RELATIVE CONTRIBUTION OF SOIL PROPERTIES TO MODIFYING THE PHYTOTOXICITY AND BIOACCUMULATION OF CADMIUM, LEAD AND ZINC TO LETTUCE

By

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Submitted to the Faculty of the Graduate College of the Oklahoma State University In partial fulfillment of The requirements for The Degree of DOCTOR OF PHILOSOPHY December 2003 RELATIVE CONTRIBUTION OF SOIL PROPERTIES TO MODIFYING THE PHYTOTOXICITY AND BIOACCUMULATION OF CADMIUM, LEAD AND ZINC TO LETTUCE

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ACKNOWLEDGMENTS

I wish to express my sincere appreciation to my major advisor, Dr. Nick Basta, for his supervision, guidance, and friendship. My sincere appreciation extends to my other committee members, Dr. Mark Payton, Dr. Hailin Zhang, and Dr. Jeffrey Hattey, whose assistance was also invaluable. Finally, I would like to thank the Environmental Institute for financial support.

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INTRODUCTION

Contamination of soil with heavy metals is a serious, common, worldwide problem. Excess metal in soil poses a threat to human health directly via inhalation, ingestion of soil, and contamination of groundwater and indirectly through consumption of plants grown on contaminated soil. Heavy metals in soil are also of great concern to soil ecosystems where organisms are in direct contact with the soil.

To adequately protect or restore soil ecosystems, it is necessary to accurately characterize soils suspected or presumed to be contaminated with heavy metal by defining levels of metal in these soils that constitute a hazard to soil organisms. Ecological soil screening levels (Eco-SSL) are levels of contaminants that, below which, pose little or no risk to ecological receptors.

Ecological Soil Screening Levels

Ecological soil screening levels are concentrations of metal or other contaminants that, if exceeded, require further investigation in a site-specific, ecological risk assessment. One approach of terrestrial ecological risk assessment (ERA) involves the use of Eco-SSLs as the first step. If a contaminant concentration exceeds Eco-SSL value, then adjustments or modifications to the ERA can be considered. One adjustment is to consider the modifying effect of soil properties on metal bioavailability and ecotoxicity.

Initially, Eco-SSLs are being derived for 17 metals (including Cd, Pb and Zn) and 7 organic contaminants, deemed to be of concern at Superfund National

Priority List sites (U.S. EPA, 2000). Ecological soil screening levels are not clean-up standards but, rather, are screening levels intended to identify contaminants that need to be evaluated in a site-specific baseline ecological risk assessment. Establishing standardized soil screening levels will streamline the risk assessment process because contaminants below the screening level will not need to be considered further. Each contaminant will have a screening level for each of four groups of ecological receptors: plants, soil invertebrates, mammals, and birds. Plant Eco-SSLs are established based on available toxicity testing data obtained from peer-reviewed literature. For the derivation of plant and soil invertebrate Eco-SSLs, the ability of soil properties to modify contaminant ecotoxicity and bioavailability is being considered (U.S. EPA, 2000). Toxicity is generally acknowledged to be poorly related to total metal content, and metal bioavailability may be a better predictor of toxicity than total content (Adriano, 2001; Allen, 2002; Suave, 2002). Bioavailability is the portion of the total metal content that is absorbed into an organism and reacts with some biological receptor causing a response. In effect, it is the dose of contaminant that is available to a receptor site. Plant uptake and bioavailability of metal are related to metal solubility in the soil solution (Basta and Gradwohl, 2000; Adriano, 2001)

Understanding how soil properties modify the availability/solubility of different metals will provide insight into potential ecological risk. Bioavailability data can be used to relate soil properties to toxicity. For the purpose of developing Eco-SSLs, four soil parameters were initially selected to be included

as potential soil modifying factors to toxicity, pH, cation exchange capacity (CEC), clay, and organic matter (OC). But because there is insufficient data available for CEC and clay, only pH and OC are currently being considered as modifying factors. These two parameters are being used qualitatively to characterize metal availability as high, medium, and low (Table 1). For metal contaminants, for example, if soil pH and OC are high the bioavailability of the contaminant is expected to be low. Alternatively, if pH and OC are low the risk is expected to be high (U.S. EPA, 2000).

The primary objective of this work was to determine the relative contribution of soil physico/chemical parameters [e.g. pH, organic carbon (OC), percent Clay, amorphous AI and Fe oxides (FEAL), and cation exchange capacity (CEC)] to modifying metal (Cd, Pb, and Zn) bioavailability. Results from this research study may provide baseline chemical and biological data that can be used in the development of Eco-SSLs or in reducing the uncertainty associated with determining site-specific ecological risk. Current routine practices of investigating the nature and extent of metal contamination involve determining total metal content in soils, which is probably not as useful in characterizing risk. The quantity of metal used as the exposure point concentration (dose) in risk calculation is an implicit percentage of the total content that may be bioavailable. Often, very high percentages (e.g., 100%) of total metals are assumed to be bioavailable. While this assumption is conservative in terms of being protective of human health and the environment, it may not be a reasonable estimate of site conditions because the actual bioavailability of metals has not been accurately

assessed. The resulting calculated risk may overestimate the true risk of exposure to site media. Overestimation of risk could result in lengthy and costly site remediation that may not be warranted (Adriano, 2001; Suave, 2002; Allen, 2002). More information is needed concerning the relative contribution of soil properties to modifying metal bioavailability.

Soil Properties Affecting Metal Bioavailability

Although currently the U.S. EPA is focusing on the modifying effects of pH and organic matter (OC) other soil properties that have the potential to modify the availability of metals to ecological receptors (including clay content, FEAL, and CEC) also need to be considered in the development of Eco-SSLs.

Soil pH is often called the "master variable." It has the potential to modify metal solubility/availability in several ways. It controls dissolution/precipitation and therefore influences speciation of minerals. It regulates the ionization of pHdependent exchange sites on organic matter and metal oxide clay minerals. This can affect contaminant as well as micronutrient availability. The ionization of pHdependent functional groups on soil organic matter also affects stable organic complex formation (Sposito, 1989; McBride, 1994; Adriano, 2001; Sparks, 2003).

Soil organic matter (SOM) typically comprises from 0.5 to 5% by weight of mineral soils (Bohn et. al., 1985; McBride, 1994; Sparks, 2003), but its importance to soil chemistry is greater than these numbers imply. Organic matter has a large surface area (800 to 900 m² g⁻¹) rich in reactive functional groups (e.g. carboxyl or phenolic) (Bohn et. al., 1985; McBride, 1994; Sparks,

2003). It is the ionization of these groups, as mediated by pH, that imparts a high pH-dependent cation exchange capacity (150 to 300 cmol_c kg⁻¹) to soil organic matter (Bohn et. al., 1985; Adriano, 2001; Sparks, 2003). Metal bound to the CEC is removed from solution and so less available to plants. This type of binding, however, is not strong and the contaminant metal, in equilibrium with the soil solution, is readily re-supplied to solution if the equilibrium is shifted through, for example, precipitation, leaching, or plant uptake. A more stable form of metal complexation with soil organic matter is through chelation or binding to ligand functional groups (i.e., amine, thiol, carbonyl). For example, Cd, Pb, and Zn are soft acids and will complex with soft functional (N- or S-) groups on organic matter (Bohn et. al., 1985; Adriano, 2001; Sparks, 2003).

Clay minerals are a highly reactive component of soil characterized by having a particle size < 2.0 μ m and a large surface area. The reactions between clay minerals and metals are primarily attributed to cation exchange (CEC) or ligand exchange (specific adsorption) reactions that occur on amorphous metal oxides (FEAL). The permanent, negatively charged portion of the soil CEC is associated with isomorphously substituted 2:1 clay minerals such as smectite and montmorillonite. These clay minerals have a large surface area and high CEC. Montmorillonite, for example, has a surface area of 600 to 800 m² g⁻¹ and a CEC of 80 to 150 cmol_c kg⁻¹. The pH-dependent CEC sites are associated primarily with amorphous metal oxides and hydroxides. These amorphous metal oxides also have a large surface area. For example, Fe and Al oxides have a

specific surface area of 70 to 250 and 100 to 220 m² g⁻¹, respectively (Bohn et. al., 1985; Adriano, 2001; Sparks, 2003).

Interactions Between Metal and Soil Properties

Cadmium

Adriano (2001) stated that soil pH is the most important soil property controlling the bioavailability of Cd to plants. Mahler et al. (1978) found lettuce tissue Cd concentrations decreased with increased pH. At a soil Cd level of 320 mg kg⁻¹, lettuce (*Lactuca sativa*) accumulated between 540 to 780 mg Cd kg⁻¹ and chard accumulated > 1500 mg Cd kg⁻¹ in soils with pH < 5.7. For soils at the same Cd level but with pH > 7.4, Cd accumulation was < 300 mg kg⁻¹ for both lettuce and chard. Naidu and Harter (1998) found that Cd extractability, in four soils, decreased with increasing pH despite the addition of organic ligands. They concluded that pH and not OM was primarily responsible for controlling Cd solubility. Mitchell et al. (1978) grew lettuce in a slightly acid (pH 5.7) and an alkaline (pH 7.5) soil spiked with Cd at rates ranging from 0.1 to 640 mg kg⁻¹. They found that at soil Cd < 40 mg kg⁻¹ uptake in lettuce was greater in the calcareous soil. At > 40 mg Cd kg⁻¹, tissue Cd was higher in lettuce grown on the acidic soil. They theorized that in the acid soil the availability of Cd may have been controlled by specific adsorption on amorphous oxides. At higher Cd rates, when all adsorption sites were filled, tissue Cd concentration increased rapidly. In the calcareous soil, they theorized that Cd availability was being controlled by precipitation (Mitchell et al., 1978). Lagerwerff (1971) saw a pH effect on Cd

uptake and yield in radish. In radish grown on a soil with pH adjusted to 5.9 and 7.2, he found radish Cd uptake was 4, 11, and 8% higher, respectively, at soil Cd rates of 29.9, 165, and 299 mg kg⁻¹; and radish yield was reduced by 32, 29, and 37%, respectively, in the more acidic soil. This decreased solubility of Cd at higher pH may have been due to the formation of carbonate, sulfate, or phosphate minerals or increased available CEC sites at high pH (Adriano, 2001). Basta et al. (1993) measured the Cd sorption maximum on two soils from two long-term cropping experiments. Using path analysis, they determined that the direct effect of pH > OC > CEC for Cd adsorption. Street et al. (1977) found that sorption of Cd occurred even in sandy soils with low OM and CEC at pH > 7.0. They attributed sorption to precipitation of CdCO₃. Soil CEC can be a significant sink for Cd. In a sequential fractionation study, Chlopecka et al. (1996) found that in 14 soils with soil Cd levels ranging from 7.2 to 102 mg Cd kg⁻¹, an average of 36% of soil Cd was found in the exchangeable fraction and 23% was bound to oxides. Very little of the Cd was associated with the organic matter fraction (Chlopecka et al., 1996). Ion exchange was also found by Haghiri (1974) to be a more important binding mechanism on OM than chelation. At a constant pH of 6.5, organic matter was added to a clay soil to provide a CEC range of 17.1 to 30.5 (Haghiri, 1974). He found that oat (Avena sativa L.) Cd uptake decreased and yield increased with increased CEC. He and Singh (1993) also found the majority of soil Cd to be exchangeable in a sequential fractionation experiment. Three soil types (pH < 5.0) spiked with 27 mg Cd kg⁻¹ and with organic matter additions of 32% resulted in the exchangeable Cd fraction increasing from 26.9

to 53.6% in a sand from 51.6 to 63.3% in a sandy loam and from 56.3 to 65.5% in a clay loam. The decrease in pH due to additions of organic matter (pH 3.67) resulted in a reduction in the proportion of Cd bound to the oxide fraction by an average of from 11 to 19% in all three soil types. Additionally, the increase in CEC due to organic matter addition was significantly ($r^2 = 0.66$) correlated to Cd uptake by ryegrass (He and Singh, 1993).

Lead

The negative relationship between pH and Pb solubility plays a major role in controlling Pb bioavailability to plants (Adriano, 2001). Many studies have reported decreased Pb uptake by plants with increased pH. A clay loam soil with an average pH of 4.75 and a Pb content of 94 mg kg⁻¹ was limed to pH 6.85 (Cox and Rains (1972). After liming, the uptake of Pb in five plant species was reduced by an average of approximately 34%. Similarly, John and Van Laerhoven (1972) raised the pH of a silty clay loam from pH 3.8 to 5.2 and spiked it with 1000 mg Pb kg⁻¹. With the increased pH, lettuce tissue Pb concentration was reduced from 233.6 to 54.3 mg Pb kg⁻¹. They suggested that the lime reduced the solubility of Pb. Lead hydrolysis is controlled by pH, with insoluble Pb(OH)₂ predominating at pH > 8.0 (Adriano, 2001). Lagerwerff (1977) found a decrease in radish uptake of Pb with an increase of pH from 5.9 to 7.2. MacLean (1969) suggested that the solubility of Pb decreased with increasing pH primarily due to the increased ability of OM and amorphous oxides to complex Pb. In a sequential fractionation study, Chlopecka et al. (1996) found that in 14 soils at

soil Pb levels ranging from 14 to 7100 mg Pb kg⁻¹ an average of only 4.7% of soil Pb was found in the exchangeable fraction, while an average of 57% was found in the oxide fraction and 14% was associated with organic matter. This indicated that Pb was forming stable compounds and will not be bioavailable (Chlopecka et al., 1996). Riffaldi et al. (1976) determined the Langmuir Pb sorption maximum for 12 soils. The Pb sorption maximums ranged from 4.2 to 23.8 mg Pb kg⁻¹. They found Pb sorption maximum was highly correlated with soil CEC (r = 0.93, P < 0.01), OM (r = 0.86, P < 0.01), clay (r = 0.67 P < 0.05) and Fe or Mn oxides (r = 0.79, P < 0.01 and r = 0.83, P < 0.827 respectively). Basta et al. (1993) measured the Pb sorption maximum on two soils from two long-term cropping experiments. Using path analysis, they determined that the direct effect of pH >OC > CEC for Pb adsorption. Complexation with OM can be a major sink for soil Pb, either by CEC or in more stable complexes with OM ligands (Adriano, 2001). Soldatini et al. (1976) measured the Langmuir P sorption maximum of 12 Tuscan soils, which ranged from 4.4 to 48.3 meg 100 g⁻¹. They found that the soil properties most closely related to Pb sorption maximum were OM and clay content, with a small contribution by Mn oxide. To determine if Pb will be sorbed by specific adsorption, a follow up study using the same 12 Tuscan soils was conducted (Riffaldi et al., 1976). Langmuir Pb sorption maximums were measured in the presence of solution Ca to block soil exchange sites. Langmuir adsorption maximum ranged from 1.5 to 23.8 meg 100 g⁻¹, and mean overall reduction of 51%. All possible regression analysis showed that OM and clay still explained a maximum of the variation in Pb sorption maximum, but the decrease

in total sorption suggested that CEC was also important for Pb sorption (Riffaldi et al., 1976). Other studies have shown decreased plant uptake of Pb with organic matter additions to soil (Scialdone et. al., 1980; Zimdahl and Foster, 1976). John (1972) found greater extractable Pb and Pb uptake in lettuce and tomato plants in sandy loam soils than in clay soils. Both pH and clay were highly correlated to tissue Pb, while OM was not related to plant uptake. In high clay soils, CEC increases with increased pH can lead to increased Pb sorption (Adriano, 2001). Zimdahl and Skogerboe (1977) measured the Langmuir Pb sorption maximum in 18 soils. They used step-wise regression to determine which soil parameters (pH, texture, clay mineral type, OM, CEC) were most important in predicting the Langmuir Pb sorption maximum. Their results showed that CEC and pH explained most of the variation in soil Pb sorption maximum. Basta and Tabatabai (1992b) reported that increased CEC resulted in significantly decreased Pb availability.

Zinc

The interactions between Zn and the soil matrix effectively control Zn bioavailability (Adriano, 2001). Zinc availability in soils has been shown to be influenced by pH, clay, CEC, and OM. Basta et al. (1993) measured the Zn sorption maximum of two soils from two long-term cropping experiments. Using path analysis, they determined that the direct effect of OC > CEC > pH for Zn adsorption. Other studies have also shown that large amounts of Zn were associated with the soil CEC. Elgabaly (1950) found that for four different types

of clay saturated with Zn-acetate solutions a mean of 54% of the added Zn was NH_4 exchangeable. In alkaline soils, Zn precipitates as carbonates or hydroxides (Adriano, 2001). Lagerwerff (1971) saw a pH effect on Zn uptake and yield in radish. In a soil with pH adjusted to 5.9 and 7.2, he found radish Zn uptake was 10, 24, and 36% higher, respectively, at soil Zn rates of 10.1, 35, and 59.8 mg kg⁻¹ at the more acidic pH. At soil Zn levels of 320 mg Zn kg⁻¹, Mitchell et al. (1978) found lettuce Zn tissue levels to be 380 and 1585 mg Zn kg⁻¹ when grown on soils with pH 7.5 or 5.7 respectively. Cavallaro and McBride (1984) found that at pH below Zn hydrolysis sorption of Zn on two clay soils was up to 70% exchangeable with CaCl₂. Further, they found that the metal oxide soil fraction was a more important sink than OM complexation in sorbing the remainder of Zn. Removal of the oxide fraction substantially reduced Zn sorption, while removal of the OM fraction did not reduce Zn sorption. Across all treatments, Zn sorption increased with pH. At pH nearing Zn hydrolysis, sorption increased sharply (Cavallaro and McBride, 1984). In a sequential fractionation study (Chlopecka et al., 1996) found that in 12 soils at soil Zn levels ranging from 60 to 300 mg Zn kg⁻¹ an average of only 12% of soil Zn was found in the exchangeable fraction, while an average of 35% of soil Zn was found in the oxide fraction and 15% was associated with organic matter.

