LABILITY AND SOLUBILITY OF TRACE METALS IN SOILS

By

Lingchen Mao

(BSc. (Hons) Environmental Science)

Thesis submitted to the University of Nottingham for the degree of Doctor of Philosophy

September 2013

ABSTRACT

The continuing need for improved assessment of risk from heavy metal contamination of the environment has prompted scientific interest in quantifying and predicting metal solubility, 'lability' and bioavailability. This has led to the development of new techniques to fractionate and speciate trace metals in soils. The objectives of the current study were to increase understanding of the effects of (i) soil properties, (ii) contaminant source and (iii) contact time on metal lability and solubility in soils. Multi-stable isotope dilution (ID) methods were used to determine the lability (E-values) of Ni, Cu, Zn, Cd and Pb in soils, alongside more traditional approaches employed for metal fractionation including single and sequential extraction procedures. Most of the work was undertaken using (i) archived soils amended by metal salts (MA soils; n=23) and (ii) topsoils collected from Nottingham, Wolverhampton and London (Urban soils; n=100). The resulting data was used first to quantify the factors affecting trace metal lability in the MA soils using a logistic (S-shape) model which described metal E-value primarily as a function of soil pH with secondary influences from other soil properties. It was apparent that mineral oxides were important fixation phases for Ni, Zn and Cd while Pb was strongly affected by organic matter in soils. This model, parameterised on soils contaminated originally by metal nitrate solution, was then applied to the Urban soils to reveal the extent to which contaminant source still controlled metal lability. A further investigation of the long-term effect of metal source on metal lability was pursued through a third dataset of rural roadside soils (n=42) which had received Pb mainly from petrol-derived and geogenic sources, defined by their isotopic signature. It was demonstrated that petrol-derived Pb remained more labile than Pb from the parent material, despite decades of contact, although both petrol-derived and geogenic Pb contributed to both the labile

and non-labile fractions. In a fourth dataset, soils that had received Pb from sewage sludge amendment (n=16), the co-existence of high phosphate concentration from sewage sludge limited the magnitude and range of Pb lability, probably through formation of Pb-phosphate minerals. No consistent agreement was found between labile fraction of Pb and any single sequential extraction (SEP) fraction in all soils contaminated by Pb from multiple sources.

Both empirical equations (extended Freundlich) and mechanistic models (WHAM-VII) were used to predict metal solubility in the MA and Urban soils. The advantage of using E-values (M_E) over metal extractable by dilute nitric acid (M_{Ext}) to represent the reactive metal fraction in predictions of solubility was very clear for WHAM-VII, but not for the ‘locally parameterised’ Freundlich model. This was almost certainly due to the strong links between pH and E-value becoming subsumed into the coefficient nominally describing the direct influence of pH on metal solubility in the extended Freundlich equation. However, overestimation of the solution concentration from WHAM-VII was observed for all five metals, and strongly correlated with soil pH. Fractionation information from WHAM suggested that the source of the model’s underestimation of metal binding most likely lay with errors in the description of metal binding by Fe and Mn oxides for Ni, Zn and Cd and humic acid (HA) for Cu. An additional factor is the absence of potential binding phases in the WHAM model, such as particulate $CaCO_3$, and the greater diversity of active adsorbents in soils at high pH values.

WHAM is based on the assumption that all metal bound to HA is labile. However, in the current study, ‘non-labile’ Cu, Zn and Pb fractions were observed in suspensions of HA extracted from grassland and peat soils. These were quantified by measuring metal E-values and EDTA-extraction of HA-bound metal using size-exclusion

chromatography (SEC) coupled to ICP-MS to separate free and HA-complexed metal forms. Evidence of time-dependent metal fixation by HA was found for all three metals during the course of a 40 and 160 day incubation study. The proportion of non-labile Cu held by HA could be 40-50%. The presence of a non-labile metal fraction held by HA may substantially invalidate the assumption of reversible equilibrium which is central to all current geochemical models of metal binding to humic substances.

