0±2.8b
Average	3.2±2.0c	4.6±2.7b	11.1±5.5a	4.2±2.2b
lsd (Year*Treatment) = ns¶				
		Lead		
1985	nd	nd	nd	nd
1987	nd	nd	1.4±1.8	nd
1988	nd	nd	nd	nd
Average	nd	nd	nd	nd
lsd (Year*Treatment) = ns				

Table 25 (cont'd)

Year	Control	Treatment 1	Treatment 2	Treatment 3
	- - - - - - - -	- - - - - - - mg kg^{-1}	- - - - - - -	- - - - - - - -
		Zinc		
1985	71±15c	151±21b	265±63a	114± 7bc
1987	87±18c	112±15b	283±51a	100± 5bc
1988	106±15bc	121± 7b	304±31a	87± 5c
Average	90±20c	124±22b	284±49a	101±11c
lsd (Year*Treatment) = 62				

†nd means not detectable. Concentrations of Cd, Cr, and Pb less than 0.5, 0.2, and 1.0 mg kg^{-1}, respectively, were below analytical detection.
‡Values in rows followed by the same letter were not significantly different at p = 0.05.
§Fisher's (protected) least significant difference at p = 0.05.
¶ns means not significant at p = 0.05.

extractable elemental concentrations. This information is useful when examining plant uptake data of trace elements. Although significant differences among treatment and years were observed for plant uptake of some trace elements (Tables 19, 20, and 25), HCl extractable trace elements alone would not have necessarily predicted them. This indicated that factors other that soil elemental concentrations affected plant uptake (e.g., yearly weather conditions and plant genetic differences).

Table 27 lists the concentration of trace elements extracted using several different techniques, including 0.1*M* HCl. Three techniques that used chelating agents [i.e., AB-DTPA, DTPA-TEA, and EDTA-$Ca(NO_3)_2$] extracted similar amounts of the six trace elements. AB-DTPA, however, was more effective in extracting analytically measurable quantities of

Table 26. Concentrations of Cd, Cr, Cu, Ni, Pb, and Zn in surface soils from 1986 to 1990 extracted with 0.1*M* HCl.

Year	Control	Treatment 1	Treatment 2	Treatment 3
	- - - - - - - - - - - -	mg kg^{-1}	- - - - - - - - - - - -	
		Cadmium		
1986	0.3±0.1c†	8.6±1.9a	2.2±0.5b	2.9±0.3b
1987	0.7±0.2c	7.4±1.0a	3.1±0.3b	2.9±0.3b
1988	0.4±0.0c	6.2±0.8a	2.9±0.0b	2.7±0.3b
1989	0.6±0.1c	6.4±1.0a	3.0±0.1b	2.6±0.2b
1990	0.6±0.1c	6.9±0.5a	3.1±0.1b	2.8±0.3b
Average	0.5±0.2c	7.1±1.4a	2.9±0.4b	2.8±0.3b
lsd‡ (Year*Treatment) = 0.9				
		Chromium		
1986	3±1c	50± a	29±12b	46± 2a
1987	10±4c	52±12a	19± 7bc	26± 3b
1988	3±1c	43± 6a	18± 4b	18± 5b
1989	8±2c	49± 8a	23± 7b	23± 2b
1990	7±1c	60± 6a	28± 6b	27± 2b
Average	6±3d	51± 9a	23± 8c	28±10b
lsd (Year*Treatment) = 8				
		Copper		
1986	11± 4b	58± 6a	38±27ab	NA§
1987	30±14c	66±16b	59±29bc	181±16a
1988	12± 3c	51± 7b	61±21b	150±35a
1989	23± 5d	55±14c	79±28b	166±20a
1990	22± 3d	59±10c	87±22b	173± 9a
Average	20±10c	58±11b	65±29b	167±23a
lsd (Year*Treatment) = ns¶				
		Nickel		
1986	11± 1	NA	NA	NA
1987	22± 9c	45±10c	164±19a	111±15b
1988	9± 2d	35± 6c	161±28a	115±12b
1989	25±12c	38±10c	150±20a	106± 5b
1990	16± 2d	38± 8c	154±14a	113± 6b
Average	17± 9d	39± 9c	157±20a	111±10b
lsd (Year*Treatment) = ns				