Statistical Techniques

The relationships between and among soil properties make it difficult to discern which soil property or combination of properties is affecting metal

bioavailability. Ionization of pH-dependent exchange sites on clay and organic matter is strongly and positively related to soil pH. Solubility of metal is negatively related to pH. Because amorphous Fe and Al oxide (FEAL) is a measure of metal oxide clay minerals, it is strongly positively correlated with clay. Similarly, because cation exchange sites occur on both clay and OC surfaces, it is positively correlated with both. Nonpolar soil organic matter will bind with nonpolar clay surfaces; consequently, there is a positive correlation between clay and OC.

The independent variables an investigator chooses to include in an experiment and subsequent statistical analysis implies a causal hypothesis to be tested. In other words, the investigator is theorizing that each independent variable may be having a causal effect on the dependent variable either directly or indirectly in combination with (mediated by or via) another variable also included in the model.

Correlation and multiple regression techniques are routinely used to examine relationships between and among variables. However, these techniques provide little information regarding the relative contribution of each parameter included in a model. The relative contribution of each independent variable is further obscured by intercorrelations between independent variables.

Path analysis (PA), a form of structural equation modeling (SEM), is a technique that can be used to augment traditional regression analysis and provide insight into the relative contribution of each parameter in explaining the variation of the dependent variable (Maruyama, 1998). A geneticist, Sewall

Wright, developed path analysis in 1918. He used PA to determine the relative importance of different genetic paths from parents to offspring (Wright, 1921, 1934). Since then, PA has been used extensively in plant breeding (Singh and Chowdhury, 1982; Boe and Ross, 1983; Gravois and Helms, 1992; Rodomiro and Langie, 1997) and ecology (Grace and Pugesek, 1998). Basta et al. (1993) were the first to use PA to describe the sorption of heavy metals by soil.

Structural equation modeling, including PA, uses a series of equations to describe a hypothesized structure of relationships. To use this technique, the investigator must first develop a conceptual model to be tested, specifying theorized relationships among variables. Through the use of PA, these hypothesized structures are tested and either rejected or accepted as consistent with the data (plausible) (Maruyama, 1998). Causation is not established simply because a path model is consistent with the data, but it can be said that the conceptual model may be valid. Failure of a path model to fit the data, however, results in rejection of the mechanistic hypothesis (Grace and Pugesek, 1998).

Structural equation modeling attempts to partition correlation (hypothesized causal effects) into direct and indirect effects, called decomposition of effects. As mentioned previously, PA begins with a conceptual model to be tested that specifies theoretical relationships among variables. This model is called the path model or diagram (Maruyama, 1998). An example of a path diagram is presented in Figure 1. In this example the independent (predictor) variables (pH, OC, and clay) are numbered 1 through 3. The dependent variable (number 4) is some biological endpoint (i.e., bioaccumulation, dry matter growth, or

germination). The direct effect of an independent variable on the dependent variable is denoted with a single-headed arrow. These are called the path coefficients (P_{ii}) and are estimated as the standard partial regression coefficients (slopes) from standardized least squares regression. The arrow represents a hypothesis that a direct causal relationship exists. It implies that a change in the independent variable will result in a change in the biological endpoint. Intercorrelation between the independent variables (rig) is denoted with doubleheaded arrows. These are simple correlation coefficients between independent variables. The indirect effect of an independent variable, or its effect due to (via) its intercorrelation with another variable, is the product of the path coefficient and the simple correlation coefficient ($P_{ij} * r_{iq}$). An example of an indirect effect is the effect of pH on metal bioavailability via (due to or mediated by) the intercorrelation between pH and CEC due to the pH dependence of cation exchange sites. While pH may have a direct effect on metal bioavailability due to its influence on solubility (speciation), it may also have an indirect effect on metal availability through its effect on the ionization of pH-dependent exchange sites (i.e., carboxyl and phenolic groups). So it can be said that pH has an indirect effect via (mediated by) CEC. The residual is that part of the variation of the dependent variable not explained by the model and is expressed as (Figure 1):

Residual =
$$(1 - R^2)^{\frac{1}{2}}$$
 (1)

where R^2 is the coefficient of determination from the multiple regression. The path total (r_{ij}) is the sum of direct and indirect effects and should be equal (considering round-off error) to the simple correlation coefficient between the

independent and dependent variable because the path analysis represents a partitioning of this correlation into its direct and indirect effects (Figure 1). The path total can be expressed as:

$$\mathbf{r}_{ij} = (\Sigma \ \mathsf{P}_{ij} * \mathbf{r}_{iq}) \tag{2}$$

where *i* is the independent variable, *j* is the dependent variable, and *q* is an index over all variables with a direct path to *i* and *j* (Maruyama, 1998). In the example shown in Figure 1, the results of the partitioning of the effects of the independent (predictor) variables can be expressed by the following normal equations:

Total effect of pH =
$$P_{14}$$
 + ($r_{12} * P_{14}$) + ($r_{13} * P_{14}$) (3)

Total effect of OC =
$$P_{24}$$
 + ($r_{12} * P_{24}$) + ($r_{23} * P_{24}$) (4)

Total effect of Clay =
$$P_{34} + (r_{13} * P_{34}) + (r_{23} * P_{34})$$
 (5)

Some additional requirements necessary for a valid path model (Loehlin,

1987) include the following:

- Unidirectional causal flow.
- Studentized (standardized) variables, so they are unitless and covariances, and correlations are identical.
- The effect of each independent variable should be unitary.
- The direct effects should be linear.

Path analysis is a technique that can be used to augment traditional regression analysis. By partitioning correlations into direct and indirect effects, the relative importance of hypothesized causal effects can be determined and appropriate emphasis given to each parameter included in the model. Another way that partitioning helps the investigator is when a low correlation can be

shown to be due to the opposition of a positive direct or indirect effect with a negative one. While the total correlation may be statistically insignificant, the partitioned effects may be guite strong and merit consideration that might otherwise have been lost (Singh and Chowdury, 1982). While PA cannot prove a mechanistic hypothesis, it can confirm or refute the plausibility of the investigator's proposed model. Disproving a model is valuable in that it provides mechanistic insight and guidance for future experimentation. For example, Singh and Chowdhury (1982) were trying to determine which parameters contributed the most to the production of oil in mustard (Brassica juncea). They started out with a daunting number of parameters (eight) to consider for plant selection including primary branching, secondary branching, plant height, pods per main shoot, seeds per pod, seed weight, percent oil, and seed yield per plant. Of the eight original parameters included, the results of the PA showed that for breeding mustard for maximum yield selection should be based on three of the original eight parameters: secondary branching, plant height, and seed weight. This insight into the parameters most strongly contributing to increased yield provided guidance for subsequent plant selection (Singh and Chowdhury, 1982).

Objective

While there is no question that soil properties significantly modify the bioavailability and toxicity of metals to biological receptors, there is still a great deal of confusion as to the relative contribution of an individual soil property to modify metal bioavailability and ecotoxicity. Most studies consider only one or a

very few soil properties. This may be a shortcoming considering the tremendous intercorrelation between soil properties. To add to the confusion, many soil properties have more than one potential mechanism of effect. For example, clay has a CEC component and an amorphous metal oxide (specific adsorption) component. Often called the "master variable," pH affects solubility (precipitation) and the availability of pH-dependent cation exchange sites. While the task of unraveling this mechanistic muddle is daunting, it is imperative for making remediation decisions that will be protective of human and ecological health and, at the same time, be monetarily sound.

The objective of this study was to examine the relationship between soil chemical parameters (pH, OC, CEC, clay, and FEAL) and metal bioavailability in an attempt to provide a measure of the relative contribution of each soil property, individually and in combination, on modifying the bioavailability of metals (Cd, Pb, and Zn).

To estimate the bioavailability of metal in soil, the response of lettuce (*Lactuca sativa*) biological endpoints [bioaccumulation, dry matter growth (DMG) and germination], to Cd, Pb, and Zn-spiked soils were determined. Twenty one soils were spiked at only one level of each metal. This insured that any variation in lettuce response to metal would be due to the modifying effects of the soil properties.

MATERIALS AND METHODS

Selection of Soils

Forty soils were collected from Oklahoma and Iowa representing a wide range of soil properties including pH, OC, and percent clay, FEAL, and CEC. Chemical and physical properties were measured on collected soils, both A and B horizons were considered, and a sub-set of 21 soils were selected to represent a wide range in soil properties. Table 2 lists the soil taxonomic classification and series names of selected soils.

Background metal levels (Cd, Pb, Zn) in selected soils were determined by acid digestion using microwave (CEM MDS 2100, CEM Corporation, Matthews, NC, USA) according to U.S. EPA Method 3051(U.S. EPA 1994). Metal levels were within the range considered typical (Adriano, 2001) for uncontaminated soils (Table 2).

Soil Contaminant Spiking

Soils were spiked with reagent grade Cd(NO₃)₂ at 300 mg Cd kg⁻¹, Pb(NO₃)₂ at 2000 mg Pb kg⁻¹, or Zn(NO₃)₂ at 300 mg Zn kg⁻¹. Soil spiking contaminant levels were established from initial rangefinder plant and earthworm bioassays. Soils were spiked with only one metal to avoid competitive adsorption effects (Basta and Tabatabai, 1992a). Spiking solutions were prepared using metal salts and deionized water. One liter of spiking solution was mixed with 5.0 kg of soil. Additional deionized water was added to form a saturated paste and was thoroughly mixed. The spiked soils underwent three wet-dry cycles at 105 °

18

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C for 24 h. Heavy metals added as salt to soil can result in "salt effect" where metal availability is greater in spiked soil than non-spiked contaminated soil. The three wet-dry cycles minimize the salt effect by increasing the reaction between the soil matrix and metal contaminants (Logan and Chaney, 1983).

Soil salinity, as measured by the electrical conductivity (EC) of a watersaturated soil paste (Rhoades, 1996), was measured in spiked soils to insure that metal salt spiking had not increased salinity enough to inhibit germination. Electrical conductivity of spiked soils was determined after the second wet-dry cycle. Soils that had EC > 1.5 dS m⁻¹ were leached with deionized water until the soil EC < 1.5 dS m⁻¹. Spiked soils that had EC < 1.5 dS m⁻¹ were not leached.

The final metal concentration of spiked soils was confirmed by acid digestion using microwave according to U.S. EPA Method 3051 to be within 10% of total spiked metal level.

Soil Chemical and Physical Properties

All analyses were performed on duplicate samples of air-dried soil (< 2 mm). Soil pH was determined in 1:1 soil: deionized water suspension using a combination pH electrode (Thomas, 1996). Because metal salt addition can cause acidification due to metal hydrolysis (Basta and Tabatabai, 1992b), soil pH was measured on control (unspiked) and on metal-spiked soils. Soil pH measured on metal-spiked soils was used for all statistical analyses using soil pH. Soil organic carbon (OC) was determined by oxidation of organic C by acid dichromate reduction (Heanes, 1984). Soil texture was determined by the

hydrometer method, following pretreatment with hydrogen peroxide (H_2O_2) to remove OM (Gee and Bauder, 1986). Hydrometer readings were taken at 30 sec and 7 h 14 sec to determine the sand and clay content, respectively. Silt was determined by difference (100% - sand - clay = silt).

Amorphous Fe and Al oxide content was determined by acid ammonium oxalate extraction (McKeague and Day, 1996) and CEC was determined using the unbuffered salt (BaCl₂) extraction method (Sumner and Miller, 1996)

Metal content in pore water was measured in a saturated paste (Rhoades, 1996). Pastes were allowed to equilibrate for 48 h, then centrifuged at 10,000 rpm for 15 min. The supernatant was filtered through a 0.45 μ m membrane filter, acidified with trace metal HCI, and analyzed by ICP.

Lettuce Bioassay

Soil (800 g) was mixed with 50%, by volume, vermiculite in 1 L pots. To prevent metals from leaching, the pots were not allowed to drain. Twenty lettuce (*Lactuca sativa var. Paris Island Cos*) seeds were planted per pot. Three replicates of each metal treatment were planted in a completely randomized design. Plants were grown in a controlled environment growth chamber with 18 h of light day⁻¹, daytime temperatures of 20°C, and night temperatures of 18.5°C. Soil plant nutrition levels were tested. All soils had adequate levels of plant nutrients after fertilizer addition of Miracle GroTM (15% N + 30% P₂O₅ + 15% K₂O). To balance nitrogen due to metal-NO₃ salt additions with soil spiking, an additional 200 mg kg⁻¹ of N was applied to the control pots as NH₄NO₃. Percent

germination was determined at 7 days. Pots were thinned to 5 plants per pot at 14 days. Lettuce was harvested after 40 days, rinsed in deionized water, and dried at 70°C for 48 h and crushed by hand. The dried material was weighed to determine dry matter growth (DMG). Dry lettuce tissue (0.25 g) was predigested for 4 h in 10 mL of nitric acid. Predigested samples were digested at 140°C for 4h, or until clear (Zarcinas et al., 1987). Filtered (0.45 μ m) solutions were analyzed for metals by ICP-AES. To account for differences in lettuce biological endpoints due to differences in soil quality (i.e., acidity, texture), dry matter growth and germination are presented relative to their controls.

Statistical Analyses

Statistical analyses were performed using PC SAS Version 8.2 (SAS Institute Inc., Cary, NC). Prior to analysis, all data was studentized (standardized). This allows the data to be expressed in standard form with a mean of zero and a standard deviation of one (Loehlin, 1987). Because the simple or combined relationships between the biological endpoints and soil properties may not be linear, all statistical analyses were performed on the studentized (R) and studentized and then linearized data sets (Steele et al., 1997) to find the best fit model. Data set transformations used to linearize the studentized data sets were log₍₁₀₎ (log) or square root (SR). The best fit model for each relationship is presented.

The relationship between lettuce biological endpoints [metal bioaccumulation, relative dry matter growth (RDMG), and relative germination

(RG)] and soil properties (pH, OC, clay, FEAL, and CEC) were examined using simple linear regression. Simple regressions were also determined to show the intercorrelations between the independent variables (Steele et al., 1997).

The combined relationships among each biological endpoint and soil properties were further examined using multiple linear regression (MR) and path analysis (PA). For the purpose of comparison, two potential mechanistic models were considered (Figure 2). The first model includes pH, OC, and clay as independent variables; it was included because it is commonly used and these parameters are readily available from soil survey information. The second model considers clay as its two potentially separate effects: FEAL (amorphous Fe and Al oxide) and CEC; and so uses pH, OC, FEAL, and CEC as independent variables. It was hoped that this would provide further mechanistic insight and would conform better to the rule of unitary effects necessary for subsequent path analysis.

Multiple linear regression was used to examine the combined effects of soil properties on metal toxicity and bioavailability to biological endpoints. Empirical models that explain a maximum of the variation (R²) in the dependent variables due to the combined effects of soil properties were derived (Steele et. al., 1997). The multiple regression function has the format

$$Y = a + b_1 x_1 + b_2 x_2 + b_3 x_3 \dots b_p x_p$$
(6)

where, Y is the biological endpoint, *a* is the *y* intercept, and b_i is the partial slope for the *i*th independent variable (i = 1,....p).

Path analysis was used to partition correlations into direct and indirect effects. The diagram of the two path models considered here is shown in Figure 2. The example path diagram (Figure 1) shows that the direct effect (P_{ij}) of each independent variable on the dependent variable is denoted with a single-headed arrow and is the standard partial regression coefficient obtained from the MR. The simple correlations (r_{iq}) between the independent variables are denoted with double-headed arrows. The indirect effects are the product of the simple correlation coefficient and the partial regression coefficient ($r_{iq} \times P_{ij}$). The path total (r_{ij}) is the sum of the direct and indirect effects of each independent variable on the dependent variable.

The results of the path analysis are presented in a tabular form suggested by Williams et al. (1990). Results for each biological endpoint are presented as a matrix in a separate sub-table. Each sub-table contains the direct effect (italics) on the diagonal, the indirect effects off diagonal, and the path total. The significance levels for the direct effects (path coefficients) are derived from the Pvalues associated with the partial slopes from the multiple regressions. The significance levels for the path total are the correlation coefficients (r) from the simple correlations.

RESULTS

Soil Properties

Soil chemical and physical properties of the 21 selected soils are summarized in Table 3. The distribution in soil pH is shown in Figure 3. Soil pH for the control soils (Fig. 3A) ranged from 4.0 to 8.0 with a mean of 7.1. Metal

spiking generally reduced pH due to metal hydrolysis and ion exchange. The mean pH for spiked soils was 5.7 for Cd (Fig. 3B), 5.6 for Pb (Fig. 3C) and 5.8 for Zn (Fig. 3D). The distribution of soil properties is shown in Figure 4. Soil OC ranged from 0.50 to 3.0% with a mean of 1.46% (Fig. 4 A). The CEC ranged from 3.01 to 32.4 cmol_c kg⁻¹ with a mean of 15.6 cmol_c kg⁻¹ (Fig. 4B). The percent clay content of the selected soils ranged from 5.0 to 71.3%, with a mean of 32.2% (Fig. 4 D). The amorphous Al and Fe oxide (FEAL) content ranged from 0.009 to 0.195 mol kg⁻¹, with a mean of 0.056 mol kg⁻¹ (Fig. 4C). Because all of the soils were spiked with an equal amount of each metal, it is assumed that significant differences in bioavailability should be due to the modifying effects of soil properties rather than metal concentration.