To my parents, *MINGQIANG* and *CAIMIN*, who are dearest to my heart

ACKNOWLEDGEMENT

I would like to offer my sincerest gratitude to my supervisors, Dr Liz Bailey and Dr Scott Young for their supervision and support throughout my PhD study with patience, motivation, enthusiasm and immense knowledge. This thesis would not have been possible without their guidance.

I thank the University of Nottingham for providing research funding during the first three years of my PhD.

I would also like to thank all my colleagues in the Environmental Science department, University of Nottingham for support and sharing over the course of my PhD. A special thank to Mr Darren Hepworth, Mr John Corrie, Mrs Emma Hooley and Dr Sue Grainger for their technical and administrative support.

Last but not least, I would like to express my thanks to my parents for loving and encouraging me during my pursuit of academic achievements; my friends for their spiritual support during my time in Nottingham.

CONTENTS

LIST OF TABLES

LIST OF FIGURES

1 CHAPTER 1: INTRODUCTION

1.1 Overview

Assessment of risk from heavy metal contamination of the environment has increasingly emphasised the need to consider metal 'reactivity' in soils (Fairbrother et al., 1999; Lock and Janssen, 2001). This has led to greater interest in quantifying metal solubility, 'lability' and bioavailability and the development of new techniques to fractionate and speciate trace metals in soils (e.g. Meers et al., 2006; Sterckeman et al., 2009; Huang et al., 2011; Zhang et al., 2001).

Metal held in the soil solid phase is distributed in several operationally defined fractions (Degryse et al., 2009). 'Inert' forms of metal are strongly incorporated into primary minerals, precipitated within secondary alteration products or possibly occluded within organic compounds. Inert metal may only become available to biological and geochemical reactions through very slow processes that alter the solid substrate, such as weathering. 'Non-labile' forms of metal are present as pure or mixed solids, in crystal lattices, or in the internal pores of sorbents (such as mineral (hydr)oxides and humus) possibly following extended intra-particle diffusion from the soil solution. Non-labile metal does not contribute to the immediate solid↔solution distribution that occurs within the labile metal pool but participates in reactions through kinetically moderated processes over periods of months or years. The 'labile' metal fraction exists in the soil pore water and adsorbed on solid particulate surfaces. It is immediately reactive in response to changes in equilibrium conditions and subject to kinetically controlled transfers with the non-labile metal pool. Therefore, the labile pool for trace metals in soils may provide a more appropriate basis for expressing a solid-liquid partition coefficient in soil (Welp and Brummer, 1999; Degryse et al., 2003), on which to base transport calculations and models of plant uptake, rather than the total soil metal content (Streck and Richter, 1997a, b).

Free metal ions in solution, and their inorganic complexes, are generally considered the most reactive species in terms of reaction with the solid phase and are able to exchange with those reactively bound to suspended colloids (Degryse et al., 2009). Free metal ions in solution have also been suggested as the pool most readily available for plant uptake. Thus, the 'Free Ion Activity' (FIA) has been suggested to be the most appropriate determinant of plant toxicity (Di Toro et al., 2001; Thakali et al., 2006) and plant uptake of metals (Hough et al., 2005). However, not all metal assigned to the solution phase is labile. Metal ions in solution may be bound to suspended 'nano-particulate' colloids (< 0.1 μm) including clay, organic matter and sesquioxides (Tack, 2010; Sposito, 2008) either in a labile or non-labile form (Hamon and McLaughlin, 2002). A non-labile metal fraction has been detected in soil solutions associated with dissolved organic matter (DOM) and (hydr)oxides by several researchers (Lombi et al., 2003; Ma et al., 2006a; Nolan et al., 2009). Trace metals in such suspended colloidal particles (SCP-metal) are presumably not readily available for plant uptake but may enhance metal mobility in the environment.