Table 26 (cont'd)

Year	Control	Treatment 1	Treatment 2	Treatment 3
	- - - - - - - - - - - - - - -	mg kg^{-1}	- - - - - - - - - - - -	
		Lead		
1986	4.2±1.0b	5.2±3.0b	2.9±2.1b	17.6±4.3a
1987	9.5±3.3a	8.6±2.3a	4.8±3.1a	7.7±3.2a
1988	5.4±1.5a	8.4±0.9a	5.7±0.7a	6.7±1.6a
1989	8.4±1.8a	8.6±1.5a	7.5±2.4a	10.5±0.9a
1990	6.7±0.7a	8.4±1.2a	7.2±1.9a	8.5±0.6a
Average	6.8±2.6bc	7.8±2.2b	5.6±2.6c	10.2±4.6a
lsd (Year*Treatment) = 3.2				
		Zinc		
1986	36± 7a	352±60a	NA	384±NAa
1987	125±25c	410±80b	1920±130a	438±65b
1988	49± 5c	305±43b	2050±170a	405±49b
1989	150±40c	320±72b	1730±160a	401±31b
1990	120±20d	360±59c	2080±110a	482±73b
Average	96±51d	350±69c	1950±190a	429±60b
lsd (Year*Treatment) = 124				

†Values in rows followed by the same letter were not significantly different at p = 0.05.
‡Fisher's (protected) least significant difference at p = 0.05.
§NA means not available.
¶ns means not statistically significant at p = 0.05.

Cr. In general, 0.1*M* HCl method extracted the greatest amount of trace elements and TCLP the least. Only 0.1*M* HCl was effective in extracting more than an average of 50% of the total Cd and Zn. The other methods generally extracted less than 40% of the total Cd, Cr, Cu, Pb, Ni, and Zn. All methods were ineffective in extracting more than about 10% of the total Cr and Pb and only 0.1*M* HCl extracted, on average, about 30% of the total soil Ni.

Table 27. Cadmium, Cr, Cu, Ni, Pb, and Zn concentrations from soil testing procedures performed on samples collected in 1990.

	Cd	Cr	Cu	Ni	Pb	Zn
	- - - - - - - - - - - -	- - - - - - - - - - - -	mg kg^{-1}	- - - - - - - - - - - -	- - - - - - - - - - - -	- - - - - - - -
			AB-DTPA			
Control	0.3±0.1	0.2±0.0	22± 3	5.4±1.5	4.1±0.6	50±13
Treatment 1	2.8±0.3	0.4±0.0	49± 8	17±4	6.0±1.2	125±23
Treatment 2	1.1±0.1	0.2±0.0	66± 5	67±3	3.3±0.6	520±16
Treatment 3	0.8±0.1	0.6±0.0	160±10	36±6	14± 1	110±15
LSD†	0.3	0.1	13	6.0	1.5	29
			DTPA-TEA			
Control	0.2±0.1	<0.04	12±2	4±1	2.1±0.6	38±11
Treatment 1	2.2±0.2	0.04±0.02	30±5	14±3	3.3±0.8	95±16
Treatment 2	0.8±0.0	<0.04	54±4	52±4	0.4±0.1	510±20
Treatment 3	0.6±0.1	0.06±0.00	105±7	29±5	8.2±0.6	78±13
LSD	0.2	-	9	5	1	25
			EDTA-$Ca(NO_3)_2$			
Control	0.4±0.1	<0.1	19±2	8±2	6.8±0.9	74±18
Treatment 1	3.4±0.4	<0.1	45±8	21±4	9.6±1.6	180±33
Treatment 2	0.9±0.1	<0.1	68±5	39±3	4.0±1.2	715±41
Treatment 3	0.9±0.1	<0.1	145±9	32±4	14±1	130±17
LSD	0.4	-	12	5	2	50
			HCl			
Control	0.6±0.1	7.1±1.3	22± 3	16± 2	6.7±0.7	115± 19
Treatment 1	6.9±0.5	60±6	59±10	38± 8	8.4±1.2	365± 59
Treatment 2	3.1±0.1	28±6	87±22	155±14	7.2±1.9	2080±105
Treatment 3	2.8±0.3	27±2	175± 9	115± 6	8.5±0.6	480± 73
LSD	0.5	7	19	13	NS‡	116