Lettuce: Toxicity and Bioaccumulation

Control Soils

Lettuce bioassay endpoints for the control soils are summarized in Tables 4, 9, and 14. With the exception of Zn, all of the lettuce tissue metal (Cd, Pb) concentrations were below the detection limits of 0.5 mg kg⁻¹ (Table 4) for Cd and 1.25 mg kg⁻¹ (Table 9) for Pb. Lettuce tissue Zn concentrations, for the control soils (Table 14), ranged from 11.9 to 33.8 mg kg⁻¹, within the normal tissue concentration for most plants of 8 to 125 mg kg⁻¹ (Brady and Weil, 1996). Dry matter growth ranged from 3.59 to 8.27 g, with a mean of 5.91 g. Lettuce germination ranged from 50 to 91.7% (of 20 seeds planted) with a mean of

77.5% (Table 4). The distribution by soil for germination and DMG is shown in Figure 5.

Metal-spiked Soils

Lettuce bioassay biological endpoints, for lettuce grown in metal-spiked soils, are summarized in Tables 4, 9, and 14. Although sufficient macro, and micronutrients were supplied to all soils so that they would not be limiting, soil quality (i.e., acidity, texture) may affect dry matter yield and germination. To differentiate between soil quality effects and effects due to the metal contaminants, dry matter growth (DMG) and germination of lettuce grown on metal-spiked soils are presented here relative to the control. Relative dry matter growth (RDMG) is calculated as:

(DMG (g) in metal-spiked soil / DMG (g) in control soil) * 100 = RDMG (7)

Similarly relative germination (RG) is calculated as:

(Germ. in metal-spiked soil / Germ. in control soil) * 100 = RG (8)

Because the lettuce metal tissue concentrations for the control soils were so low, the tissue concentrations for the lettuce grown on metal-spiked soils are not reported relative to controls. If lettuce plants grown on the metal-spiked soils were deemed to be too unhealthy or necrotic, tissue concentrations were considered unreliable and are not included. Significance levels reported here are denoted as, *P < 0.05, **P < 0.01, and ***P < 0.001.

Cadmium

Relative germination in Cd-spiked (300 mg kg⁻¹) soils ranged from 71.1 to 133% with a mean of 91.7% of the germination in the control soils (Table 4). ANOVA showed no significant effect of metal on germination.

Relative dry matter growth (RDMG), significantly lower in Cd-spiked soils, ranged widely from 1.04 to 55.6% with a mean of 11.4% of lettuce grown in the control soils (Table 4). Cadmium contamination substantially decreased DMG in lettuce. The wide range in RDMG indicates that the toxicity of Cd was modified by soil properties.

Lettuce tissue Cd concentrations ranged from 57.7 to 403 mg Cd kg⁻¹, with a mean of 156 mg Cd kg⁻¹ (Table 4), significantly higher than lettuce grown in control soils. Again, the wide range in tissue concentrations indicates that the availability of Cd was modified by soil properties. The distribution by soil of lettuce bioassay endpoints grown in Cd- spiked (300 mg kg⁻¹) soil is shown in Figure 6.

To examine the modifying effects of soil properties on phytotoxicity and bioaccumulation due to Cd, the simple linear correlation coefficients (r) for the relationship between lettuce bioassay endpoints (studentized or studentized and then linearized) and soil properties (OC, clay, pH, FEAL, and CEC) were determined (Table 5).

For relative dry matter growth (RDMG), there was a significant (r = 0.476*) relationship with percent OC, and between log RDMG and percent clay (r = 0.544^*). There was a highly significant (r = 0.808^{***}) correlation between RDMG and FEAL, but no significant correlation between RDMG and pH or CEC. For lettuce tissue Cd, there was a significant negative relationship between log lettuce tissue Cd and percent clay ($r = -0.720^{**}$), pH ($r = -0.649^{**}$), and CEC (r = -0.702**). There was no significant correlation between tissue Cd and percent OC or FEAL. Inverse relationships between Cd bioaccumulation and soil pH were found. Increasing Cd solubility and availability in acidic soils (pH < 6) resulted in increased Cd bioavailability and phytotoxicity. Increased clay content decreased Cd bioavailability and phytotoxicity by adsorbing Cd from soil solution and decreasing its availability. Increased CEC with increased pH significantly reduced Cd bioaccumulation. Although many of the soil properties appear to be significantly affecting lettuce biological endpoints, the intercorrelation between the soil properties may be responsible for some of these ostensible effects.

To determine how the combined effects of soil properties may modify soil Cd phytotoxicity, two multiple linear regression models were used to examine the relationship between each lettuce bioassay endpoint (studentized or studentized and then linearized) and combined soil properties. Model I included pH, OC, and clay as independent variables. Model II included pH, OC, FEAL, and CEC as independent variables. Empirical models that explain a maximum of the variation (\mathbb{R}^2) in the dependent variables due to the combined effects of soil properties (Steele et. al., 1997) were generated (Table 6).

For RDMG, there was a significant relationship ($R^2 = 0.54^{**}$) for model I and ($R^2 = 0.660^{**}$) model II. There was also a significant relationship between log tissue Cd and soil properties model I for ($R^2 = 0.63^{**}$) and model II ($R^2 =$ 0.66^{**}) (Table 6). Though significant, the multiple regression equations may not be useful for predicting metal bioavailability or for making remediation decisions because they do not provide insight as to the relative contribution of each independent variable included in the models.

As mentioned previously, the intercorrelations between soil properties (Table 7) can complicate an investigators understanding of potential causal effects. Strong intercorrelations between independent variables can result in apparent correlations with the dependent variable that may not be well-founded. These intercorrelations affect the indirect effects of the soil properties included in the path models. Some of this ambiguity may be cleared up through this decomposition of correlations. For the 21 soils included in the study, pH was significantly correlated with clay (r = 0.55*) and CEC (r = 0.71***), but not with OC or FEAL. Organic carbon was significantly related with clay (r = 0.49*), FEAL (r = 0.61**), and CEC (r = 0.70***). Clay was also significantly related to FEAL (r = 0.61**) and highly related to CEC (0.90***). There was also a significant relationship (r = 0.54**) between FEAL and CEC. These intercorrelations between soil properties can obscure the relative contribution of each property in explaining the variation in the dependent variable.

The results of path analysis (PA) for RDMG (Table 8), for model I, showed a significant path total for OC ($r = 0.476^*$), which was primarily due to a
combination of the direct effect of OC (0.321) and the indirect effect of OC via clay (0.326). There was also a significant path total for clay ($r = 0.531^*$), which was entirely due to the direct effect (0.665^{**}) of clay. While the correlation between pH and RDMG for model I was very poor (r = -0.064), the PA shows that this can be attributed to the combination of a significant (-0.532^{*}) negative direct effect of pH opposed by a positive (0.364) indirect effect of pH via clay and to a lesser extent, via a positive (0.103) effect of OC. These strong effects merit consideration and, except for the partitioning of correlation through PA, would have been lost.

The usefulness in separating the effect of clay, in model II, into FEAL and CEC became apparent when considering the effects on RDMG. The highly significant (r = 0.808***) total correlation between RDMG and FEAL is shown to be almost entirely due to the direct effect of FEAL (0.775***). The significant (r = 0.470*) total correlation between RDMG and OC is due entirely to the indirect effect of OC (0.472) via its intercorrelation with FEAL. Even the strong effect (0.400) of CEC was via FEAL. The strong effect of FEAL compared to the very low effects of CEC demonstrates that the component of clay that is modifying the effect of Cd to RDMG is the amorphous oxide and not CEC. For model II, there are no strong direct or indirect effects of OC or pH.

For Cd tissue concentration, model I showed a significant path total (r = -0.648^{**}) for pH. This came from a strong negative direct effect of pH (-0.384) supported by a strong negative indirect effect of pH (-0.250) via its intercorrelation with clay. There was also a highly significant path total (r = -

 0.720^{**}) for the effects of clay. This included a strong direct effect of clay (-0.508*), supported by an indirect effect of clay via pH (-0.189). There were no significant direct or indirect effects of OC for tissue Cd for model I. Model II had a significant path total (r = -0.649*) for the effect of pH on tissue Cd, which was due entirely to the direct effect of pH (-0.647*). Although the path total for the effects of CEC on Cd tissue was significant (r = -0.701*), it was predominantly due to the indirect effect of CEC via pH (-0.439) and FEAL (-0.291). By comparison the direct effect of CEC (-0.104) was relatively low. There was also a strong direct effect (-0.538) for FEAL. Again, model II shows that the contribution of clay is through its oxide and not its CEC component.

For RDMG, model I showed pH, OC, and clay as all contributing. On closer inspection, model II showed that it was the amorphous oxide and not the CEC component of clay that was strongly affecting RDMG. This demonstrates the importance of the constraint of unitary effects of each predictor variable (Loehlin, 1987) for PA. The apparent contributions of OC and clay in model I may have been due to their high intercorrelations with FEAL.

For tissue uptake of Cd, model I showed a strong contribution by pH and clay. This is supported in model II which shows that it is the amorphous metal oxide content (FEAL) in these soils that is primarily responsible for modifying Cd bioavailability. In model II, pH also contributes significantly to modifying Cd bioaccumulation.

The large contribution of FEAL and pH to the variation in RDMG and tissue Cd uptake makes sense since oxides can bind Cd through specific

adsorption forming stable complexes (Mitchell et al., 1978; Bingham et al., 1980; Adriano, 2001; Sparks, 2003) and since increasing pH is associated with decreasing Cd solubility (Lagerwerff, 1971; Street et al., 1977; Mahler et al., 1978; Basta et al., 1993; Chlopecka et al., 1996; Adriano, 2001; Sparks, 2003).

Lead

The relative germination (RG) in Pb-spiked (2000 mg kg⁻¹) soils ranged from 76.9 to 165% with a mean of 111% relative to the control soils (Table 9).

Relative dry matter growth (RDMG) ranged widely from 2.5 to 88.5% with a mean of 33.3% of lettuce grown in the control soils (Table 9). This suggests that Pb contamination substantially decreased lettuce growth relative to the controls, and the wide range indicates that the bioavailability of Pb is being modified by soil properties.

Lettuce tissue Pb concentrations also ranged widely, from 3.22 to 233 mg Pb kg⁻¹, with a mean of 64.7 mg Pb kg⁻¹ (Table 9). Again, the wide range in Pb tissue concentrations suggests that the phytoavailability of Pb is being modified by soil properties. The distribution by soil of lettuce bioassay endpoints is shown in Figure 7. This shows the wide range in tissue Pb concentrations and graphically shows how increased tissue Pb concentration corresponded to a decrease in RDMG.

Simple linear correlation coefficients (r) for the relationship between the lettuce bioassay endpoints (studentized or studentized and then linearized) and

soil properties (OC, clay, pH, FEAL, and CEC) were determined (Table 10) to ascertain which soil properties may act as modifiers to soil Pb toxicity.

There was a significant relationship between log RDMG and percent OC (r = 0.438^{*}) and between RDMG and FEAL (r = 0.437^{*}). There was no significant correlation between RDMG and clay, pH, or CEC (Table 10).

Highly significant negative relationships were found between log tissue Pb and percent OC ($r = -0.728^{***}$), percent clay ($r = -0.611^{**}$), CEC ($r = -0.771^{***}$), and FEAL ($r = -0.524^{*}$). The relationship between tissue Pb and pH was not significant. Soil organic matter will absorb Pb through cation exchange reactions (Alloway, 1990) and complexation reactions between thiol (e.g. –SH) functional groups thereby decreasing Pb solubility and bioavailability (McBride, 1994). Clay adsorbs Pb by ion exchange and specific adsorption reactions on amorphous metal oxide (FEAL) surfaces, thereby decreasing Pb bioavailability (Table 10).

To determine if the combined effects of soil properties may modify soil Pb phytotoxicity, two multiple linear regression models were used to examine the relationship between each lettuce bioassay endpoint (studentized or studentized and then linearized) and the combined soil properties. Model I included OC, clay, and pH as independent variables. Model II included pH, OC, FEAL, and CEC as independent variables (Figure 2). Empirical models that explain a maximum of the variation (R^2) in the dependent variables due to the combined effects of soil properties (Steele et. al., 1997) were generated (Table 11).

No significant (P < 0.05) relationship was found between Pb RDMG and soil properties for either model (Table 11). The relationship between lettuce log

tissue Pb and soil properties was highly significant for both model I ($R^2 = 0.64^{***}$) and model II ($R^2 = 0.67^{***}$) (Table 11). Although significant, the regression equations may not be useful for predicting metal bioavailability or for making remediation decisions because they do not provide any insight as to the relative contribution of each independent variable included in the models toward explaining the variation in biological endpoints.

As mentioned previously, the intercorrelations between soil properties (Table 12) can complicate an investigator's understanding of potential causal effects. Strong intercorrelations between independent variables can result in apparent correlations with the dependent variable that may not be wellfounded. These intercorrelations affect the indirect effects of the soil properties included in the path models. It is through this decomposition of correlations that some of this ambiguity may be cleared up. For the 21 soils used in this study, there was a significant correlation between pH and clay (r = 0.53^*) and CEC (r = 0.69^{***}), while there was no significant relationship between pH and OC or FEAL. Organic carbon was significantly related to clay (r = 0.49^*) and CEC (r = 0.61^{**}), but not with FEAL. Clay was also significantly related to FEAL (r = 0.61^{**}).

The path analysis for square root (SR) RDMG in model I showed no significant path totals. There was, however, a significant positive direct effect of OC (0.528*), but when combined with the negative indirect of OC (-0.154) via pH the overall correlation was not significant. Similarly, the opposing negative indirect effect of clay via pH and positive indirect effect of clay via OC accounted

for the insignificant total correlation with clay. For model II, there was a significant path total ($r = 0.437^*$) for FEAL, which was due to the combined direct effect of FEAL (0.242) and the indirect effect of FEAL via OC (0.243). There was also a strong, although not significant direct effect of OC (0.399).

For tissue Pb, model I shows a strong significant path total for OC (r = -0.728***), which is primarily due to the direct effect of OC (-0.548*) with a small contribution indirectly (-0.120) via clay. Similarly, the significant ($r = -0.611^{**}$) path total for clay is primarily due to the combined direct effect of clay (-0.244) and the indirect effect of clay via OC (-0.269). The strong, although not significant, path total for pH is primarily due to its intercorrelation with OC and clay. In model II, the value of separating the effects of clay into FEAL and CEC was demonstrated in that it becomes apparent that the effect of clay is primarily due to the CEC component. This is supported by the strong path total for CEC (r = -0.773^{***}), which is primarily attributable to the direct effect of CEC (-0.440) in conjunction with the indirect effect of CEC via OC (-0.225). The apparent effects of CEC and OC are corroborated by the strong path total for OC (-0.690***). This correlation is primarily due to the combined direct effect of OC (-0.365) and the indirect effect of OC via CEC (-0.268). Even the significant path total for FEAL (r = -0.524^*) is due to the combined indirect effects of FEAL via OC (-0.222) and CEC (-0.242), while the direct effect of FEAL is negligible (-0.057). There is no evidence of a strong effect of pH for log tissue Pb for model II. Again, the major effects of pH are indirect via OC (-0.118) and CEC (-0.304).

For RDMG, PA shows that OC and FEAL are decreasing Pb toxicity to lettuce. Many studies have suggested that Pb can form stable complexes with OC (MacLean et al., 1969; Riffaldi et al., 1976; Soldatini et al., 1976; Zimdahl and Foster, 1976; Scialdone et al., 1980; Adriano, 2001; Sparks, 2003) and with metal oxide surfaces (Riffaldi et al., 1976; Soldatini et al., 1976; Cholpecka et al., 1996; Adriano, 2001; Sparks, 2003).

The PA clearly shows that it is the CEC component of clay that is modifying the effects of Pb on tissue uptake. At the high spike concentration used in this study (2000 mg Pb kg⁻¹), it may be that more stable complexation sites became filled and excess Pb moved to CEC sites. Organic carbon also contributed significantly to modifying the effects of Pb. Many studies have suggested that soil organic matter will adsorb Pb (MacLean et al., 1969; Riffaldi et al., 1976; Soldatini et al., 1976; Zimdahl and Foster, 1976; Scialdone et al., 1980; Adriano, 2001; Sparks, 2003), forming stable complexes. Lead bioavailability has also been shown to be reduced by increased CEC (Riffaldi et al., 1976; Zimdahl and Skogerboe, 1977; Basta and Tabatabai, 1992a; Adriano, 2001; Sparks, 2003)

Zinc

The relative germination (RG) in Zn-spiked (300 mg kg⁻¹) soils ranged from 87.2 to 190% with a mean of 110% of the germination in the control soils (Table 14), suggesting that Zn spiking did not inhibit germination.

Zinc contamination substantially decreased lettuce growth. Relative dry matter growth (RDMG) ranged widely from 3.4 to 85.4% with a mean of 47.4% of the DMG found in lettuce grown on the control soils (Table 14). The wide range in RDMG shows that the phytotoxicity of Zn is being modified by soil properties.

Lettuce tissue Zn concentrations also ranged widely from 18.4 to 2038 mg Zn kg⁻¹, with a mean of 332 mg Zn kg⁻¹ (Table 14), also showing that the bioavailability of Zn was modified by soil properties. The distribution, by soil, of lettuce bioassay endpoints is shown in Figure 8. This figure shows the wide range in tissue Zn concentrations.

Simple linear correlation coefficients (r) for the relationships between lettuce bioassay endpoints (studentized or studentized and then linearized) and soil properties (OC, clay, pH, FEAL, and CEC) were determined (Table 15) to find out which soil properties may modify soil Zn toxicity and bioavailability.