Published literature (Degryse et al., 2004, Tack, 2010, Hammer et al., 2006) generally suggests that the lability of trace metals in soils is the net result of three factors: (i) soil properties, (ii) source characteristics and (iii) contact time. Relevant soil properties include soil physicochemical characteristics such as pH and redox potential and the proportions of soil constituents such as Fe/Mn (hydr)oxide, or organic matter present. For example, trace metal lability is generally lowest in alkaline soils with high mineral oxide and carbonate contents (Buekers et al., 2007; Tye et al., 2003) and greatest in acidic peat soils. Trace metal binding to soil organic matter (SOM) is also strongest at high pH (Tack, 2010) but it appears that the effect of humus on lability is not consistent for all trace metals (Degryse et al., 2009). Furthermore, in alkaline conditions, and at low electrolyte concentrations, humic acids (HAs) and fulvic acids (FAs) are more likely to disperse as colloidal suspensions and contribute to the range of trace metal complexes in the solution phases (Karathanasis, 1999) - especially for trace metals such as Cu that have a particularly strong affinity for humus.

Sources of contamination (natural or anthropogenic) may vary greatly in their intrinsic metal lability, and those characteristics may persist for a long time following contact with soil. For example, previous studies have suggested that Pb from petrol and coal combustion particulates is more soluble in dilute acid extractions than Pb from parent material which tends to be associated with the residual pool of sequential extraction procedures (SEPs) (Erel et al., 1997; Li et al., 2011). Conversely, minespoil-derived Pb, Cd and Zn appear to remain much less labile than indigenous soil metal long after disposal of spoil in mine sites around the UK (Marzouk et al., 2013).

Time-dependent fixation or, conversely, slow release of trace metals from contaminant sources may take place in soils. The rates of these opposing reactions may be strongly affected by soil properties such as soil pH and Eh (Tye et al., 2003). For example metal sulphide ore minerals are likely to weather through oxidation in a topsoil environment whereas Pb deposited as relatively soluble PbO particulates from vehicle fuel combustion may slowly become occluded within soil phosphate minerals (Kaste et al., 2006; Atkinson et al., 2011).

In this chapter, a general introduction to the analytical methods used to determine trace metal fractionation is presented to underpin the later chapters which deal with how the three factors discussed above (soil, source and time) may affect the lability and solubility of soil-borne metals.

1.2 Analysis and Fractionation of Trace metals in Soils

Hot acid digestion (e.g $HF/HClO_4/HNO_3$ acid digestion) mobilizes essentially all forms of trace metal in soils. Therefore, a range of techniques have been developed to assess the mobility and solubility of trace element fractions in soil solid fraction. Popular methods including chemical fractionation procedures utilizing single and sequential extractions (Ure, 1996; Thornton et al., 2008; Atkinson et al., 2011); isotopic dilution techniques (Tongtavee et al., 2005; Tye et al., 2003; Young et al., 2000); diffusive gradient in thin film technique (DGT) (Zhang et al., 2001; Zhang and Young, 2005); and fractionation based on particle size e.g. size exclusion chromatography (SEC) (Laborda et al., 2009; Schmitt et al., 2001) or Field-flow fractionation (FFF) (Laborda et al., 2011). In addition, sophisticated geochemical speciation models such as WHAM (Tipping et al., 2003) or NICA-Donna (Bonten et al., 2008) may also be used to predict metal fractionation and speciation in the solid and solution phases of soil suspensions, respectively.

1.2.1 Single chemical extraction

Single extraction procedures are methodologically attractive; a wide range of protocols have been applied to assess the lability of cationic trace metals in soils in many published studies (Table 1-1). The methods vary with respect to the chemical nature and concentration of extracting solutions, solution:soil ratio, shaking speed, temperature and extraction time. Most of the reagents used for extraction can be categorized into three groups: dilute acids, chelating agents and neutral salts (Laing, 2010a).

Table 1-1: Selected methods for single chemical extraction.