Table 27 (cont'd)

	Cd	Cr	Cu	Ni	Pb	Zn
	$mg\ L^{-1}$					
			TCLP			
Control	<0.05	0.02±0.01	0.05±0.01	0.2±0.0	0.1±0.1	1.9±0.2
Treatment 1	0.05±0.01	0.04±0.01	0.14±0.03	0.5±0.1	0.1±0.0	3.9±0.8
Treatment 2	<0.05	0.04±0.01	0.45±0.04	2.8±0.3	0.3±0.1	35± 2
Treatment 3	<0.05	0.06±0.02	0.82±0.06	1.8±0.2	0.3±0.1	4.3±0.5
LSD	-	0.01	0.04	0.2	0.1	0.9
			Total			
	$mg\ kg^{-1}$					
Control	<2.5	110±12	50± 8	52± 7	59± 7	220± 27
Treatment 1	7.7±0.9	515±68	110±21	94±17	80±10	460± 88
Treatment 2	4.4±0.1	565±28	285±13	415±49	165±12	2440±240
Treatment 3	4.8±0.6	820±118	515±64	430±49	175±20	800±140
LSD	1.3	128	62	62	22	250

†Fisher's (protected)least significant difference at p = 0.05.
‡NS = not significant at p = 0.05

The objective of using a soil test for trace elements is to quickly assess their potential bioavailability in soil. On this account, AB-DTPA and 0.1*M* HCl did as well as any other method in their correlation with water-soluble, exchangeable, and acid-soluble soil fractions (Table 28). The ability of 0.1*M* HCl to extract a high percentage of the Cd and Zn corresponded with results of sequentially extracting these two elements. Cadmium and Zn tended to remain in soil chemical fractions that continue to be plant available, such as the water-soluble, exchangeable, and acid-soluble fractions(Figure 8 and 13).

The 0.1*M* HCl extracting solution was less effective at removing Cr and Pb than Cd and Zn because of their tendency to reside in soil chemical forms that were less available. Chromium resided in the organic and Fe oxide fractions (Figure 15) and Pb occupied primarily the residual fraction (Figure 18). Both Cu and Ni have an availability that appeared intermediate in relation to the other two groups of elements as indicated by their fractionation in soil (Figure 16 and 17).

Simple correlation analysis of soil extraction methods with the water-soluble, exchangeable, and acid-soluble soil fractions resulted in many similar coefficients (Table 28). Cadmium, Cu, Ni, and Zn extracted using AB-DTPA, DTPA-$Ca(NO_3)_2$, EDTA-TEA, 0.1*M* HCl, and TCLP were highly correlated with water-soluble and exchangeable soil fractions, indicating that these soil testing methods provide information that can be used to help predict bioavailability. Data for Cr and Pb, however, were not as well correlated with these fractions. Chromium and Pb were in soil components not readily plant available and the correlations indicated this. When using testing methods to help determine the bioavailability of trace elements in a potentially contaminated soil, relative comparisons made with a similar soil that is not contaminated may be useful when plant uptake and sequential extraction data is lacking.

Results of TCLP indicated that the soils of this study would not be classified as hazardous wastes. A material can be regulated as hazardous when the concentrations of Cd, Cr, or Pb exceed 1.0, 5.0, and 5.0 mg L^{-1} (Federal Register, 1990b), respectively. None of the soils in

Table 28. Correlation coefficients, probability of significance, and number of pairs of soil test values with water-soluble, exchangeable, and acid-soluble fractions of trace elements.