There were significant negative relationships between RDMG and percent clay ($r = -0.474^*$) and between RDMG and pH ($r = -0.546^{**}$) and CEC (-0.532*) (Table 15). There was no significant correlation between RDMG and percent OC or FEAL. Unlike Pb, Zn and Cd generally have a lower affinity for soil organic matter and metal oxide complexation (Sparks, 2003).

Highly significant negative relationships were found between log tissue Zn and percent clay (r = -0.793^{***}), pH (r = -0.887^{***}), CEC (r = -0.86^{***}). The relationship between tissue Zn and percent OC and FEAL was not significant (P < 0.05) (Table 15).

To determine if the combined effects of soil properties may modify soil Zn phytotoxicity, two multiple linear regression models were used to examine the relationship between each lettuce bioassay endpoint (studentized or studentized and then linearized) and soil properties. Model I included pH, OC, and clay as independent variables. Model II included pH, OC, FEAL, and CEC as independent variables. Empirical models that explain a maximum of the variation (\mathbb{R}^2) in the dependent variables due to the combined effects of soil properties (Steele et al., 1997) were generated (Table 16).

There was no significant relationship (P < 0.05) between RDMG and the combined soil properties for either model. The relationship between lettuce log tissue Zn and soil properties was highly significant for both models I and II ($R^2 = 0.91^{***}$) (Table 16). Although very significant, the regression equations may not be useful for predicting metal bioavailability or for making remediation decisions because they do not provide any insight as to the relative contribution of each independent variable included in the models.

As mentioned previously, the intercorrelations between soil properties (Table 17) can complicate an investigators understanding of potential causal effects. Strong intercorrelations between independent variables can result in apparent correlations with the dependent variable that may not be well founded. These intercorrelations affect the indirect effects of the soil properties included in the path models. It is through this decomposition of correlations that some of this ambiguity may be cleared up.

For the 21 soils in this study, pH was significantly correlated with OC (r = 0.45^*), and clay (r = 0.59^{**}) and strongly correlated with CEC (r = 0.75^{***}). Soil pH was not significantly correlated with FEAL. Organic carbon was significantly correlated with clay (r = 0.49^*), and FEAL (r = 0.61^{**}) and strongly correlated with CEC (r = 0.70^{***}). Clay was also significantly related to FEAL (r = 0.61^{**}) and very strongly correlated with CEC (r = 0.90^{***}). FEAL was also significantly related to CEC (r = 0.55^{**}).

The results of the path analysis for RDMG showed a significant effect of pH for both models. In model I, the direct effect of pH (-0.433) in conjunction with the indirect effect of pH via clay (-0.159) accounts for the majority of the significant (r = -0.546**) total effect of pH. Similarly, the direct effect of clay (-0.269) supported by the indirect effect of clay via pH (-0.255) accounts for the majority of the significant effect of clay ($r = -0.474^*$). There was little contribution from OC directly or indirectly to RDMG in model I. It appears that only pH and clay are contributed substantially to explaining the variation in RDMG in model I. The results of the PA for RDMG for model II demonstrate the usefulness of splitting the effects of clay into FEAL and CEC. In this model, the significant total effect of pH (r = -0.546**) was due entirely to the indirect effect of pH via its intercorrelation with CEC (-0.599). The negative effect of CEC to RDMG is again demonstrated by the direct effect of CEC (-0.793), which accounts for greater than the CEC path total ($r = -0.532^*$) because it is confounded by a positive indirect effect (0.218) via FEAL. Although not significant, the path total for FEAL shows the opposition of a strong positive direct effect of FEAL (0.396) by the

negative indirect effect of FEAL via CEC (-0.436), demonstrating that two strong but opposing effects can lead to an overall low correlation (r = 0.020). While there may be some evidence that FEAL may be contributing to the explanation of the variance in RDMG in model II, CEC stands out most strongly and pH and OC do not contribute directly or indirectly to any extent in model II. The apparent contribution of pH and clay in model I may be primarily due to the strong intercorrelation between pH ($r = 0.75^{***}$) and clay ($r = 0.90^{***}$) with CEC. This would not have been apparent if we had compared f the two models, demonstrating the importance of the constraint of unitary effects of each predictor variable (Loehlin, 1987). It is difficult to explain why the effects of pH and clay are negative for RDMG.

This same concurrence of effects for pH, clay, and CEC repeats for the bioavailability of Zn to lettuce tissue. In model I, we see a highly significant (r = -0.887^{***}) total correlation with pH, because the significant direct effect (r = -0.671^{***}) of pH supported by an indirect effect of pH (-0.266) via clay. Similarly, there is a highly significant (r = -0.792^{***}) correlation with clay because to the significant direct effect (r = -0.452^{***}) of clay supported by the indirect effect (-0.395) of clay via pH. There is no significant direct or indirect contribution by OC. In model II, a highly significant (r = -0.887^{***}) correlation with pH is shown to be the result of the significant direct effect (r = -0.518^{**}) of pH supported by a strong indirect effect (-0.503) of pH via CEC. Similarly, there is a highly significant (r = -0.861^{***}) correlation with CEC due to a significant direct effect (r = -0.665^{***}) of CEC supported by an indirect effect (-0.391) of CEC via pH. In

model II, the effect of OC on bioaccumulation of Zn in lettuce tissue was significant (r = 0.303*), but this positive effect was confounded by a strong negative indirect effect (-0.406) of OC via CEC and a negative indirect effect (-0.232) of OC via pH. Consequently, the total correlation between OC and Zn bioaccumulation is not significant. For model I, both pH and clay contributes to explaining the variation in tissue Zn, while the direct or indirect effects of OC contributed little. On closer inspection, model II showed that it is the CEC component of clay that is primarily accounting for variation in tissue Zn. The soil CEC and pH account for most of the reduction in Zn uptake in lettuce. Increasing pH decreases Zn solubility and bioavailability (Bingham et al., 1964; Lagerwerff, 1971; Mitchell et al., 1978; Cavallaro and McBride, 1984; Adriano, 2001; Sparks, 2003). Zinc sorbed to CEC sites is also less bioavailable (Elgabaly, 1950; Cavallro and McBride, 1984; Basta et al., 1993; Adriano, 2001; Sparks, 2003).

DISCUSSION

Cadmium

Soil properties significantly modified the bioaccumulation of Cd in lettuce. Results from statistical analyses are summarized in Table 19. ANOVA analysis showed Cd significantly affected Cd phytotoxicity and bioaccumulation. Simple correlation analysis showed bioaccumulation was highly correlated with pH, clay, and CEC. Multiple regression equations were significant (P < 0.01) for both model I and model II, but provided no information as to the relative contribution of each parameter included. Path analysis was used to partition simple correlations

and provide further information. The path analysis for model I (pH, OC, clay) showed a significant negative effect of clay on Cd bioaccumulation. The PA for model II (pH, OC, FEAL, CEC) showed the modifying effects of pH and FEAL were responsible for Cd bioaccumulation. Comparison of PA models I and II suggest the significant clay direct effect for Model I was due to the FEAL (amorphous oxide component) and not CEC component of clay. Other properties (e.g., OC, CEC) did not affect Cd bioaccumulation.

Many studies have shown that the solubility and therefore availability of Cd is reduced as pH increases (Lagerwerff, 1971; Street et al., 1977; Mahler et al., 1978; Basta et al., 1993; Chlopecka et al., 1996; Adriano, 2001; Sparks, 2003) and that Cd can form stable complexes on oxide surfaces thereby reducing bioavailability (Mitchell et al., 1978; Bingham et al., 1980; Adriano, 2001; Sparks, 2003). Results from our work are consistent with these findings.

Soil properties significantly modified the toxicity of Cd to lettuce in this study as measured by RDMG. At the same Cd spike concentration of 300 mg Cd kg⁻¹, lettuce RDMG ranged from 1.04 to 55.6%, a 53.4-fold range in lettuce response. Results from statistical analyses are summarized in Table 19. ANOVA analysis showed Cd significantly affected lettuce RDMG. Simple correlation analysis showed RDMG was correlated with FEAL (P < 0.01) and OC and clay (P < 0.05). Multiple regression equations were significant (P < 0.01) for both model I and model II, but provided no information as to the relative contribution of each parameter included. Path analysis was used to partition simple correlations and provide further information. A highly significant direct

effect of clay on RDMG was found. The path analysis for model I (pH, OC, clay) showed a significant negative effect of pH confounded by positive direct effects of clay. The PA for model II (pH, OC, FEAL, CEC) showed a highly significant direct effect of FEAL on RDMG. Model I showed the importance of clay. The path analysis for model II showed FEAL was the most important factor. Comparison of PA models I and II suggests the significant clay direct effect in PA model I was due to the FEAL (amorphous oxide component) and not CEC component of clay. Other properties (e.g., OC, CEC) did not affect RDMG.

Results from our study show FEAL reduced Cd bioavailability and bioaccumulation and therefore reduced phytotoxicity (e.g., increased lettuce RDMG). Our results show pH was negatively correlated with bioaccumulation of Cd. Decreased tissue Cd should also reduce Cd phytotoxicity. One would expect a positive effect of soil pH on RDMG. Our negative effect of pH on RDMG is difficult to explain. However, the effect of FEAL on RDMG was much greater than soil pH. Our results suggest FEAL is the most important modifying property on phytotoxicity (e.g., RDMG).

Lead

The range of Pb bioaccumulation of 3.22 to 233 mg Pb kg⁻¹ represents a remarkable 72-fold range in lettuce response. ANOVA analysis showed Pb significantly affected phytotoxicity and bioaccumulation (Table 20). Simple correlation analysis shows bioaccumulation was highly correlated with pH, clay, FEAL, and CEC. Only pH was not significantly correlated to tissue Pb. Multiple

regression equations were highly significant (P < 0.001) for both model I and model II, but provide no information as to the relative contribution of each parameter included. The path analysis for model I showed OC and clay as contributing to most of the reduction in Pb uptake. This result was further clarified in model II where it was shown that the effects were due to OC and CEC. This indicates that it is the CEC portion of clay and not the oxide that is responsible for modifying uptake of tissue Pb. The PA for model II shows that pH and FEAL did not contribute to explaining the variation in tissue Pb. Many studies have shown that Pb will form stable complexes with OC (Riffaldi et al., 1976; Soldatini et al., 1976; Zimdahl and Foster, 1976; Alloway, 1990; McBride, 1994; Adriano, 2001; Sparks, 2003) and that Pb bound to cation exchange sites (Riffaldi et al., 1976; Zimdahl and Skogerboe, 1977; Basta and Tabatabai, 1992b; Adriano; 2001; Sparks, 2003) is less bioavailable.

Soil properties significantly modified the toxicity of Pb to lettuce in this study. At the same Pb spike concentration of 2000 mg Pb kg⁻¹, lettuce RDMG ranged from 2.5 to 88.5% of lettuce grown in control soils. This represents a 35-fold range in lettuce response. ANOVA analysis showed Pb significantly affected RDMG (Table 20). Simple correlation analysis shows RDMG was correlated with OC and FEAL. Soil pH was not significantly correlated to RDMG. Neither multiple regression model I or II was significant (P < 0.05). The path analysis for model I showed a direct effect of OC. The path analysis for model II showed the combined effects of OC and FEAL affected RDMG. The positive relationship of RDMG with FEAL and OC is not surprising. Lead will bind to specific adsorption

sites on metal oxides (Riffaldi et al., 1976; Soldatini et al., 1976; Chlopecka et al., 1996; Adriano, 2001; Sparks, 2003) and ligand functional groups on OC (MacLean et al., 1969; Riffaldi, et al., 1976; Soldatini et al., 1976; Zimdahl and Foster, 1976; Scialdone et al., 1980; Adriano, 2001; Sparks, 2003) forming stable complexes.

Zinc

The range of Zn uptake by lettuce was from 18.4 to 2038 mg Zn kg⁻¹, representing a 243-fold range in lettuce response to Zn. This vividly illustrates that soil properties had a profound modifying effect on Zn bioaccumulation. ANOVA analysis showed Zn significantly affected bioaccumulation (Table 21). Simple correlation analysis showed bioaccumulation was highly negatively correlated with pH, clay, and CEC, but not FEAL and OC. Multiple regression equations were significant (P < 0.01) for both model I and model II, but provided no information as to the relative contribution of each parameter included. Path analysis was used to partition simple correlations and provide further information. The path analysis for model I (pH, OC, clay) showed a significant negative effect of clay and pH on Zn bioaccumulation. The PA for model II (pH, OC, FEAL, CEC) shows the modifying effects of pH and CEC were responsible for reductions in Zn bioaccumulation. Comparison of PA models I and II suggest that the effect of clay was entirely due to its CEC component.

Over all, both models I and II showed that increased CEC and pH were responsible for decreasing Zn uptake in lettuce. Many studies have shown that

Zn solubility decreases with increased pH (Bingham et al., 1964; Lagerwerff, 1971; Mitchell et al., 1978; Cavallaro and McBride, 1984; Adriano, 2001; Sparks, 2003) and that Zn sorption on cation exchange sites will reduce the bioavailability and toxicity of Zn (Elgabaly, 1950; Cavallaro and McBride, 1984; Basta et al., 1993; Adriano, 2001; Sparks, 2003).

Soil properties substantially modified Zn toxicity to lettuce. At the same Zn spike level of 300 mg Zn kg⁻¹, lettuce dry matter growth in Zn-spiked soils ranged from 3.4 to 85.4% of lettuce grown in control soils. This represents a 25-fold range in response to Zn. ANOVA analysis showed Zn significantly affected RDMG. Simple correlation analysis shows RDMG was negatively correlated with pH, clay, and CEC, but not FEAL and OC. Neither multiple regression model I or II was significant (P < 0.05). For model I, the path analysis showed strong negative effects of pH and clay with no contribution of OC. Path analysis was used to partition simple correlations between soil properties and RDMG. Path analysis did not discover any significant direct effects between soil properties and RDMG.

CONCLUSIONS

Metal spiking affected metal bioaccumulation and phytotoxicity, but not relative germination in this study. Several soil properties were correlated with metal bioaccumulation and phytotoxicity. The relative contribution (e.g., importance) of specific soil properties to modify the phytotoxicity and bioaccumulation of Cd, Pb, and Zn was metal dependent.

Increased pH and FEAL decreased Cd bioavailability and bioaccumulation. The most important soil property to increase RDMG in Cd-contaminated soil was FEAL. Iron and aluminum oxide clay strongly adsorbed Cd, decreasing its bioavailability and phytotoxicity.

Organic C and CEC but not soil pH decreased Pb bioavailability and bioaccumulation. The most important soil property to increase RDMG in Pb-contaminated soil was OC and FEAL. Soil organic matter (e.g., OC) formed strong complexes with Pb, decreasing its solubility, bioavailability, and phytotoxicity.

Soil pH and CEC decreased Zn bioavailability and bioaccumulation. Soil pH and CEC decreased the solubility of Zn. Zinc additions to soil reduced lettuce RDMG. The effect of soil properties for modifying Zn toxicity for RDMG were inconclusive.

Path analysis is a useful statistical tool to augment regression analysis. Although path analysis cannot prove a causal relationship, it can refute one. The detailed examination provided by path analysis provides insight into underlying chemical mechanisms affecting contaminant solubility, bioavailability, and ecotoxicity. Path analysis is useful for ecological studies involving soils with a wide range of physiochemical properties and can assist in site ERA and remediation decisions. Further studies on the use of path analysis in soil chemical/ecotoxicity studies are needed.

To adequately protect or restore soil ecosystems, it is necessary to accurately characterize soils suspected or presumed to be contaminated with

heavy metal by defining levels of metal in these soils that constitute a hazard to soil organisms.

Soil chemical properties greatly affected contaminant solubility, bioavailability, and toxicity to ecological receptors (e.g., lettuce). Soil physico/chemical parameters [e.g. pH, organic carbon (OC), % clay, amorphous AI & Fe oxides (FEAL) and cation exchange capacity (CEC)] should be measured for site-specific adjustments in ecological risk assessments (ERA). Specifically, soil properties should be used to modify metal (Cd, Pb, and Zn) bioavailability and ecotoxicity in site ERA. Results from this research study may provide baseline chemical and biological data that can be used in the development of Eco-SSLs or in reducing the uncertainty associated with determining site-specific ecological risk.

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Soil pH	<u> </u>	% Organic Ma	tter
-	< 2	2 to 6	6 to 10
4 < Soil pH • 5.5	Very High	High	Medium
5.5 < Soil pH < 7	High	Medium	Low
7 • Soil pH • 8.5	Medium	Low	Very Low
·			

Table 1. Proposed qualitative characterization of potential bioavailability from Eco-SSL guidelines (U.S. EPA, 2000).