Group	**Extractants**	**Concentration (mol L^{-1})**	**Reference**
Dilute acids	Nitric acid	0.43	Tipping et al., (2003)
		0.22	Almas et al., (2007)
		2.0	Novozamsky et al., (1993)
	CH_3COOH	0.10-0.43	Ure, (1996); Ure et al., (1993)
	HCl	0.1-1.0	Novozamsky et al., (1993)
Chelating agents	EDTA	0.05	Ure, (1996)
	DTPA	0.005	Lindsay and Norvell, (1978)
Salt solutions	$CaCl_2$	0.01 - 1	Young et al., (2000) Novozamsky et al., (1993) Ure, (1996)
	$NaNO_3$	0.1	Gupta and Aten, (1993)
	NH_4NO_3		Novozamsky et al., (1993)
	$Ca(NO_3)_2$	0.10	Meers et al., (2007)
	$AlCl_3$	0.3	Hughes and Noble, (1991)
	$MgCl_2$	0.02-0.1	Makino et al., (2006)

Dilute acids, typically nitric acid, hydrochloric acid and acetic acid, can partially dissolve trace elements associated with a range of discrete fractions such as the exchangeable pool, carbonates, iron and manganese (hydr)oxides and organic matter. Among these, nitric acid may be particularly useful as no interference from the counterion in the mobilization of metals is expected (Laing, 2010a). In recent studies, the metal fraction extracted with nitric acid, at a range of concentrations, has been successfully used as an input variable to geochemical models to predict solubility in soil (Weng et al., 2002; Tipping et al., 2003; Almas et al., 2007). However, mobilization of non-labile metals by HNO_3 is expected, especially in calcareous and/or heavily contaminated soils with a small fraction of labile metals (Sinaj et al., 2004). Degryse et al. (2009) hypothesized that non-labile metals (Zn) that are occluded in carbonate or soluble Zn-containing minerals will be solubilised by the acid extract. This was also demonstrated for Zn, Cd and Pb by Marzouk et al. (2013) comparing a large number of acidic organic soils with neighbouring calcareous soils directly affected by Pb minespoil.

Neutral salt solutions are often used to extract exchangeable trace elements which are assumed to be retained mainly by electrostatic attraction on charged surfaces in soils. This fraction has also been assumed to represent the ‘bioavailabile’ or ‘plant-available’ pool of trace metals (Novozamsky et al., 1993) and the fraction that controls toxic response and metal uptake by soil organisms (Weltje, 1998; Peijnenburg et al., 1999). Salts of Ca, Ba and Mg have been suggested as the highly effective and selective agents in extracting exchangeable trace metals. However, they may cause serious background problems (interferences) during determination by spectroscopic techniques (Laing, 2010a).

Chelating agents, such as Ethylenediaminetetraacetic acid (EDTA) and diethylene triamine pentaacetic acid (DTPA), are able to dissolve not only the exchangeable element fraction but also metals forming complexes with organic matter, fixed within soil iron and manganese (hydr)oxides or bound to carbonates. Although this is a much more powerful extractant than neutral salts, the use of EDTA, at an appropriate concentration, may be the best choice when trying to estimate the labile pool of several metals using a single extractant (Degryse et al., 2009; Welp & Brummer, 1999; Tongtavee et al., 2005; Gabler et al., 2007). Degryse et al. (2009) suggested the use of concentrations ranging from approximate 50 to 100 mmol EDTA per kg soil, because smaller concentrations may not completely extract the labile pool of metal and larger concentrations will release large amounts of the non-labile metal pool (Degryse et al., 2004). However, problems may still arise for some elements that are unstable in EDTA solutions, such as Ti and Mo (Gabler et al., 2007). Comparisons between EDTA extractable and E-values (isotopic exchangeable fraction; see Section 1.2.3) suggested that EDTA may release non-labile metals including Ni, Cu, Cd and Pb in soils, especially soils with high pH values (> pH 7) (Nakhone and Young et al., 1993; Gable et al., 2007; Marzouk et al., 2013). A relatively weak solution of DTPA (0.005 mol L^{-1}), at pH 7.3 was thought to avoid the release of non-labile metal through carbonate dissolution (Lindsay and Norvell, 1978). However, underestimation of the lability may occur comparing with E-values (Marzouk et al., 2013) simply through lack of complexation capacity.