	AB-DTPA	DTPA-TEA	EDTA	HCL	TCLP	Total
			Cadmium, Acid-Soluble			
r†	0.9433	0.9461	0.9580	0.9374	0.6813	0.9549
Prob‡	0.0001	0.0001	0.0001	0.0001	0.5229	0.0001
N§	12	12	12	12	3	12
			Chromium, Acid-Soluble			
r	0.6083	-0.5688	0.3630	0.7377	0.6703	0.8395
Prob	0.0124	0.1827	0.6370	0.0011	0.0045	0.0001
N	16	7	4	16	16	16
			Copper, Water-Soluble			
r	0.9553	0.9854	0.9698	0.9627	0.9871	0.9808
Prob	0.0001	0.0001	0.0001	0.0001	0.0001	0.0001
N	13	13	13	13	13	13
			Copper, Exchangeable			
r	0.9183	0.9063	0.9127	0.9289	0.8886	0.8648
Prob	0.0001	0.0001	0.0001	0.0001	0.0001	0.0003
N	12	12	12	12	12	12
			Copper, Acid-Soluble			
r	0.9708	0.9943	0.9823	0.9790	0.9965	0.9888
Prob	0.0001	0.0001	0.0001	0.0001	0.0001	0.0001
N	16	16	16	16	16	16
			Nickel, Water-Soluble			
r	0.9503	0.9658	0.9622	0.9348	0.9589	0.7401
Prob	0.0001	0.0001	0.0001	0.0001	0.0001	0.0092
N	11	11	11	11	11	11
			Nickel, Exchangeable			
r	0.9872	0.9878	0.9300	0.9435	0.9650	0.7983
Prob	0.0001	0.0001	0.0001	0.0001	0.0001	0.0002
N	16	16	16	16	16	16
			Nickel, Acid-soluble			
r	0.9537	0.9604	0.9231	0.9930	0.9890	0.9501
Prob	0.0001	0.0001	0.0001	0.0001	0.0001	0.0001
N	16	16	16	16	16	16

Table 28 (cont'd)

	AB-DTPA	DTPA-TEA	EDTA	HCL	TCLP	Total
			Lead, Exchangeable			
r	0.9961	0.9977	0.9978	-0.1771	-0.2802	0.6356
Prob	0.0566	0.0430	0.0423	0.8867	0.8192	0.5615
N	3	3	3	3	3	3
			Lead, Acid-Soluble			
r	0.3010	0.2360	-0.0156	0.0970	0.8499	0.8804
Prob	0.2573	0.3971	0.9542	0.7208	0.0001	0.0001
N	16	15	16	16	16	16
			Zinc, Water-Soluble			
r	0.5212	0.5307	0.4993	0.5910	0.5350	0.5676
Prob	0.1853	0.1760	0.2078	0.1229	0.1719	0.1422
N	8	8	8	8	8	8
			Zinc, Exchangeable			
r	0.9868	0.9915	0.9854	0.9793	0.9894	0.9490
Prob	0.0001	0.0001	0.0001	0.0001	0.0001	0.0001
N	16	16	16	16	16	16
			Zn, Acid-Soluble			
r	0.9858	0.9823	0.9792	0.9915	0.9844	0.9937
Prob	0.0001	0.0001	0.0001	0.0001	0.0001	0.0001
N	16	16	16	16	16	16

†Pearson correlation coefficient
‡Significance probability of the correlation under the null hypothesis that the correlation is zero
§Number of observations used to calculate the coefficient

this study were greater than these maximum concentrations (Table 28). Currently, no maximum concentrations of Cu, Ni, and Zn would classify a soil as hazardous. Plant uptake and soil data from previous sections, however, indicated that the soils had levels of Zn, and possibly Ni and Cr, that were toxic. Also, plant toxicities to high levels of soil Cu are known to occur (Jones, 1991).