			Backo	round Metal	Levels
Soil Taxonomic Classification	Soil Series H	lorizon	Cd	Pb	Zn
				mg kg ⁻¹ -	
Fine-loamy, mixed, superactive, calcareous, mesic Typic Endoaquoll	Canisteo	А	< 0.50	6.61	55.4
Fine mixed thermic Aquic Argiudoll	Dennis	А	< 0.50	7.39	43.6
Tale, mixed, thermic Aquic Argiduoli	Dennis	В	< 0.50	13.7	56.8
Loamy, mixed, active, thermic Arenic Haplustalf	Dougherty	А	< 0.50	7.69	24.5
Coarse-loamy, mixed, superactive, mesic Cumulic Hapludoll	Hanlon	Α	< 0.50	6.08	47.6
Fine, mixed, superactive, thermic Udertic Paleustoll	Kirkland	А	< 0.50	9.07	41.1
Fine, smectitic, mesic Typic Endoaquert	Luton	А	< 0.50	12.3	150
Fina-loamy mixed superactive thermic Aridic Calciustell	Mancie	Α	< 0.50	4.41	40.1
Fine-idanty, mixed, superactive, thermic Andic Calciuston	IVIALISIC	В	< 0.50	< 2.50	34.8
Fine-silty, mixed, active, thermic Udic Paleustoll	Norge	А	< 0.50	11.6	38.7
Fine smeetitic thermic Typic Enjagert	Osago	A	< 0.50	14.4	145
File, Sinectilic, mennic, Typic Epiager	Usaye	В	< 0.50	14.3	134
Fina silty mixed superactive thermic Dechie Argustell	Pond Crook	А	< 0.50	10.0	48.0
Fine-sity, mixed, superactive, thermic Facilic Arguston	FUIL CIEEK	В	< 0.50	8.74	46.2
Sandy mixed masia Lamellia Hanlystolf	Brott	А	< 0.50	2.88	28.2
Sandy, mixeu, mesic Lamellic Haplustall	Plan	В	< 0.50	2.53	14.9
Fine, smectitic, mesic Aridic Argiustoll	Richfield	В	< 0.50	12.6	64.3
Fina amastitia tharmia Overauja Vartia Arajudall	Cummit	A	< 0.50	12.7	56.9
Fine, smeculic, inermic Oxyaquic Venic Argiudoli	Summu	В	< 0.50	7.60	58.1
Fine, mixed, thermic Mollic Albaqualfs	Taloka	А	< 0.50	7.34	26.5
Fine-loamy, mixed, active, thermic Udic Argiustoll	Teller	А	< 0.50	11.4	26.1
				Range	
		Min.	< 0.50	< 2.50	14.9
		Max.		14.4	150
			-	Typical range	e ^a
		Min.	0.08	2.00	10.0
		Max.	0.94	200	300

Table 2. Taxonomic classifications and background metal levels of selected soils and typical range of metal occurring in uncontaminated soil.

^a Typical range of metal occurring in uncontaminated soil (Adriano, 2001).

Soil	pН	pH-	spiked So	oil ^a	OCp	CEC ^c	Clay	FEAL ^d
	Control	Cd	Pb	Zn	%	cmol _c kg ⁻¹	%	mol kg ⁻¹
1	7.5	7.6	7.6	7.7	3.00	30.5	38.8	0.057
2	5.6	4.8	4.8	4.7	1.90	9.77	23.8	0.083
3	6.1	5.5	5.2	5.3	0.80	14.6	45.0	0.066
4	5.3	4.7	4.5	4.6	1.20	3.33	11.3	0.016
5	7.4	6.6	6.7	6.4	1.60	16.3	17.5	0.044
6	5.6	4.7	4.8	4.8	1.45	14.0	31.3	0.061
7	7.1	6.8	6.6	6.8	2.00	32.4	71.3	0.069
8	7.8	7.6	7.7	7.7	1.50	16.5	30.0	0.026
9	8.0	7.6	7.8	7.2	0.65	11.7	35.0	0.011
10	4.0	3.9	3.8	3.9	1.20	4.57	17.5	0.045
11	6.6	6.1	5.9	5.8	2.60	28.3	55.7	0.128
12	6.8	5.8	5.9	6.2	2.00	27.5	61.3	0.195
13	5.2	4.4	4.1	5.8	1.90	10.7	28.8	0.058
14	6.0	5.7	5.2	5.4	0.80	12.5	32.5	0.049
15	6.5	5.2	4.6	5.3	0.90	4.40	5.00	0.010
16	6.4	5.2	5.2	4.4	0.50	3.40	6.25	0.009
17	7.7	6.8	6.4	6.9	1.10	22.4	41.3	0.033
18	7.2	7.1	7.0	7.3	2.40	29.4	45.7	0.089
19	7.0	6.6	6.5	6.4	1.25	27.6	56.8	0.036
20	5.1	4.5	4.2	4.3	1.20	4.85	11.3	0.032
21	4.5	3.9	4.3	4.1	0.85	3.01	10.0	0.011
Mean	7.1	5.7	5.6	5.8	1.46	15.6	32.2	0.056
Min.	4.0	3.9	3.8	3.9	0.50	3.01	5.00	0.009
Max.	8.0	7.6	7.8	7.7	3.00	32.4	71.3	0.195

Table 3. Control soil pH and pH of metal-spiked soils and soil chemical and physical properties related to metal bioavailability.

^a Metal spike levels: Cd 300 mg kg⁻¹, Pb 2000 mg kg⁻¹, and Zn 300 mg kg⁻¹. ^b Soil organic carbon. ^cCation exchange capacity. ^d Amorphous Fe and Al oxide.

	(Control Soil		····	Cd-spiked So	
Soil	Tissue	DMG ^a	G⁵	Tissue	RDMG ^c	RG ^d
	mg kg ⁻¹	g	%	mg kg ⁻¹	%	%
1	< 0.50	7.00	78.4	124	1.99	85.1
2	< 0.50	6.87	72.5	242	17.5	80.5
3	< 0.50	6.43	85.0	122	1.76	86.3
4	< 0.50	4.39	77.5	Na	1.66	96.8
5	< 0.50	8.05	83.4	110	1.30	75.0
6	< 0.50	4.86	71.7	Na	29.2	95.4
7	< 0.50	5.07	76.7	57.7	22.1	120
8	< 0.50	4.41	80.0	68.0	4.54	100
9	< 0.50	3.59	86.7	74.0	7.06	71.1
10	< 0.50	5.58	62.5	237	2.44	125
11	< 0.50	6.99	76.7	64.2	48.8	84.8
12	< 0.50	5.40	91.7	67.7	55.6	74.6
13	< 0.50	8.01	71.7	237	27.8	90
14	< 0.50	6.70	50.0	Na	1.04	79.1
15	< 0.50	6.02	81.7	403	1.35	73.5
16	< 0.50	5.38	86.7	215	1.86	76.9
17	< 0.50	5.21	81.7	251	3.78	93.9
18	< 0.50	8.27	86.7	68.9	1.37	71.1
19	< 0.50	4.35	88.4	116	4.53	88.7
20	< 0.50	6.10	55.0	Na	1.74	124
21	< 0.50	5.45	83.4	192	2.39	133
					·	
Mean	< 0.50	5.91	77.5	156	11.4	91.7
Min.	< 0.50	3.59	50.0	57.7	1.04	71.1
Max.	< 0.50	8.27	91.7	403	55.6	133

Table 4. Bioassay endpoints for lettuce grown in control soils and Cd-spiked (300 mg kg⁻¹) soils.

^a Lettuce dry matter growth. ^b Percent germination of 20 seeds. ^c Relative dry matter growth. ^d Relative germination.

	Simpl	le Linear Corre	lation C	oefficients (r)
	Ť	ïssue Cd		RDMG ^a
Soil Parameter	T^{c}	r	Т	r
Organic carbon	log	- 0.410	R	0.476 *
Clay	log	- 0.720 **	log	0.544 *

log

log

- 0.464

- 0.649 **

R

R

R

0.808 ***

- 0.065

0.399

Table 5. Simple linear correlation coefficients (r) for lettuce biological endpoints vs soil properties for soils spiked with Cd 300 mg kg⁻¹.

pН

Amorphous AI and Fe oxide

Cation exchange capacitylog- 0.702 **Fa Relative dry matter growth.b Best fit data set transformation: studentized (R), log10 (log).

	T ^a	Multiple Linear Regression	R^2
		Model I	
Tissue Cd	log	- 0.38(pH) – 0.048(OC) – 0.51(Clay)	0.63 **
RDMG ^b	R	- 0.53(pH) + 0.32(OC) + 0.67(Clay	0.54 **
		Model II	
Tissue Cd	log	- 0.65(pH) + 0.19(OC) – 0.54(FEAL) – 0.10(CEC)	0.66 **
RDMG	Ř	- 0.13(pH) – 0.001(OC) + 0.77(FEAL) + 0.07(CEC)	0.66 **
^a Best fit data	set tran	sformation: studentized (R) log ₁₀ (log)	

Table 6. Comparison of multiple regression equations and R² for lettuce biological endpoints for two potential mechanistic models for soils spiked with Cd at 300 mg kg⁻¹.

on: studentized (R), log₁₀ (log). ^b Relative dry matter growth.

	Simple Correlation Coefficients (r)									
	рĤ	OCa	Clay	FEAL ^b	CEC ^c					
рН	1.00	0.32	0.55 *	0.02	0.71 ***					
00		1.00	0.49 *	0.61 **	0.70 ***					
Clay			1.00	0.61 **	0.90 ***					
FEÁL				1.00	0.54 **					
CEC					1.00					

Table 7. Intercorrelations between soil properties for soils spiked with Cd at 300 mg kg⁻¹.

^a Organic carbon.
 ^b Amorphous Al and Fe oxide.
 ^c Cation exchange capacity.

	· · · ·	Мс	odel I		
	pН	OC ^a	Clay	Total	
Log ^b Tissue					
рН	- 0.384	- 0.014	- 0.250	- 0.648 **	
ÖC	- 0.115	- 0.048	- 0.248	- 0.411	
Clay	- 0.189	- 0.023	- 0.508 *	- 0.720 **	
R RDMG					
pН	- 0.532 *	0.103	0.364	- 0.065	
OC	- 0.171	0.321	0.326	0.476 *	
Clay	- 0.291	0.157	0.665 **	0.531 *	
		Мо	del II		

Table 8. Results of path analysis for soils spiked with Cd 300 mg kg⁻¹. Direct effects (*italics*, on the diagonal), indirect effects (off diagonal), and path totals.

		IVIC	aerii		
	pН	OC	FEAL ^c	CEC ^d	Total
Log Tissue					······································
рН	- 0.647 *	0.056	0.013	- 0.071	- 0.649 *
OC	- 0.193	0.188	- 0.331	- 0.064	- 0.400
FEAL	0.015	0.116	- 0.538	- 0.056	- 0.463
CEC	- 0.439	0.133	- 0.291	- 0.104	- 0.701 *
R RDMG					
рН	- 0.130	- 0.001	0.017	0.048	- 0.066
ÖC	- 0.042	- 0.001	0.472	0.041	0.470 *
FEAL	- 0.003	- 0.001	0.775 **	0.037	0.808 ***
CEC	- 0.092	- 0.001	0.426	0.067	0.400

^a Organic carbon.
 ^b Best fit data set transformations: studentized (R), log₁₀ (log), square root (SR).
 ^c Amorphous Al and Fe oxide.
 ^d Cation exchange capacity.

	C	ontrol Soil	P	b-spiked So	 oil	
Soil	Tissue	DMG ^a	G⁵	Tissue	RDMG [℃]	RG₫
	mg kg ⁻¹	g	%	mg kg ⁻¹	%	%
1	< 1.25	7.00	78.4	8.72	47.4	95.7
2	< 1.25	6.87	72.5	88.7	18.3	124
3	< 1.25	6.43	85.0	160	2.50	98.1
4	< 1.25	4.39	77.5	114	26.9	123
5	< 1.25	8.05	83.4	9.20	39.5	96.0
6	< 1.25	4.86	71.7	50.4	88.5	112
7	< 1.25	5.07	76.7	16.5	22.5	109
8	< 1.25	4.41	80.0	60.0	24.3	83.3
9	< 1.25	3.59	86.7	59.5	3.60	100
10	< 1.25	5.58	62.5	61.3	49.3	133
11	< 1.25	6.99	76.7	3.22	44.3	117
12	< 1.25	5.40	91.7	15.3	75.9	96.4
13	< 1.25	8.01	71.7	43.1	80.4	102
14	< 1.25	6.70	50.0	95.7	14.3	116
15	< 1.25	6.02	81.7	233	3.70	102
16	< 1.25	5.38	86.7	112	3.20	107
17	< 1.25	5.21	81.7	37.8	18.1	102
18	< 1.25	8.27	86.7	30.3	3.50	76.9
19	< 1.25	4.35	88.4	13.4	37.5	102
20	< 1.25	6.10	55.0	37.7	65.7	164
21	< 1.25	5.45	83.4	108	30.5	165
Mean		5.91	77.5	64.7	33.3	111
Min.		3.59	50.0	3.22	2.50	76.9
Max.		8.27	91.7	233	88.5	165

Table 9. Bioassay endpoints for lettuce grown in control soils and Pb-spiked 2000 mg kg⁻¹ soils.

^a Lettuce dry matter growth. ^b Percent germination of 20 seeds. ^c Relative dry matter growth. ^d Relative germination.

Table 10. Simple linear correlation coefficients for lettuce biological endpoints vs soil parameters for soils spiked with Pb 2000 mg kg⁻¹.

Soil Parameter	Simp Ti	Simple Linear Correlation Coefficients Tissue Pb RDMG ^a				
	T ^b	r	Т	r		
Organic carbon	log	- 0.728 ***	log	0.438 *		
Clay	log	- 0.611 **	R	0.09		
Amorphous AI and Fe oxide	log	- 0.524 *	R	0.437 *		
рH	log	- 0.491	R	- 0.289		
Cation exchange capacity	log	- 0.771 ***	R	0.09		

^a Relative dry matter growth.
 ^b Best fit data set transformations: studentized (R), log10 (log).

Table 11.	Comparison	of multiple re	gression eq	uations an	id R ² for	lettuce	biological	endpoints for
two poten	tial mechanis	tic models for	soils spiked	d with Pb a	it 2000 r	ng kg ⁻¹ .	4	

	T ^a	Multiple Linear Regression Equation	R ²
		Model I	
Tissue Pb	log	- 0.18(pH) – 0.55(OC) – 0.24(Clay)	0.64 ***
RDMG⁵	SŘ	- 0.48(pH + 0.53(OC) + 0.10(Clay)	0.35
		Modei II	
Tissue Pb	log	- 0.07(pH) – 0.36(OC) – 0.06(FEAL) – 0.44(CEC)	0.67 ***
RDMG	R	- 0.39(pH) + 0.40(OC) + 0.24(FEAL) – 0.06(CEC)	0.36
^a Best fit data ^b Relative dry	set tra matter	nsformation: studentized (R), log10 (log), square root (SR) [,] growth.	

·	Simple Correlation Coefficients (r)							
	рН	OCa	Clay	FEAL ^b	CEC°			
pН	1.00	0.32	0.53 *	0.04	0.69 ***			
ÖC		1.00	0.49 *	0.61 **	0.70 ***			
Clay			1.00	0.61 **	0.90 ***			
FEAL				1.00	0.55 **			
CEC		•			1.00			

Table 12. Intercorrelations between soil properties for soils spiked with Pb at 2000 mg $\mbox{kg}^{-1}.$

^a Organic carbon.
 ^b Amorphous Fe and Al oxide.
 ^c Cation exchange capacity.

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Model I								
	pН	OCª	Clay	Total				
Log ^b Tissue			······································		······			
рН	- 0.185	- 0.177	- 0.129	- 0.491				
ÖC	- 0.060	- 0.548 **	- 0.120	- 0.728 ***				
Clay	- 0.098	- 0.269	- 0.244	- 0.611 **				
SR RDMG								
pН	- 0.476	0.171	0.052	- 0.253				
ÖC	- 0.154	0.528 *	0.048	0.422				
Clay	- 0.251	0.259	0.098	0.106				
Model II								
	pH	00	FEAL ^c	CECd	Total			
Log Tissue								
рН	- 0.066	- 0.118	- 0.003	- 0.304	- 0.491			
00	- 0.022	- 0.365	- 0.035	- 0.268	- 0.690 **			
FEAL	- 0.003	- 0.222	- 0.057	- 0.242	- 0.524 *			
CEC	- 0.046	- 0.255	- 0.032	- 0.440	- 0.773 ***			
R RDMG								
pН	- 0.390	0.129	0.011	- 0.039	- 0.289			
oc	- 0.126	0.399	0.147	- 0.034	0.386			
FEAL	- 0.017	0.243	0.242	- 0.031	0.437 *			

Table 13. Results of path analysis for soils spiked with Pb 2000 mg kg⁻¹. Direct effects (*italics*, on the diagonal), indirect effects (off diagonal), and path totals.

CEC -^a Organic carbon.

CEC

^b Best fit data set transformations: studentized (R), log₁₀ (log), square root (SR).
 ^c Amorphous AI and Fe oxide.
 ^d Cation exchange capacity.