1.2.2 Sequential extraction procedure (SEP)

In contrast to single extractions, sequential extraction procedures (SEPs) provide an estimation of several fractions of metal expected to be present in soil, rather than simply a distinction between the labile and non-labile fractions. Typically, SEPs use a series of chemical extractants with gradually increasing extraction power. The number of steps may vary from 3 to 7, depending on the purpose of the study (Table 1-2). Many SEPs currently used are modifications of the approaches suggested by Tessier et al. (1979) or Ure et al. (1993) (also known as the ‘BCR’ method), but they vary in the number of fractions and reagents used. The BCR method involves only three steps which are intended to extract metals bound to carbonate, amorphous Fe oxides and organic matter, while the Tessier method partitions metals into five operationally-defined geochemical fractions including exchangeable, carbonate (acid-soluble), Fe and Mn oxides (reducible), organically bound (oxidisable) and residual (inert).

Table 1-2: Selected sequential extraction procedures for the fractionation of trace metals in soils.

	Extraction step						
	F1	**F2**	**F3**	**F4**	**F5**	**F6**	**F7**
Ure etl al. (1993) (BCR)	HOAc	$NH_2OH.HCl$	H_2O_2/ NH_4OAc				
Tessier et al., (1979)	$MgCl_2$	NaOAc	$NH_2OH.HCl$/ HOAc	H_2O_2/ NH_4OAc			
Elliot et al. (1990)	$MgCl_2$	NaOAc	NH_4Ox/ HOx	$Na_4P_2O_7$			
Maclaren and Crawford (1973)	$CaCl_2$	HOAc	$K_4P_2O_7$	NH_4Ox/ HOx	DCB*		
Miller et al. (1986)	$Ca(NO_3)_2$/ $Pb(NO_3)_2$	HOAc/ $Ca(NO_3)_2$	$NH_2OH.HCl$	$K_4P_2O_7$	NH_4Ox/ HOx	NH_4Ox/ HOx	
Krishnamurti et al. (1995)	$Mg(NO_3)_2$	NaOAc	$Na_4P_2O_7$	$NH_2OH.HCl$	H_2O_2/ $Mg(NO_3)_2$	NH_4Ox	NH_4Ox/ AA

*DCB: Dithionite, citrate and bicarbonate extraction

One of the main criticisms for SEPs is the lack of complete specificity in each step of the sequential extraction schemes (Biester and Scholz, 1997; Young et al., 2005; Kim and Fergusson, 1991). For example, Schramel et al. (2000) suggested that BCR-F2 may be incomplete because of the poor reducing power of hydroxylamine-hydrochloride (HA-HCl) for crystalline Fe oxides. Furthermore, ‘re-adsorption’ has been found when there is a change in pH from the previous step (Bermond, 2001). Modified methods have been developed from the Tessier procedure to obtain a more discrete dissolution of the target fractions in soils and sediments (Li et al., 1995, Arunachalam et al., 1996, Ahnstrom and Parker, 2001, Rigol et al., 1999, Li and Thornton, 2001, Brazauskiene et al., 2008). Despite the amount of fractionation information SEPs can provide, the application of such results to risk assessment may be limited because labile is unlikely to correspond to any single SEP fraction (Atkinson et al., 2011; Ahnstrom and Parker, 2001). Therefore, care should be taken when drawing conclusions concerning bioavailability to plants, or bioaccessibility following soil ingestion, based on SEPs results (Laing, 2010a). Considerable effort has gone into the standardisation of SEPs including the reagents used and the order of their application (Clark, 2000; Stalikas et al., 1999). It is widely accepted that the schemes in use are largely ‘operational’ in nature and therefore their validity lies in comparative studies which requires strict adherence to standard approaches and the availability of certified reference materials for quality control (Laing, 2010a).