This discussion has no interest in weighing the merits of using TCLP to determine the toxicity characteristics of a material associated with landfilling it. The TCLP testing method and elemental concentrations used to characterize wastes as hazards, however, are simply not appropriate for purposes of determining trace element availability in soils. The soil in this study would have had to contain significantly higher concentrations of Cd, Cr, and Pb in order to fail TCLP. Concentrations of Cd residing in soils of Treatment 1 already were at levels that may make plants especially vulnerable to its uptake. Additionally, a soil may be contaminated with phytotoxic levels of Cu, Cr, Ni, and Zn and not be considered a hazardous waste under TCLP guidelines. However, the soil concentration of these elements would still be of concern for the purpose of crop production. TCLP was not intended as a soil test and should not be used in that fashion. If one is deciding the merits of treating a soil as a hazardous waste, TCLP may be of value, especially in a regulatory function. Soil passing TCLP, however, may still pose a hazard depending on its use. The quantities of trace elements determined by soil testing methods have been correlated with plant uptake. These tests should give better insights into the nature of the hazard to plants and to animals using the plants as a source of food than does TCLP.

SUMMARY AND CONCLUSIONS

Surface soils to which municipal wastewater sludges were applied from 1977 to 1986 were sequentially extracted and trace elements were measured in each of eight fractions: (1)water-soluble; (2)exchangeable, 0.5*M* $Ca(NO_3)_2$; (3)acid-soluble, 0.44*M* CH_3COOH + 0.1*M* $Ca(NO_3)_2$; (4)Mn oxide, 0.1*M* $NH_2OH{\cdot}HCl$; (5)organic, 0.1*M* $Na_4P_2O_7$; (6)amorphous and (7)crystalline Fe oxides, 0.175*M* $(NH_4)_2C_2O_4$; and (8)residual, $HCl/HNO_3/HF$. Results of chemical fractionation revealed that Cd, Cu, and Zn resided primarily in the exchangeable and acid-soluble fractions, Cr in the organic and Fe oxide fractions, Ni in the acid-soluble and Fe oxide fractions, and Pb in the residual fraction. Loadings of trace elements to the soil resulted in Cd, Ni, and Zn occupying soil fractions that were available for plant uptake (i.e., water-soluble, exchangeable, and acid-soluble fractions). Copper loadings occupied primarily the acid-soluble fraction, although elevated levels were in both the water-soluble and exchangeable fractions. Compared to soils from control plots, loadings of Cr resulted in this element being found in greater concentrations in the organic and Fe oxide fractions. Lead loadings remained mostly in the residual fraction of soil, which is unavailable for plant uptake.

Plant uptake of trace elements was highly variable from year to year, plant part, and crop. Significantly greater yields of corn grain and sorghum-sudangrass occurred for plants grown on sludge-treated plots, whereas soybean grain yields on treated plots were equal to or less than those of controls to which no sludge was applied. Greater concentrations of Cd, Cr, Cu, Ni, and Zn occurred in corn diagnostic tissue; Cd, Ni, and Zn in corn grain; Cd, Cu, Ni, and Zn in corn stover; Ni in soybean grain; and Cd, Cr, Cu, Ni, and Zn in sorghum-sudan in plants of some sludge-treated plots compared with control plots.

Results of chemical soil fractionation and plant analysis suggested that soil loadings of Cd, Ni, and Zn increased their environmental availability, whereas Cr and Cu additions increased the environmental availability of these two elements to a smaller extent. Loadings of Pb to the levels seen in this study did not appear to significantly increase its environmental availability.

The TCLP method and guidelines are inappropriate to use when testing soils for potentially hazardous concentrations of trace elements. Soil test methods for trace elements that exhibit plant uptake (Cd, Cu, Ni, and Zn) generally correlated well with the most labile soil fractions (i.e., water-soluble, exchangeable, and acid-soluble). Only AB-DTPA and HCl correlated well with acid-soluble Cr. None correlated well with Pb, an indication of its low plant availability.

LIST OF REFERENCES

LIST OF REFERENCES

Baker, D.E., and M.C. Amacher. 1982. Nickel, copper, zinc and cadmium. *In* A.L. Page et al. (ed.) Methods of soil analysis. Part 2. 2nd ed. Agronomy 9:323-336.

Bartlett, R., and B. James. 1979. Behavior of chromium in soils. III. Oxidation. J. Environ. Qual. 8:31-35.