0.133

- 0.056

0.085

0.278

- 0.270
| ··· ··· · | С | ontrol Soil | | Zn-spiked Soil | | | |
|-----------|---------|------------------|------|---------------------|-------------------|--------|--|
| Soil | Tissue | DMG ^a | G⁵ | Tissue | RDMG ^c | RG^d | |
| | mg kg⁻¹ | g | % | mg kg ⁻¹ | % | % | |
| 1 | 11.9 | 7.00 | 78.4 | 25.4 | 18.0 | 87.2 | |
| 2 | 24.9 | 6.87 | 72.5 | 333 | 29.1 | 101 | |
| 3 | 16.2 | 6.43 | 85.0 | 110 | 3.40 | 88.2 | |
| 4 | 22.8 | 4.39 | 77.5 | 2038 | 48.7 | 101 | |
| 5 | 24.1 | 8.05 | 83.4 | 96.0 | 37.3 | 104 | |
| 6 | 16.8 | 4.86 | 71.7 | 190 | 45.1 | 105 | |
| 7 | 22.3 | 5.07 | 76.7 | 28.5 | 20.9 | 115 | |
| 8 | 12.2 | 4.41 | 80.0 | 26.4 | 27.2 | 113 | |
| 9 | 16.6 | 3.59 | 86.7 | 28.0 | 58.0 | 94.2 | |
| 10 | 16.6 | 5.58 | 62.5 | 739 | 69.0 | 131 | |
| 11 | 33.2 | 6.99 | 76.7 | 69.7 | 80.3 | 109 | |
| 12 | 33.8 | 5.40 | 91.7 | 64.3 | 60.7 | 98.2 | |
| 13 | 20.2 | 8.01 | 71.7 | 249 | 70.5 | 96.0 | |
| 14 | 17.3 | 6.70 | 50.0 | 148 | 58.2 | 107 | |
| 15 | 18.2 | 6.02 | 81.7 | 932 | 72.8 | 112 | |
| 16 | 18.2 | 5.38 | 86.7 | 574 | 68.8 | 102 | |
| 17 | 18.6 | 5.21 | 81.7 | 38.9 | 26.3 | 104 | |
| 18 | 23.6 | 8.27 | 86.7 | 27.6 | 26.4 | 108 | |
| 19 | 14.5 | 4.35 | 88.4 | 18.4 | 20.0 | 94.3 | |
| 20 | 24.8 | 6.10 | 55.0 | 598 | 85.4 | 142 | |
| 21 | 17.0 | 5.45 | 83.4 | 631 | 69.0 | 190 | |
| | | | | | | | |
| Mean | 20.2 | 5.91 | 77.5 | 332 | 47.4 | 110 | |
| Min. | 11.9 | 3.59 | 50.0 | 18.4 | 3.40 | 87.2 | |
| Max. | 33.8 | 8.27 | 91.7 | 2038 | 85.4 | 190 | |

Table 14. Bioassay endpoints for lettuce grown in control soils and Zn-spiked (300 mg kg^{-1}) soils,

^a Lettuce dry matter growth.
 ^b Percent germination of 20 seeds.
 ^c Relative dry matter growth.
 ^d Relative germination.

	Simple Linear Correlation Coefficients (r)					
Soil Parameter	T	ïssue Zn	RDMG ^a			
	ТÞ	r	Т	r		
Organic carbon	log	- 0.411	R	- 0.224		
Clay	log	- 0.793 ***	R	- 0.474 *		
Amorphous AI and Fe oxide	R	- 0.368	R	0.020		
рН	log	- 0.887 ***	R	- 0.546 **		
Cation exchange capacity	log	- 0.860 ***	R	- 0.532 *		
^a Polativo day matter growth		······				

Table 15. Simple linear correlation coefficients for lettuce biological endpoints vs soil parameters for soils spiked with Zn at 300 mg kg⁻¹.

^a Relative dry matter growth.
 ^b Best fit data set transformations: studentized (R), log₁₀ (log), square root (SR).

	Ta	Multiple Linear Regression	R ²
		Model I	
Tissue Zn	log	- 0.671(pH) + 0.112(OC) – 0.452(Clay)	0.91 ***
RDMG⁵	R	- 0.433(pH) + 0.102 (OC) - 0.269(Clay)	0.34
		Model II	
Tissue Zn	log	- 0.518(pH) + 0.303(OC) – 0.028(FEAL) – 0.665(CEC)	0.91 ***
RDMG	Ř	- 0.041(pH) + 0.106(OC) + 0.396(FEAL) - 0.793(CEC)	0.43
^a Best fit data ^b Relative dry	a set tra / matte	nsformations: studentized (R), log₁₀ (log). r growth.	

Table 16. Comparison of multiple regression equations and R^2 for lettuce biological endpoints for two potential mechanistic models.

	Simple Correlation Coefficients (r)								
	pН	OC ^a	Clay	FEAL ^b	CEC℃				
pН	1.00	0.45 *	0.59 **	0.12	0.75 ***				
ÖC		1.00	0.49 *	0.61 **	0.70 ***				
Clay			1.00	0.61 **	0.90 ***				
FEAL				1.00	0.55 **				
CEC					1.00				

Table 17. Simple correlation coefficients (r) for the intercorrelations between soil properties.

^a Organic carbon.
 ^b Amorphous Fe and Al oxide.
 ^c Cation exchange capacity.

Model I										
	рН	OC ^a	Clay	Total						
Log ^b Tissue				· · · · · · · ·						
рН	- 0.671 ***	0.050	- 0.266	- 0.887 ***						
ÖC	- 0.301	0.112	- 0.222	- 0.411						
Clay	- 0.395	0.055	- 0.452 ***	- 0.792 ***						
R RDMG										
pН	- 0.433	0.046	- 0.159	-0.546 **						
ÖC	- 0.194	0.102	- 0.132	- 0.224						
Clay	- 0.255	0.050	- 0.269	- 0.474 *						
		Mo								
	ъЦ			CECd	Total					
Log Tissue	рп	00			TOLAI					
pH	- 0.518 **	0.137	- 0.003	- 0.503	- 0.887 ***					
όc	- 0.232	0.303 *	- 0.017	- 0.406	- 0.352					
FEAL	- 0.060	0.185	- 0.028	- 0.366	- 0.269					
	0.201	0.044	0.010	0 665 ***	0 004 ***					

Table 18. Results of path analysis for soils spiked with Zn (300 mg kg⁻¹). Direct effects (*italics*, on the diagonal), indirect effects (off diagonal) and path totals.

CEC - 0.391 0.211 - 0.016 - 0.665 - 0.861 *** **R** RDMG - 0.041 - 0.546 ** pН 0.048 0.046 - 0.599 00 - 0.019 0.106 0.241 - 0.483 - 0.155 FEAL - 0.005 0.065 0.396 - 0.436 0.020 - 0.031 0.074 CEC 0.218 - 0.793 - 0.532 *

^a Organic carbon.

^b Best fit data set transformations: studentized (R), log₁₀ (log), square root (SR).

^c Amorphous AI and Fe oxide.

^d Cation exchange capacity.

Statistical		Lettuce Endpoint						
Test		Bioaccumulation	RDMG ^a					
ANOVA		Significant	Significant					
Simple Correlation ^b		- Clay**, - pH**, - CEC**	FEAL***, OC*, Clay*					
Multiple Regression	Model I Model II	R ² = 0.63** R ² = 0.66**	R ² = 0.54** R ² = 0.66**					
Path Analysis	Model I Model II	- Clay (d ^c)* - pH (d)*, - FEAL (d)	Clay (d)**, - pH (d)* FEAL (d)**					

Table 19. Summary of statistical analyses for the effects of Cd (300 mg kg⁻¹) on lettuce endpoints.

^a Relative dry matter growth. ^b Significant simple correlation between lettuce endpoint and properties defined in Table 3. ° Direct effect.

Statistical		Lettuce Endpoint							
Test		Bioaccumulation	RDMG ^a						
ANOVA		Significant	Significant						
Simple Correlation ^b		- OC***, - Clay**, - FEAL*, - CEC***	OC*, FEAL*						
Multiple Regression	Model I Model II	R ² = 0.64*** R ² = 0.67***	$R^2 = 0.35$ $R^2 = 0.36$						
Path Analysis	Model I Model II	[- OC (d)**+ - Clay (i ^d)]*** [- CEC (d) + - OC (i)]***	OC* [OC (i) + FEAL (d)]*						
^a Relative dry ma ^b Significant sim ^c Direct effect. ^d Indirect effect.	atter growth. ple correlatio	n between lettuce endpoint and propertie	s defined in Table 3.						

Table 20. Summary of statistical analyses for the effects of Pb (2000 mg kg⁻¹) on lettuce endpoints.

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Statistical	·	Lettuce Endpoint								
Test		Bioaccumulation	RDMG ^a							
ANOVA		Significant	Significant							
Simple Correlation ^b		- Clay***, - pH***, - CEC***	- Clay*, - pH**, - CEC*							
Multiple Regression	Model I Model II	R ² = 0.91*** R ² = 0.91***	$R^2 = 0.34$ $R^2 = 0.43$							
Path Analysis	Model I Model II	- pH (d)***, - Clay (d)*** - pH (d)**, - CEC (d)***								

Table 21. Summary of statistical analyses for the effects of Zn (300 mg kg⁻¹) on lettuce endpoints.

^a Relative dry matter growth. ^b Significant simple correlation between lettuce endpoints and properties defined in Table 3. ^c Direct effect.







Figure 2. Two path models used to examine the modifying effects of soil properties on metal phytotoxicity.

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Figure 3. pH of control and metal spiked soils.



Figure 4. Distribution, by soil, of soil properties: (A) Organic carbon, (B) cation exchange capacity, (C) amophous Fe and Al oxide, and (D) percent clay.



Figure 5. (A) Dry matter growth (DMG) and (B) percent germination (G) for lettuce grown in control soils.



Figure 6. Lettuce biological endpoints (A.) tissue Cd., (B.) relative dry matter growth (RDMG), and (C.) relative % germination



Figure 7. Lettuce biological endpoints. A. tissue Pb, B. relative dry matter growth (RDMG), and C. relative % germination.



Figure 8. Lettuce biological endpoints. A. tissue Zn, B. relative dry matter growth (RDMG), and C. relative % germination.

APPENDIX

#	Soil & Horizo	on	Metal	Rep	рΗ	Clay	OC	CEC	Al	Fe
						%	%	cmol _c /kg	mg/	kg
1	Canisteo	Α	Control	1	7.5	32.5	3.0	30.17	1186	835
1	Canisteo	Α	Control	2	7.5	45.0	3.0	30.84	1151	781
1	Canisteo	Α	Control	3					1105	770
2	Dennis	Α	Control	1	5.5	20.0	1.9	9.86	814	2753
2	Dennis	Α	Control	2	5.6	27.5	1.9	9.68	864	2960
2	Dennis	Α	Control	3					843	2920
3	Dennis	в	Control	1	6	45.0	0.80	14.53	1213	1352
3	Dennis	в	Control	2	6.1	45.0	0.80	14.68	1126	1286
3	Dennis	В	Control	3					1138	1212
4	Dougerty	Α	Control	1	5.2	15.0	1.1	3.35	255	346
4	Dougerty	Α	Control	2	5.3	7.5	1.3	3.31	270	369
4	Dougerty	Α	Control	3					272	378
5	Hanlon	Α	Control	1	7.4	15.0	1.6	16.56	459	1539
5	Hanlon	Α	Control	2	7.4	20.0	1.6	16.06	431	1507
5	Hanlon	Α	Control	3					451	1541
6	Kirkland	Α	Control	1	5.6	25.0	1.4	14	899	1547
6	Kirkland	Α	Control	2	5.6	37.5	1.5	14.01	903	1616
6	Kirkland	Α	Control	3					876	1567
7	Luton	Α	Control	1	7.1	72.5	2.0	32.48	702	2290
7	Luton	Α	Control	2	7.1	70.0	2.0	32.38	673	2365
7	Luton	Α	Control	3					728	2504
8	Mansic	Α	Control	1	7.7	25.0	1.5	16.59	607	223
8	Mansic	Α	Control	2	7.8	35.0	1.5	16.47	590	212
8	Mansic	Α	Control	3					596	231
9	Mansic	в	Control	1	8	37.5	0.80	11.68	269	114
9	Mansic	в	Control	2	8	32.5	0.50	11.65	216	90.9
9	Mansic	в	Control	3					269	269
10	Norge	Α	Control	1	4	12.5	1.2	4.58	811	778
10	Norge	Α	Control	2	4	22.5	1.2	4.55	875	872
10	Norge	Α	Control	3					823	761
11	Osage	Α	Control	1	6.6	56.3	2.6	28.51	960	5050
11	Osage	Α	Control	2	6.6	55.0	2.6	28.07	1000	5048
11	Osage	Α	Control	3		•			1007	5182
12	Osage	в	Control	1	6.7	62.5	2.0	27.45	1417	7506
12	Osage	в	Control	2	6.8	60.0	2.0	27.48	1453	7986
12	Osage	в	Control	3					1449	8196
13	Pond Creek	Α	Control	1	5.2	22.5	1.9	10.69	886	1413
13	Pond Creek	Α	Control	2	5.2	35.0	1.9	10.63	853	1552
13	Pond Creek	Α	Control	3					845	1397
14	Pond Creek	в	Control	1	6	30.0	0.70	12.5	750	1123
14	Pond Creek	в	Control	2	6	35.0	0.90	12.47	781	1195
14	Pond Creek	в	Control	3					779	1138
15	Pratt	Α	Control	1	6.5	2.5	0.90	4.4	181	157
15	Pratt	Α	Control	2	6.5	7.5	0.90	4.4	200	175
15	Pratt	Α	Control	3					182	160
16	Pratt	в	Control	1	6.4	5.0	0.40	3.36	180	173

#	Soil & Horiz	on	Metal	Rep	рΗ	Clay	OC	CEC	Al	Fe
						%	%	cmol _c /kg	mg/	kg
16	Pratt	В	Control	2	6.4	7.5	0.60	3.43	157	142
16	Pratt	В	Control	3					152	141
17	Richfield	В	Control	1	7.7	42.5	1.0	22.35	725	415
17	Richfield	В	Control	2	7.7	40.0	1.2	22.34	669	357
17	Richfield	В	Control	3					737	412
18	Summit	Α	Control	1	7.1	46.3	2.4	29.34	1414	2345
18	Summit	Α	Control	2	7.2	45.0	2.4	29.37	1364	2204
18	Summit	Α	Control	3					1268	2024
19	Summit	В	Control	1	7.1	56.3	1.2	27.72	997	978
19	Summit	В	Control	2	7.1	57.3	1.3	27.44	996	988
19	Summit	В	Control	3			×			
20	Taloka	Α	Control	1	5.1	7.5	1.3	4.86	596	1808
20	Taloka	Α	Control	2		15.0	1.1	4.83	628	1854
20	Taloka	Α	Control	3					601	1783
21	Teller	Α	Control	1	4.5	10.0	0.90	3.05	523	596
21	Teller	Α	Control	2	4.5	10.0	0.80	2.97	536	588
21	Teller	Α	Control	3					532	629
1	Canisteo	Α	Cd	1	7.8	32.5	3.0	30.17	1186	835
1	Canisteo	Α	Cd	2	7.9	45.0	3.0	30.84	1151	781
1	Canisteo	Α	Cd	3					1105	770
2	Dennis	Α	Cd	1	4.8	20.0	1.9	9.86	814	2753
2	Dennis	Α	Cd	2	4.8	27.5	1.9	9.68	864	2960
2	Dennis	Α	Cd	3					843	2920
3	Dennis	В	Cd	1	5.5	45.0	0.80	14.53	1213	1352
3	Dennis	В	Cd	2	5.5	45.0	0.80	14.68	1126	1286
3	Dennis	В	Cd	3					1138	1212
4	Dougerty	Δ	Cd	1	47	15.0	11	3 35	255	346
4	Dougerty	Δ	Cd	2	4.7	7.5	13	3 31	270	369
4	Dougerty	Δ	Cd	2		1.0	1.9	0.01	272	378
5	Hanlon	Δ	Cd	1	66	15.0	1 6	16.56	459	1539
5	Hanlon	Δ	Cd	2	6.5	20.0	1.0	16.00	431	1507
5	Hanlon	Δ	Cd	3	0.0	20.0		10.00	451	1541
6	Kirkland	Δ	Cd	1	47	25 0	14	14	899	1547
6	Kirkland	A	Cd	2	4.7	37.5	1.5	14.01	903	1616
6	Kirkland	A	Cd	3					876	1567
7	Luton	Α	Cd	1	6.7	72.5	2.0	32.48	702	2290
7	Luton	A	Cd	2	6.9	70.0	20	32.38	673	2365
7	Luton	Α	Cd	3					728	2504
8	Mansic	A	Cd	1	7.6	25.0	1.5	16.59	607	223
8	Mansic	A	Cd	2	7.6	35.0	1.5	16.47	590	212
8	Mansic	Α	Cd	3					596	231
9	Mansic	В	Cd	1	7.5	37.5	0.80	11.68	269	114
9	Mansic	В	Cd	2	7.6	32.5	0.50	11.65	216	90.9
9	Mansic	В	Cd	3					269	269
10	Norge	Ā	Cd	1	3.9	12.5	1.2	4.58	811	778
10	Norge	Α	Cd	2	3.9	22.5	1.2	4.55	875	872
10	Norge	Α	Cd	3					823	761
	-									