1.2.3 Isotopic dilution technique (ID)

Isotope dilution is perhaps the most appropriate method to determine the ‘labile’ or ‘reactive’ metal pool in soils (Degryse et al., 2009). It was initially used to quantify the availability of essential plant nutrients (Lopez and Graham, 1972; Tiller 1972 a,b).

However, more recently it has been applied to the problem of defining the reactivity of toxic metals in the environment (Young et al., 2000). The method defines an amount of metal, distributed between the solution and solid phases, which is isotopically exchangeable, known as the *E*-value (Smolders et al., 1999; Young et al., 2000). Both radioactive and stable isotopes can be used to quantify this fraction, although stable isotopes provide several advantages, such as providing the operator with a non-radioactive environment and fewer problems of waste disposal (Sterckeman et al., 2009). There is also greater experimental flexibility in using stable isotopes because they present none of the limitations on time inherent in using short-lived radioisotopes (Lombi et al., 2003; Nolan et al., 2004) and they even present the possibility of field-based experiments (Chilimba et al., 2012). However, because they are usually more analytically demanding than radioisotopes, stable isotopes have not been widely used in ID methods until recently with the development of rapid and sensitive instrumentation – principally Inductively Coupled Plasma-Mass Spectrometry (ICP-MS) (Degryse et al., 2007; Gabler et al., 2007; Nolan et al., 2004).

To measure isotopically exchangeable metal in soil, typically a soil sample is first suspended in a dilute electrolyte typically $Ca(NO_3)_2$ or $CaCl_2$ for a specific *pre-equilibrium* time. A comparatively small amount of an isotope of the analyte of interest (the ‘spike’) is then added into the suspension in a small volume. After a period of equilibration (typically 1 – 3 days) the supernatant will be separated from the solid (centrifugation and filtering) and the isotopic abundance of the spike isotope determined in spiked and non-spiked samples. The extent to which the spike isotope has mixed with the indigenous soil metal can then be used to quantify the ‘isotopically exchangeable’ metal pool in the soil; details of the calculation are described in Chapter 2.

Choice of spike isotopes and their concentrations:

There are currently no guidelines for the choice and concentration of stable isotopes to be used in ID measurements. It is an absolute requirement that the amount of enriched isotope solution (spike) added to the system must be sufficiently large to cause a quantifiable change in the isotopic ratio of the spiked soil in comparison to the natural isotopic ratio of the system (Midwood, 2007). However, it is also important not to perturb the natural metal dynamics of the soil by adding large amounts of the isotope tracer. As an example for Pb, Atkinson et al. (2011) used ^{204}Pb (1.4% IA) added at ~2.70 times the native ^{204}Pb content to determine Pb lability in soils, while Degryse et al. (2007) and Gabler et al. (2007) used Pb isotopes with higher natural abundance of ^{208}Pb (52.4% IA) at ~ 4.0 times the native ^{208}Pb content and ^{207}Pb (22.1% IA) at $^{1}/_{3}$rd of EDTA extractable ^{207}Pb content, respectively. The key considerations in determining the amount of spike to be used are (i) the 'expected' concentration of labile soil metal with which the spike will mix, (ii) the natural isotopic abundance of the spike isotope, (iii) the solubility of metal in the soil suspension used to determine E-values (Marzouk et al., 2013) and (iv) the isotopic abundance of the spike isotope in spike solution. Marzouk et al., (2013) used ^{70}Zn (0.62% IA), ^{108}Cd (0.89% IA) and ^{204}Pb (1.4% IA) on eight soils and tested five levels of spike amount (10%, 30%, 50%, 70% and 100% of the native spike isotope). Good agreement was found in E-values across this very wide range of spike isotope enrichments. Generally, a spike isotope with a low natural abundance is preferred as lower absolute amounts of spike are required to produce a measureable difference in isotopic abundance (Hamon et al., 2008, Atkinson et al., 2011). However, this decision may be over-ruled if there are analytical difficulties (e.g. isobaric interferences) with a particular isotope.