Bartlett, R., and B. James. 1988. Mobility and bioavailability of chromium in soils. p. 267-304. *In* J.O. Nriagu and E. Nieboer (ed.) Chromium in the natural and human environments. John Wiley & Sons, NY

Brown, J.R., and D. Warncke. 1988. Recommended cation tests and measures of cation exchange capacity. p. 15-16. *In* W.C. Dahnke (ed.) Recommended chemical soil test procedures for the North Central region. N. Central Reg. Publ. No. 449 (revised), October 1988, N. Dakota Agr. Exp. Sta., NDSU, Fargo, ND

Burau, R.G. 1982. Lead. p. 347-366. *In* A.L. Page, et al. (ed.) Methods of soil analysis. Part 2 2nd Ed. American Society of Agronomy, Madison, WI

Chang, A.C., A.L. Page, J.E. Warneke, and E. Grgurevic. 1984a. Sequential extraction of heavy metals following a sludge application. J. Environ. Qual. 13:33-38.

Chang, A.C., J.E. Warneke, A.L. Page, and L.J. Lund. 1984b. Accumulation of heavy metals in sewage sludge-treated soils. J. Environ. Qual. 13:87-91.

Chang, A.C., A.L. Page, and F.T. Bingham. 1982. Heavy metal absorption by winter wheat following termination of cropland sludge applications. J. Environ. Qual.11:705-708.

Day, P.R. 1965. Particle fractionation and particle-size analysis. p. 545-567. *In* C.A. Black (ed.) Methods of soil analysis. Part 1. No. 9. American Society of Agronomy Inc., Madison, WI

Dowdy, R.H., W.E. Larson, J.M. Titrud, and J.J. Latterell. 1978. Growth and metal uptake of snap beans grown on sewage sludge-amended soil: A four-year field study. J. Environ. Qual. 7:252-257.

Eckert, D.J. 1988. Recommended pH and lime requirement tests. p. 6-8. *In* W.C. Dahnke (ed.) Recommended chemical soil test procedures for the North Central region. N. Central Reg. Publ. No. 449 (revised), October 1988, N. Dakota Agr. Exp. Sta., NDSU, Fargo, ND

Elliott, H.A., B.A. Dempsey, and P.J. Maille. 1990. Content and fractionation of heavy metals in water treatment sludges. J. Environ. Qual. 19:330-334.

Emmerich, W.E., L.J. Lund, A.L. Page, and A.C. Chang. 1982a. Solid phase forms of heavy metals in sewage sludge-treated soils. J. Environ. Qual. 11:178-181.

Emmerich, W.E., L.J. Lund, A.L. Page, and A.C. Chang. 1982b. Predicted solution phase forms of heavy metals in sewage sludge-treated soils. J. Environ. Qual. 11:182-186.

Federal Register. 1990a. Environmental Protection Agency. Part II. 40 CFR Part 261 et al. 55(61): 11798-11877.

Federal Register. 1990b. Environmental Protection Agency. Part V. 40 CFR Part 261 et al. 55(126): 26986-26998.

Fujii, R., and R.B. Corey. 1986. Estimation of isotopically-exchangeable Cd and Zn in soils. Soil Sci. Soc. Am. J. 50:306-308.

Hickey, M.G., and J.A. Kittrick. 1984. Chemical partitioning of cadmium, copper, nickel and zinc in soils and sediments containing high levels of heavy metals. J. Environ. Qual. 13:372-376.

James, B. R., and R.J. Bartlett. 1983. Behavior of chromium in soils: V. Fate of organically complexed Cr(III) added to soil. J. Environ. Qual. 12:169-172.

John, M.K. 1972. Lead availability related to soil properties and extractable lead. J. Environ. Qual. 1:295-298.

Jones. J.B., Jr. 1991. Plant tissue analysis in micronutrients. p.477-521. *In* S.H. Mickelson (ed.) Micronutrients in agriculture 2nd ed. No. 4 Soil Sci. Soc. Am., Inc. Madison, WI

Kelling, K.A., D.R. Keeney, L.M. Walsh, and J.A. Ryan. 1977. A field study of the agricultural use of sewage sludge: III. Effect on uptake and extractability of sludge-borne metals. J. Environ. Qual. 6:352-358.