#	Soil & Horizo	on	Metal	Rep	рΗ	Clay	OC	CEC	Al	Fe
						%	%	cmol _c /kg	mg/	kg
11	Osage	Α	Cd	1	6.2	56.3	2.6	28.51	960	5050
11	Osage	Α	Cd	2	5.9	55.0	2.6	28.07	1000	5048
11	Osage	Α	Cd	3					1007	5182
12	Osage	в	Cd	1	5.8	62.5	2.0	27.45	1417	7506
12	Osage	В	Cd	2	5.7	60.0	2.0	27.48	1453	7986
12	Osage	В	Cd	3					1449	8196
13	Pond Creek	Α	Cd	1	4.4	22.5	1.9	10.69	886	1413
13	Pond Creek	Α	Cd	2	4.4	35.0	1.9	10.63	853	1552
13	Pond Creek	Α	Cd	3					845	1397
14	Pond Creek	В	Cd	1	5.7	30.0	0.70	12.5	750	1123
14	Pond Creek	В	Cd	2	5.6	35.0	0.90	12.47	781	1195
14	Pond Creek	В	Cd	3		-			779	1138
15	Pratt	Α	Cd	1	5.2	2.5	0.90	4.4	181	157
15	Pratt	Α	Cd	2	5.2	7.5	0.90	4.4	200	175
15	Pratt	Α	Cd	3		•	•		182	160
16	Pratt	В	Cd	1	5.2	5.0	0.40	3.36	180	173
16	Pratt	В	Cd	2	5.1	7.5	0.60	3.43	157	142
16	Pratt	в	Cd	3		•	•		152	141
17	Richfield	В	Cd	1	6.7	42.5	1.0	22.35	725	415
17	Richfield	В	Cd	2	6.8	40.0	1.2	22.34	66 9	357
17	Richfield	в	Cd	3			•		737	412
18	Summit	Α	Cd	1	7	46.3	2.4	29.34	1414	2345
18	Summit	Α	Cd	2	7.1	45.0	2.4	29.37	1364	2204
18	Summit	Α	Cd	3	•	•			1268	2024
19	Summit	В	Cd	1	6.6	56.3	1.2	27.72	997	978
19	Summit	В	Cd	2	6.5	57.3	1.3	27.44	996	988
19	Summit	В	Cd	3	•		•	•		•
20	Taloka	A	Cd	1	4.4	7.5	1.3	4.86	596	1808
20	Taloka	Α	Cd	2	4.5	15.0	1.1	4.83	628	1854
20	Taloka	Α	Cd	3	•	•	•	•	601	1783
21	Teller	Α	Cd	1	3.9	10.0	0.90	3.05	523	596
21	Teller	Α	Cd	2	3.9	10.0	0.80	2.97	536	588
21	Teller	Α	Cd	3		•	•	•	532	629
1	Canisteo	Α	PB	1	7.6	32.5	3.0	30.17	1186	835
1	Canisteo	Α	PB	2	7.6	45.0	3.0	30.84	1151	781
1	Canisteo	Α	PB	3	•	•	•	•	1105	770
2	Dennis	Α	PB	1	4.8	20.0	1.9	9.86	814	2753
2	Dennis	Α	PB	2	4.7	27.5	1.9	9.68	864	2960
2	Dennis	Α	PB	3	•	•	•	•	843	2920
3	Dennis	В	PB	1	5.2	45.0	0.80	14.53	1213	1352
3	Dennis	В	PB	2	5.2	45.0	0.80	14.68	1126	1286
3	Dennis	В	PB	3	•	•	•	•	1138	1212
4	Dougerty	A	PB	1	4.5	15.0	1.1	3.35	255	346
4	Dougerty	A	PB	2	4.5	7.5	1.3	3.31	270	369
4	Dougerty	A	PB	3	• -		•		272	378
5	Hanlon	A	PB	1	6.7	15.0	1.6	16.56	459	1539
5	Hanion	Α	PB	2	6.6	20.0	1.6	16.06	431	1507

#	Soil & Horize	on	Metal	Rep	рΗ	Clay	OC	CEC	Al	Fe
						%	%	cmol _c /kg	mg/	kg
5	Hanlon	Α	PB	3		•	•		451	1541
6	Kirkland	Α	PB	1	4.8	25.0	1.4	14	899	1547
6	Kirkland	Α	PB	2	4.8	37.5	1.5	14.01	903	1616
6	Kirkland	Α	PB	3		•			876	1567
7	Luton	Α	PB	1	6.5	72.5	2.0	32.48	702	2290
7	Luton	Α	PB	2	6.7	70.0	2.0	32.38	673	2365
7	Luton	Α	PB	3		•			728	2504
8	Mansic	Α	PB	1	7.7	25.0	1.5	16.59	607	223
8	Mansic	Α	PB	2	7.7	35.0	1.5	16.47	590	212
8	Mansic	Α	PB	3					596	231
9	Mansic	В	PB	1	7.9	37.5	0.80	11.68	269	114
9	Mansic	В	PB	2	7.7	32.5	0.50	11.65	216	90.9
9	Mansic	В	PB	3		•	•		269	269
10	Norge	Α	PB	1	3.8	12.5	1.2	4.58	811	778
10	Norge	Α	PB	2	3.8	22.5	1.2	4.55	875	872
10	Norge	Α	PB	3	•				823	761
11	Osage	Α	PB	1	5.9	56.3	2.6	28.51	960	5050
11	Osage	Α.	PB	2	5.9	55.0	2.6	28.07	1000	5048
11	Osage	Α	PB	3					1007	5182
12	Osage	В	PB	1	5.9	62.5	2.0	27.45	1417	7506
12	Osage	В	PB	2	5.9	60.0	2.0	27.48	1453	7986
12	Osage	В	PB	3	•	•		×	1449	8196
13	Pond Creek	Α	PB	1	41	22.5	1.9	10.69	886	1413
13	Pond Creek	Α	PB	2	4.1	35.0	1.9	10.63	853	1552
13	Pond Creek	Α	PB	3	•	•	•		845	1397
14	Pond Creek	В	PB	1	5.2	30.0	0.70	12.5	750	1123
14	Pond Creek	В	PB	2	5.1	35.0	0.90	12.47	781	1195
14	Pond Creek	В	PB	3	•	•		•	779	1138
15	Pratt	Α	PB	1	4.6	2.5	0.90	4.4	181	157
15	Pratt	Α	PB	2	4.6	7.5	•	4.4	200	175
15	Pratt	Α	PB	3		•		•	182	160
16	Pratt	В	PB	1	5.1	5.0	0.40	3.36	180	173
16	Pratt	В	PB	2	5.2	7.50	0.60	3.43	157	142
16	Pratt	В	PB	3	•	•	•	•	152	141
17	Richfield	В	PB	1	6.3	42.5	1.0	22.35	725	415
17	Richfield	В	PB	2	6.4	40.0	1.2	22.34	669	357
17	Richfield	В	PB	3	•			•	737	412
18	Summit	Α	PB	1	6.9	46.3	2.4	29.34	1414	2345
18	Summit	Α	PB	2	7	45.0	2.4	29.37	1364	2204
18	Summit	Α	PB	3	•	•	•	•	1268	2024
19	Summit	В	PB	1	6.4	56.3	1.2	27.72	997	978
19	Summit	B	PB	2	6.5	57.3	1.3	27.44	996	988
19	Summit	В	PB	3		<u> </u>				
20	l aloka	A	PB	1	4.2	7.5	1.3	4.86	596	1808
20	i aloka	A	PB	2	4.1	15.0	1.1	4.83	628	1854
20	I aloka	A	PB	3					601	1783
21	Teller	Α	PB	1	4.3	10.0	0.90	3.05	523	596

#	Soil & Horizon		Metal	Rep	pН	Clay	OC	CEC	Al	Fe
						%	%	cmol _c /kg	mg/	kg
21	Teller	Α	PB	2	4.3	10.0	0.80	2.97	536	588
21	Teller	Α	PB	3					532	629
1	Canisteo	Α	ZN	1	7.7	32.5	3.0	30.17	1186	835
1	Canisteo	Α	ZN	2	7.7	45.0	3.0	30.84	1151	781
1	Canisteo	Α	ZN	3					1105	770
2	Dennis	Α	ZN	1	4.7	20.0	1.9	9.86	814	2753
2	Dennis	Α	ZN	2	4.7	27.5	1.9	9.68	864	2960
2	Dennis	Α	ZN	3					843	2920
3	Dennis	в	ZN	1	5.3	45.0	0.80	14.53	1213	1352
3	Dennis	в	ZN	2	5.3	45.0	0.80	14.68	1126	1286
3	Dennis	в	ZN	3					1138	1212
4	Dougerty	Α	ZN	1	4.6	15.0	1.1	3.35	255	346
4	Dougerty	Α	ZN	2	4.6	7.5	1.3	3.31	270	369
4	Dougerty	Α	ZN	3					272	378
5	Hanlon	Α	ZN	1	6.4	15.0	1.6	16.56	459	1539
5	Hanlon	Α	ZN	2	6.4	20.0	1.6	16.06	431	1507
5	Hanlon	Α	ZN	3					451	1541
6	Kirkland	Α	ZN	1	4.8	25.0	1.4	14	899	1547
6	Kirkland	Α	ZN	2	4.7	37.5	1.5	14.01	903	1616
6	Kirkland	Α	ZN	3					876	1567
7	Luton	Α	ZN	1	6.7	72.5	2.0	32.48	702	2290
7	Luton	Α	ZN	2	6.8	70.0	2.0	32.38	673	2365
7	Luton	Α	ZN	3					728	2504
8	Mansic	Α	ZN	1	7.6	25.0	1.5	16.59	607	223
8	Mansic	Α	ZN	2	7.8	35.0	1.5	16.47	590	212
8	Mansic	Α	ZN	3					596	231
9	Mansic	В	ZN	1	7.1	37.5	0.80	11.68	269	114
9	Mansic	В	ZN	2	7.2	32.5	0.50	11.65	216	90.9
9	Mansic	В	ZN	3					269	269
10	Norge	Ā	ZN	1	3.9	12.5	1.2	4.58	811	778
10	Norae	Α	ZN	2	3.9	22.5	1.2	4.55	875	872
10	Norge	Α	ZN	3					823	761
11	Osage	Α	ZN	1	5.8	56.3	2.6	28.51	960	5050
11	Osage	Α	ZN	2	5.8	55.0	2.6	28.07	1000	5048
11	Osage	A	ZN	3					1007	5182
12	Osage	В	ZN	1	6.2	62.5	2.0	27.45	1417	7506
12	Osage	В	ZN	2	6.2	60.0	2.0	27.48	1453	7986
12	Osage	В	ZN	3					1449	8196
13	Pond Creek	Ā	ZN	1	5.8	22.5	1.9	10.69	886	1413
13	Pond Creek	Α	ZN	2	5.7	35.0	1.9	10.63	853	1552
13	Pond Creek	Α	ZN	3					845	1397
14	Pond Creek	В	ZN	1	5.4	30.0	0.70	12.5	750	1123
14	Pond Creek	B	ZN	2	5.4	35.0	0.90	12.47	781	1195
14	Pond Creek	B	ZN	3					779	1138
15	Pratt	Ā	ZN	1	5.3	2.5	0.90	4.4	181	157
15	Pratt	Α	ZN	2	5.3	7.5	0,90	4.4	200	175
15	Pratt	Α	ZN	3			•		182	160

#	Soil & Horizon		Metal	Rep	рΗ	Clay	OC	CEC	Al	Fe
						%	%	cmol _c /kg	mg/kg	
16	Pratt	В	ZN	1	4.4	5.0	0.40	3.36	180	173
16	Pratt	В	ZN	2	4.4	7.5	0.60	3.43	157	142
16	Pratt	В	ZN	3					152	141
17	Richfield	В	ZN	1	6.9	42.5	1.0	22.35	725	415
17	Richfield	в	ZN	2	6.9	40.0	1.2	22.34	669	357
17	Richfield	В	ZN	3					737	412
18	Summit	Α	ZN	1	7.3	46.3	2.4	29.34	1414	2345
18	Summit	Α	ZN	2	7.3	45.0	2.4	29.37	1364	2204
18	Summit	Α	ZN	3					1268	2024
19	Summit	в	ZN	1	6.4	56.3	1.2	27.72	997	978
19	Summit	в	ZN	2	6.4	57.3	1.3	27.44	996	9 88
19	Summit	в	ZN	3						
20	Taloka	Α	ZN	1	4.3	7.5	1.3	4.86	596	1808
20	Taloka	Α	ZN	2	4.3	15.0	1.1	4.83	628	1854
20	Taloka	Α	ZN	3	-		*		601	1783
21	Teller	Α	ZN	1	4.1	10.0	0.90	3.05	523	596
21	Teller	Α	ZN	2	4.1	10.0	0.80	2.97	536	588
21	Teller	Α	ZN	3					532	629

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	Soil & Horizon			REP		DMG	-	Tissue Concentration		
			Metal		G		As	Cd	Pb	Zn
					g	g		r	ng/kg	
1	Canisteo	Α	Control	1	14	6.93	0.00	0.2	0.00	11.3
1	Canisteo	Α	Control	2	20	7.64	0.00	0.1	0.00	12.9
1	Canisteo	Α	Control	3	13	6.44	0.00	0.2	0.00	11.5
2	Dennis	Α	Control	1	13	7.20	0.00	0.7	0.00	20.6
2	Dennis	Α	Control	2	٠		-			
2	Dennis	Α	Control	3	16	6.54	0.00	1.1	0.00	29 .1
3	Dennis	В	Control	1	18	6.15	0.00	0.6	0.00	15.4
3	Dennis	В	Control	2	14	6.27	0.00	0.3	0.00	18.4
3	Dennis	В	Control	3	19	6.87	0.00	0.0	0.00	14.9
4	Dougerty	Α	Control	1	12	3.99	0.00	0.1	0.00	24.0
4	Dougerty	Α	Control	2	19	4.79	0.00	0.0	0.00	21.5
4	Dougerty	Α	Control	3						
5	Hanlon	Α	Control	1	15	8.14	0.00	0.3	0.00	23.5
5	Hanlon	Α	Control	2	19	8.62	0.00	0.3	0.00	27.9
5	Hanlon	Α	Control	3	16	7.40	0.00	0.2	0.00	21.0
6	Kirkland	Α	Control	1	17	5.11	0.00	0.4	0.00	17.8
6	Kirkland	Α	Control	2	13	4.18	0.00	0.0	0.00	16.2
6	Kirkland	Α	Control	3	13	5.29	0.00	0.0	0.00	16.4
7	Luton	Α	Control	1	16	5.35	0.00	0.2	0.00	21.2
7	Luton	Α	Control	2	15	5.00	0.00	0.4	0.00	28.0
7	Luton	Α	Control	3	15	4.86	0.00	0.1	0.00	17.7
8	Mansic	Α	Control	1	17	3.53	0.00	0.0	0.00	11.0
8	Mansic	Α	Control	2		4.66	0.00	0.0	0.00	13.1
8	Mansic	Α	Control	3	15	5.05	0.00	0.0	0.00	12.5
9	Mansic	В	Control	1	18	3.56	0.00	0.0	0.00	10.0
9	Mansic	В	Control	2	15	4.44				
9	Mansic	В	Control	3	19	2.76	0.00	0.0	0.00	23.1
10	Norge	Α	Control	1		5.89	0.00	0.2	0.00	14.4
10	Norge	Α	Control	2	13	7.05	0.00	0.3	0.00	18.2
10	Norge	Α	Control	3	12	3.80	0.00	0.0	0.00	17.2
11	Osage	Α	Control	1	15	5.52	0.00	0.4	0.00	37.3
11	Osage	Α	Control	2	16	8.34	0.00	0.1	0.00	29.7
11	Osage	Α	Control	3	15	7.12	0.00	0.3	0.00	32.5
12	Osage	В	Control	1	20	6.41	0.00	0.5	0.00	22.8
12	Osage	В	Control	2	20	4.35	0.00	0.0	0.00	21.1
12	Osage	В	Control	3	15	5.56	0.00	0.5	0.00	57.6
13	Pond Creek	Α	Control	1	20	6.43	0.00	0.1	0.13	21.0
13	Pond Creek	Α	Control	2	15	8.59	0.00	0.0	0.00	17.4
13	Pond Creek	Α	Control	3	15	9.02	0.00	0.2	0.00	22.3
14	Pond Creek	В	Control	1	12	6.03	0.00	0.9	0.00	24.3
14	Pond Creek	В	Control	2	16	5.23	0.00	0.2	0.00	16.0
14	Pond Creek	В	Control	3	15	8.86	0.00	0.2	0.00	11.5
15	Pratt	Α	Control	1	15	6.11	0.00	0.2	0.00	20.8
15	Pratt	Α	Control	2	16	3.10	0.00	0.2	0.00	14.3
15	Pratt	Α	Control	3	18	8.86	0.00	0.2	0.00	19.5
16	Pratt	В	Control	1	18	4.25	0.00	0.1	0.00	19.8