Composition of suspending electrolyte and duration of pre- equilibrium:

Choice of suspending solution is critical in the isotopic dilution method. It must dissolve enough metal from the soil to achieve reliable measurement of the isotopes in the solution phase but must not mobilise non-labile metal ions from the binding phases (Young et al., 2006). In various studies, the choices of suspending solutions has included: 0.01 M $CaCl_2$ for Cd and Zn (Nakhone and Young, 1993); 0.05 M $CaCl_2$ for Zn (Tiller et al., 1972a); 0.01 M $Ca(NO_3)_2$ with 3×10^{-5} M EDTA for Cd and Zn (Fujii and Corey, 1986); 0.1 M $Sr(NO_3)_2$ for Cd (Ahnstrom and Parker 2001); 0.1 M $Ca(NO_3)_2$ and $CaCl_2$ for Cd (Young et al., 2000) and water (Lombi et al., 2003); more recently, 0.01 M $Ca(NO_3)_2$ was used for stable isotopes in measuring E-values for Cr, Cu, Mo, Ni, Ti, Zn, Cd and Pb (Marzouk et al., 2013; Gabler et al., 2007). The advantage of using neutral salts over deionised water is the target metal concentration will be increased in the suspension as a result of displacement from adsorption sites by cation exchange. In the case of $CaCl_2$ metals which form complexes with chloride, such as Cd, are also solubilised by this mechanism whereas adsorption surfaces comprising compounds of Ca, Fe, Mn and Al remain intact. Increasing solubility is important for metals in soils with very low solubility if the solution concentration is close to the detection limit of analytical instruments. Hamon et al., (2008) also suggested that divalent cation salts may help flocculate colloids from suspension which facilitates filtration of the samples and diminishes the potential for colloidal interferences. For soil samples with very low metal solubility, chelating reagents, such as dilute EDTA solution, may be used to bring a greater proportion of the soil metal into solution, providing metal ions are not removed from the non-labile fraction. Atkinson et al. (2011) compared Pb E-values measured in 0.01 M $Ca(NO_3)_2$ and different concentrations of EDTA and found that dilute EDTA ($<5\times10^{-4}$ M) can be used as to increase the concentration of metal in solution without apparently mobilizing the non-labile pool. The solid: solution ratio used to measure E-values has also been tested; Young et al. (2000) found no substantial difference for ratios ranging from 0.02 to 0.08 g mL^{-1}.

Two equilibrium times must be considered in ID experiments: pre-equilibrium time (before spiking isotopes) and post-equilibrium time (after spiking). Prior to isotopic labelling, pre-establishment of equilibrium between soil and an electrolyte may improve the reliability of measured E-values. In previous studies, pre-equilibrium times used have ranged from 30 min to 5 day (Fujii and Corey, 1986; Smolders et al., 1999; Young et al., 2000). Longer pre-equilibration times increase the possibility of redox changes (specifically reduction), in the absence of sample aeration (Young et al., 2007). Reduction reactions normally raise the pH and cause Fe^{III} and Mn^{IV} dissolution which are likely to decrease and increase the E-values of cationic trace elements respectively. Gerard et al. (2000) suggested that the pre-equilibration period can be relatively short as the establishment of ‘equilibrium’ is reported to be fairly rapid. After spiking, the duration of the equilibrium time should be long enough for the spiked isotopes to dynamically equilibrate with the labile metal pool in the soil suspension. However, extending the post-spike equilibration period increases the chances of fixation in which the spike isotopes move into non-labile fraction by, for example, solid phase diffusion or other time-dependent processes (Young et al., 2007). It was suggested by Hamon et al., (2008) that a post-equilibration time of 1 to 3 days is sufficient for isotopic equilibrium to be achieved. Similarly, Young et al., (2000) found very little change in Cd E-values after 2 days of isotopic equilibration time.