Knuden, D., and D. Beegle. 1988. Recommended phosphorus tests. p. 12-15. *In* W.C. Dahnke (ed.) Recommended chemical soil test procedures for the North Central region. N. Central Reg. Publ. No. 449 (revised), October 1988, N. Dakota Agr. Exp. Sta., NDSU, Fargo, ND

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.

Miller, J.E., J.J. Hassett, and D.E. Koeppe. 1975. The effect of soil properties and extractable lead on lead uptake by soybeans. Commun. Soil Sci. Plant Anal. 6(4):339-347.

Miller, W.P., D.C. Martins, and L.W. Zelazny. 1986. Effect of sequence in extraction of trace metals from soils. Soil Sci. Soc. Am. J. 50:598-601.

Misra, S.G., and G. Pandey. 1976. Evaluation of suitable extractant for available lead in soils. Plant Soil 45:693-696.

Murphy, L.S., and L.M. Walsh. 1972. Correction of micronutrient deficiencies with fertilizers. *In* J.J. Mortvedt (ed.) Micronutrients in agriculture. Soil Sci. Soc. Am., Inc., Madison, WI

Nelson, J.L., L.C. Boawn, and F.G. Viets, Jr. 1959. A method for assessing zinc status of soils using acid-extractable zinc and "titratable alkalinity" values. Soil Sci. 88:275-283.

Rhoades, J.D. 1982. Cation exchange capacity. p.149-157. *In* A.L. Page et al. (ed.) Methods of soil analysis, Part 2. Agronomy 9 (2nd Ed.):149-165. Am. Soc. of Agron., Inc., Madison, WI

Risser, J.A., and D.E. Baker. 1990. Testing soils for toxic metals. p.275-298. *In* R.L. Westerman (ed.) Soil testing and plant analysis. 3rd Ed. No. 3. Soil Science Society of America, Inc., Madison, WI

Sabbe, W.E. 1980. Handbook on reference methods for soil testing. The Council on Soil Testing and Plant Analysis. Univ. of Georgia, Athens, GA

SAS. 1985. SAS user's guide: Statistics, version 5 ed. SAS Institute Inc., Cary, NC

Schulte, E.E. 1988. Recommended soil organic matter tests. p.29-32. *In* W.C. Dahnke (ed.) Recommended chemical soil test procedures. North Central Region Publication No. 221 (revised). Bulletin No 499. North Dakota Agricultural Experiment Station, Fargo, ND

Shuman, L.M. 1979. Zinc, manganese, and copper in soil fractions. Soil Sci. 127:10-17.

Sims, J.T., and J.S. Kline. 1991. Chemical fractionation and plant uptake of heavy metals in soils amended with Co-composted sewage sludge. J. Environ. Qual. 20:387-359.

Soltanpour, P.N., J.B. Jones, Jr., and S.M. Workman. 1982. Optical emission spectrometry. p. 29-65. *In* A.L. Page et al. (ed.) Methods of soil analysis, Part 2. Agronomy 9 (2nd Ed.) Am. Soc. of Agron., Inc., Madison, WI

Sposito, G., L.J. Lund, and A.C. Chang. 1982. Trace metal chemistry in arid-zone field soils amended with sewage sludge: I. Fractionation of Ni, Cu, Zn, Cd, and Pb in solid phases. Soil Sci. Soc. Am. J. 46:260-264.

Stover, R.C., L.E. Sommers, and D.J. Silviera. 1976. Evaluation of metals in wastewater sludge. J. Water Pollu. Con. Fed.48:2165-2175.

Tessier, A., P.G.C. Campbell, and M. Bisson. 1979. Sequential extraction procedure for the speciation of particulate trace metals. Anal. Chem. 51:884-851.

Whitney, D.A. 1988. Micronutrient soil tests for zinc, iron, manganese, and copper. p. 20-22. *In* W.C. Dahnke (ed.) Recommended chemical soil test procedures for the North Central region. N. Central Reg. Publ. No. 449 (revised), October 1988, N. Dakota Agr. Exp. Sta., NDSU, Fargo, ND