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#	# Soil & Horizon		Metal	Rep	G	DMG	As	Cd	Pb	Zn
						g		mg/kg		
16	Pratt	В	Control	2	14	7.00	0.00	0.0	0.00	17.8
16	Pratt	В	Control	3	20	4.85	0.00	0.0	0.00	17.0
17	Richfield	В	Control	1	18	4.56	0.00	0.2	0.00	17.4
17	Richfield	В	Control	2	16	4.96	0.00	0.2	0.00	17.6
17	Richfield	В	Control	3	15	6.13	0.00	0.1	0.00	20.8
18	Summit	Α	Control	1	16	10.33	0.00	0.3	0.00	24.7
18	Summit	Α	Control	2	18	9.23	0.00	0.1	0.00	16.2
18	Summit	Α	Control	3	18	5.26	0.00	0.2	0.00	30.0
19	Summit	В	Control	1	17	4.59	0.00	0.0	0.00	13.5
19	Summit	В	Control	2	17	4.07	0.00	0.0	0.00	17.6
19	Summit	В	Control	3	19	4.39	0.00	0.0	0.00	12.5
20	Taloka	Α	Control	1	11	5.87	0.00	0.0	0.00	21.2
20	Taloka	Α	Control	2	11	6.73	0.00	0.0	0.00	28.4
20	Taloka	Α	Control	3		5.71				
21	Teller	Α	Control	1	14	5.26	0.00	0.2	0.00	15.9
21	Teller	Α	Control	2	9	6.46	0.00	0.0	0.00	10.4
21	Teller	Α	Control	3	7	4.62	0.00	0.3	0.00	24.8
1	Canisteo	Α	Cd	1	13	0.15	1.31	127.8	6.64	22.0
1	Canisteo	Α	Cd	2	14	0.16	0.00	120.4	0.00	33.3
1	Canisteo	Α	Cd	3	13	0.12	0.00	122.5	1.89	68.9
2	Dennis	Α	Cd	1	12	0.67	0.00	219.9	0.00	17.0
2	Dennis	Α	Cd	2	9	0.65	0.00	284.3	0.00	18.0
2	Dennis	Α	Cd	3	14	2.27	0.00	222,1	0.00	23.9
3	Dennis	В	Cd	1	12	0.14	0.00	101.4	0.00	5.2
3	Dennis	в	Cd	2	14	0.08	0.00	173,2	0.00	14.4
3	Dennis	в	Cd	3	18	0.12	0.00	91.1	0.00	6.7
4	Dougerty	Α	Cd	1	20	0.08	0.00	1781.3	0.00	80.1
4	Dougerty	Α	Cd	2	9	0.07	0.00	1747.4	0.00	95.7
4	Dougerty	Α	Cd	3	16	0.07	0.00	1572.3	0.00	70.1
5	Hanlon	Α	Cd	1		0.09	0.00	96.9	0.00	5.7
5	Hanlon	Α	Cd	2	12	0.15	0.00		0.00	9.7
5	Hanlon	Α	Cd	3	13	0.08	0.00	123.7	0.00	11.6
6	Kirkland	Α	Cd	1	13	1.13	0.00		0.00	26.9
6	Kirkland	Α	Cd	2	14	0.41	0.00		0.00	40.7
6	Kirkland	A	Cd	3	14	2.70	0.00		0.00	14.1
7	Luton	A	Cd	1	17	1.08	0.00	62.4	0.00	11.9
7	Luton	Α	Cd	2	18	1.17	0.00	55.5	0.00	16.1
7	Luton	Α	Cd	3	20		0.00	55.3	0.00	11.0
8	Mansic	Α	Cd	1	18	0.24	0.00	66.0	0.00	2.2
8	Mansic	A	Cd	2	14	0.18	0.00	86.9	0.00	4.8
8	Mansic	Α	Cd	3	16	0.17	0.00	51.2	0.00	32
9	Mansic	В	Cd	1	12	0.25	0.00	72.3	0.91	7.0
9	Mansic	B	Cd	2	11	0.19	0.00	86.7	3.23	31.4
9	Mansic	B	Cd	3	14	0.32	0.00	62.7	0.00	1.6
10	Norae	Ā	Cd	1	15	0.19	0.00	272.6	0.00	23 7
10	Norge	Α	Cd	2	15	0.11	0.00	200.9	0.00	11.8
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#	Soil & Horizo	on	Metal	Rep	G	DMG	As	Cd	Pb	Zn
						g	ا خدر ہو ہو گیا ہو	n	ng/kg	
10	Norge	Α	Cd	3	17	0.11	0.00	•	2.21	61.3
11	Osage	Α	Cd	1	12	2.83	0.00	60.6	0.00	24.6
11	Osage	Α	Cd	2	15	4.02	0.00	65.4	0.00	29.4
11	Osage	Α	Cd	3	12	3.40	0.00	66.5	0.00	26.9
12	Osage	в	Cd	1	14	3.32	0.00	56.8	0.00	15.8
12	Osage	в	Cd	2	14	3.54	0.00	58.6	0.00	16.8
12	Osage	В	· Cd	3	13	2.16	0.00	87.6	0.00	14.9
13	Pond Creek	Α	Cd	1	10	2.89	0.00	136.8	0.00	21.7
13	Pond Creek	Α	Cd	2	-17	2.06	0.00	284.6	0.00	37.4
13	Pond Creek	Α	Cd	3	18	1.74	0.00	288.3	0.00	39.4
14	Pond Creek	В	Cd	1	12	0.07	0.00	•	0.55	73.4
14	Pond Creek	В	Cd	2	11	0.09	0.00		2.12	13.1
14	Pond Creek	В	Cd	3	11	0.06	0.00		2.63	105.6
15	Pratt	Α	Cd	1	12	0.08	0.00	268.3	0.00	15.2
15	Pratt	Α	Cd	2	13	0.07	0.00	542.1	0.00	16.2
15	Pratt	Α	Cd	3	11	0.09	0.00	399.5	0.00	9.5
16	Pratt	В	Cd	1	13				•	
16	Pratt	в	Cd	2	12	0.11	0.00	216.4	0.00	6.2
16	Pratt	В	Cd	3	15	0.10	0.00	212.8	0.00	13.2
17	Richfield	в	Cd	1	14	0.13	0.00	212.0	0.00	12.1
17	Richfield	в	Cd	2	18	0.21	0.00	292.9	0.00	8.9
17	Richfield	в	Cd	3	14	0.25	0.00	247.4	0.00	8.0
18	Summit	Α	Cd	1	12	-	•	-		
18	Summit	Α	Cd	2	15	0.09	0.00	97.9	2.18	60.4
18	Summit	Α	Cd	3	10	0.13	0.00	39.4	0.00	5.2
19	Summit	в	Cd	1	15	0.38	0.00	102.5	0.00	9.1
19	Summit	в	Cd	2	17	0.11	0.00	128.0	0.00	56.0
19	Summit	в	Cd	3	15	0.11	1.54	118.2	1.08	94.6
20	Taloka	Α	Cd	1	17	0.08	0.00	1461.2	0.00	32.4
20	Taloka	Α	Cd	2	10	0.10	0.00	956.8	0.00	26.1
20	Taloka	Α	Cd	3	14	0.14	0.00	1178.5	0.00	36.4
21	Teller	Α	Cd	1	10	0.07	2.29	254.9	0.63	91.3
21	Teller	Α	Cd	2	18	0.20	0.00	128.9	0.17	29.2
21	Teller	Α	Cd	3	12			•		
1	Canisteo	Α	PB	1	14	3.68	1.46	0.2	8.09	10.5
1	Canisteo	Α	PB	2	16	4.79	3.73	0.3	9.36	6.7
1	Canisteo	Α	PB	3	15	1.49	0.00	0.8		12.3
2	Dennis	Α	PB	1	16	1.73	0.00	1.0	77.44	54.5
2	Dennis	Α	PB	2	20	1.54	0.91	1.0	102.31	37.9
2	Dennis	Α	PB	3	18	0.51	0.00	0.9	86.40	34.3
3	Dennis	В	PB	1	17	0.14	0.00	0.0	159.94	32.9
3	Dennis	в	PB	2	15	0.13	0.00	0.0	159.84	38.1
3	Dennis	В	PB	3	18	0.21	0.00	1.4	•	23.9
4	Dougerty	Α	PB	1	18	2.01	0.00	0.1	109.21	29.2
4	Dougerty	Α	PB	2	20	0.71	0.00	0.2	83.30	42.8
4	Dougerty	Α	PB	3	19	0.82	0.00	0.1	150.20	42.8

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#	Soil & Horizon		Metal	Rep	G	DMG	As	Cd	Pb	Zn
				-		g		r	ng/kg	بد بد و م می و و م
5	Hanlon	Α	PB	1	17	3.32	0.00	0.0	6.79	15.7
5	Hanlon	Α	PB	2	15	3.05	0.00	0.3	11.84	22.1
5	Hanlon	Α	PB	3	16	3.16	0.00	0.2	8.97	20.1
6	Kirkland	Α	PB	1	13	2.24	0.00	0.8	45.92	27.8
6	Kirkland	Α	PB	2	15	5.24	0.00	0.4	53.35	22.6
6	Kirkland	Α	PB	3	20	5.42	0.00	0.4	52.04	22.6
7	Luton	Α	PB	1	19	0.57	0.00	1.6	15.65	25.7
7	Luton	Α	PB	2	14	1.36	0.00	1.2	14.35	18.4
7	Luton	Α	PB	3	17	1.49	0.00	1.0	19.44	15.4
8	Mansic	Α	PB	1	7	0.77	0.00	0.1	89.38	8.3
8	Mansic	Α	PB	2	18	1.38	0.00	0.0	35.08	4.2
8	Mansic	Α	PB	3	15	1.05	0.00	0.0	56.18	4.8
9	Mansic	в	PB	1	18	0.20	0.00	0.0	49.88	7.9
9	Mansic	в	PB	2	15	0.11	0.00	0.0	69.08	6.0
9	Mansic	в	PB	3	19	0.09	0.00	0.0		7.5
10	Norge	Α	PB	1	15	3.85	0.00	0.0	72.51	28.6
10	Norge	Α	PB	2	17	2.04	0.00	0.2	56.35	18.3
10	Norge	Α	PB	3	18	2.37	0.00	0.0	54.91	17.9
11	Osage	Α	PB	1	19	1.14	0.00	0.1	3.84	19.0
11	Osage	Α	PB	2	17	4.42	0.00	0.1	2.47	18.3
11	Osage	Α	PB	3		3.76	0.00	0.1	3.35	18.2
12	Osage	в	PB	1	18	5.55	0.00	0.4	11.63	9.7
12	Osage	в	PB	2	17	3.53	0.00	0.8	22.86	18.2
12	Osage	в	PB	3	18	3.18	0.00	0.6	11.32	18.0
13	Pond Creek	Α	PB	1		7.30	0.00	0.1	49.27	22.0
13	Pond Creek	Α	PB	2	18	7.45	0.00	0.2	37.47	21.2
13	Pond Creek	Α	PB	3	16	4.59	0.00	0.2	42.48	15.5
14	Pond Creek	в	PB	1	16					
14	Pond Creek	в	PB	2	17	0.76	0.00	1.7	95.21	31.9
14	Pond Creek	в	PB	3	17	1.16	0.00	1.7	96.10	23.6
15	Pratt	Α	PB	1	18	0.28	0.00	0.0	208.46	20.1
15	Pratt	Α	PB	2	14	0.19	0	0.5	244.20	56.2
15	Pratt	Α	PB	3	18	0.21	0.00	0.5	246.94	54.3
16	Pratt	в	PB	1	18	0.24	0.00	0.0	100.90	5.0
16	Pratt	в	PB	2	19	0.13	0.00	0.0		44.5
16	Pratt	в	PB	3		0.15	0.00	0.1	122.13	9.1
17	Richfield	В	PB	1	16	0.41	0.00	4.0	55.37	11.6
17	Richfield	В	PB	2	15	1.10	0.00	3.9	26.17	10.2
17	Richfield	В	PB	3	19	1.32	0.00	2.9	31.72	9.5
18	Summit	Α	PB	1	16	0.24	0.00	0.6	27.37	26.2
18	Summit	Α	PB	2	8	0.24	0	1.8	49.05	37.2
18	Summit	Α	PB	3	16	0.40	0.00	1.0	14.33	16.5
19	Summit	в	PB	1	18	1.91	0.00	0.7	12.54	11.9
19	Summit	в	PB	2	16	1.56	0.00	0.6	17.59	11.3
19	Summit	в	PB	3	20	1.41	0.00	1.1	10.08	13.8
20	Taloka	Α	PB	1	18	2.93	0.00	0.1	41.88	30.5

								-	Tissue	
#	Soil & Horizo	on	Metal	Rep	G	DMG	As	Cd	Pb	Zn
						g			mg/kg	
20	Taloka	Α	PB	2	18	3.40	0.00	0.0	43.93	22.6
20	Taloka	Α	PB	3		5.71	0.00	0.0	27.23	18.5
21	Teller	Α	PB	1	13	0.49	0.00	0.3	138.42	22.0
21	Teller	Α	PB	2	20	2.70	0.00	0.0	79.10	11.9
21	Teller	Α	PB	3		1.78	0.00	0.2	107.31	14.5
1	Canisteo	Α	ZN	1	8	0.87	0.00	0.0	0.00	23.3
1	Canisteo	Α	ZN	2	19	1.12	0.26	0.0	0.00	25.0
1	Canisteo	Α	ZN	3	14	1.81	0.00	0.0	0.07	27.9
2	Dennis	Α	ZN	1	16	1.18	0.00	0.4	0.00	218.4
2	Dennis	Α	ZN	2	14	2.80	0.00	0.6	0.00	320.5
2	Dennis	Α	ZN	3	14	2.01	0.00	0.8	0.00	459.2
3	Dennis	В	ZN	1	16	0.12	0.11	1.3	0.24	139.2
3	Dennis	В	ZN	2	13	0.18	0.00	0.1	0.00	127.5
3	Dennis	В	ZN	3	16	0.35	0.00	0.1	0.00	62.8
4	Dougerty	Α	ZN	1	15	2.65	0.00	3.3	0.00	2587
4	Dougerty	Α	ZN	2	16	1.97	0.00	0.5	0.00	1407
4	Dougerty	Α	ZN	3	16	1.79	0.00	1.4	0.00	2120
5	Hanlon	Α	ZN	1	17	3.71	0.00	0.0	0.00	99.7
5	Hanlon	Α	ZN	2	17	2.28	0.00	0.2	0.00	92.3
5	Hanlon	Α	ZN	3	18		0.00	0.0	0.00	
6	Kirkland	Α	ZN	1	15	3.17	0.00	0.3	0.40	198.9
6	Kirkland	Α	ZN	2	14	1.21	0.00	0.1	0.00	191.4
6	Kirkland	Α	ZN	3	16		0.00	0.2	0.00	178.9
7	Luton	Α	ZN	1	16	0.83	0.00	0.2	0.00	35.3
7	Luton	Α	ZN	2	19	1.14	0.00	0.0	0.00	25.1
7	Luton	Α	ZN	3	18	1.20	0.00	0.0	0.00	25.2
8	Mansic	Α	ZN	1	20	0.60	0.00	0.0	0.00	
8	Mansic	Α	ZN	2	17	1.96	0.00	0.0	0.00	24.9
8	Mansic	Α	ZN	3	17	1.04	0.00	0.0	0.00	27.8
9	Mansic	В	ZN	1	17	3.09	0.00	0.0	0.00	
9	Mansic	В	ZN	2	18	1.41	0.00	0.0	0.00	23.8
9	Mansic	в	ZN	3	14	1.75	0.00	2.3	0.00	32.2
10	Norae	Α	ZN	1	18	3.67	0.00	0.4	0.00	501.9
10	Norae	Α	ZN	2	14	4.23	0.00	0.5	0.00	883.9
10	Norge	Α	ZN	3	17	3.64	0.00	0.5	0.00	832.0
11	Osage	Α	ZN	1	15	6.18	0.00	0.6	0.00	72.7
11	Osage	Α	ZN	2	17	3.09	0.00	0.8	0.00	64.6
11	Osage	A	ZN	3	18	7.56	0.00	0.9	0.00	71.9
12	Osage	В	ZN	1	20	2.76	0.00	0.3	0.00	67.0
12	Osage	В	ZN	2	17	3.16	0.00	0.4	0.00	61.6
12	Osage	в	ZN	3	17	3.91	0.00	0.6	0.00	
13	Pond Creek	Ā	ZN	1	17	4.55	0.00	0.4	0.00	223.0
13	Pond Creek	Α	ZN	2	15	6.25	0.00	0.7	0.00	291.2
13	Pond Creek	Α	ZN	3	16	6.15	0.00	0.6	0.40	232.9
14	Pond Creek	В	ZN	1	14	3.61	0.00	0.7	0.00	181.2
14	Pond Creek	B	ZN	2	17	3.95	0.00	0.2	0.00	123.2
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							Tissue				
#	Soil & Horizon		Metal	Rep	G	DMG	As	Cd	Pb	Zn	
						g	mg/kg				
14	Pond Creek	В	ZN	3	15	4.15	0.00	0.5	0.00	139.5	
15	Pratt	Α	ZN	1	18	2.82	0.00	0.8	0.00	1310	
15	Pratt	Α	ZN	2	20	3.95	0.00	0.6	0.00	682.9	
15	Pratt	Α	ZN	3	17	6.36	0.00	0.3	0.00	803.3	
16	Pratt	В	ZN	1	19	3.23	0.00	0.2	0.00	726.0	
16	Pratt	В	ZN	2	18	3.70	0.00	0.0	0.00	583.7	
16	Pratt	В	ZN	3	16	4.15	0.00	0.0	0.00	411.3	
17	Richfield	В	ZN	1	14	2.54	0.00	0.2	0.00	43.0	
17	Richfield	В	ZN	2	18	0.54	0.00	0.1	0.00	38.3	
17	Richfield	В	ZN	3	19	1.02	0.00	0.1	0.00	35.3	
18	Summit	Α	ZN	1	17	2.18	0.00	0.0	0.00	27.6	
18	Summit	Α	ZN	2	20		0.00	0.0	0.00		
18	Summit	Α	ZN	3	19		0.00	0.0	0.00		
19	Summit	В	ZN	1	14	1.17	0.00	0.0	0.00	20.0	
19	Summit	В	ZN	2	18	0.71	0.00	0.0	0.00	17.5	
19	Summit	В	ZN	3	18	0.72	0.38	0.2	1.18	17.7	
20	Taloka	Α	ZN	1	13	5.13	0.00	0.2	0.00	576.6	
20	Taloka	Α	ZN	2	19	5.44	0.00	0.1	0.00	470.8	
20	Taloka	Α	ZN	3	15	5.06	0.00	0.3	0.00	746.6	
21	Teller	Α	ZN	1	18	1.39	0.00	0.2	0.00	743.3	
21	Teller	Α	ZN	2	19	3.93	0.00	0.2	0.00	526.8	
21	Teller	Α	ZN	3	20	5.94	0.00	0.3	0.13	622.2	



Elizabeth Ann Dayton

Candidate for the Degree of

Doctor of Philosophy

Thesis: RELATIVE CONTRIBUTION OF SOIL PROPERTIES TO MODIFYING THE PHYTOTOXICITY AND BIOACCUMULATION OF CADMIUM, LEAD AND ZINC TO LETTUCE

Major Field: Soil Science

Biographical:

Education: Received Bachelor of Science degree in Environmental Science from University of Massachusetts, Amherst, Massachusetts, in May 1995. Received the Master of Science degree in Soil Science from Oklahoma State University, Stillwater, Oklahoma, in May 1999. Completed the requirements for the Doctor of Philosophy degree with a major in Soil Science at Oklahoma State University in Stillwater, Oklahoma, in December 2003.

Experience: Employed by Oklahoma State University, Department of Plant and Soil Sciences as a Graduate Research Assistant, 1996 to present.