Chelex resin purification to avoid inclusion of SCP-metal in E-values:

Overestimation of E-values may occur when there is SCP-metal which passes through standard 0.2 µm filters and contains metal with which the spike isotope has not fully mixed. Hamon and Mclaughlin (2002) found that P availability was overestimated in 6 out of 8 soils due to the existence of non-labile P in a colloidal phase. Other studies have involved trace metals, including Ni, Cu, Zn and Pb. Lombi et al. (2003) and Ma et al. (2006b) reported a significant proportion of non-labile Cu and Zn in SCP phases under alkaline conditions when soil was amended with lime, beringite or red mud. Nolan et al. (2009) studied isotopically exchangeable Ni in the soil solution and found there was detectable non-labile Ni in water extracts (<9%). In these studies, an extra step of 'resin purification' was used to sample the labile trace metal ions in the soil solution and thereby determine the correct isotopic abundance of the spike isotope in the soil labile pool. An ion-exchange chelex resin was introduced into separated supernatant solutions following normal phase separation (typically centrifugation and sub-micron filtration); the resin was then washed and eluted with acid to determine the isotopic abundance of the spike isotope and hence E-value of the soil. The method is based on the assumptions that (i) the resin is not strong enough to extract non-labile metal from SCP forms; (ii) the resin can be cleaned of all colloidal material prior to isotopic assay; (iii) nonlabile metal is not dissolved through the presence of the resin (Young et al., 2007).

Comparison of ID and soil extractants to measure labile metal:

One advantage of the ID method over the other approaches, such as single extraction by nitric acid or EDTA, is that it causes minimum disturbance to solid phases (Young et al., 2005). In some circumstances, such as relatively acidic organic soils, extraction of the labile pool with competing metals (Lofts et al. 2001) or with dilute acid (Tipping et al., 2003) may provide a useful estimate of labile metal content. However, it has been reported by several authors that dilute acids extract more metal than the E-values, especially in the case of calcareous soils (Sinaj et al., 2004; Degryse et al., 2009; Marzouk et al., 2013). Nakhone and Young (1993) compared 4 chemical extractants with isotopically exchangeable Cd (Cd_E) measured on 33 soils contaminated by a wide range of sources. They found that the pools of Cd resolved generally followed the sequence: 'total' (HNO_3 digestion) > 0.05 M EDTA > E-value > 0.01 M $CaCl_2$ > 1.0 M KNO_3. More recently, Gabler et al., (2007) compared EDTA-extracted metal and E-values on a large number of unpolluted soils and found good agreement between the two methods for Cd and Zn. However, EDTA extracted non-labile Ni, Cu and Pb (> E-values), while for Cr, Ti and Mo, there were poor correlations between the two methods. The problem for Ti and Mo can be explained by the fact that it is not possible to form stable EDTA complexes during extraction, while in the case of Cr, there may have been unresolved problems with speciation, as Cr^{III} (Cr^{3+}) and Cr^{VI} (CrO_4^{2-}) may co-exist in soil and adding a single form of spike to determine a single 'labile Cr pool' is not meaningful.

1.2.4 Fractionation using size exclusion chromatograph (SEC)

Size based fractionation techniques, interfaced with a multi-element specific detector such as ICP-MS, can be applied to study the trace metal distribution on particles with < 0.2 µm diameter in soil solution (Jackson et al., 2005). Size exclusion chromatography is applicable to the separation of dissolved macromolecules over molecular weight (MW) ranges of ca. 200 – 100,000 Da. The size exclusion column is tightly packed with porous polymer gels (stationary phase) designed to have pores of different sizes. Samples are injected into an eluent stream (the mobile phase) at the top of the column. Colloids and molecules that are larger than the pores of the gel phase will be eluted first, whilst small molecules are likely to access to all the pores and retention time is longer. Recently, SEC coupled with inductively coupled plasma mass spectrometry (ICP-MS) has been applied to the study of trace element distribution within dissolved organic matter and isolated HAs (Jackson et al., 2005; Kozai et al., 2013; Liu and Cai, 2010; Liu et al., 2011; Pelekani et al., 1999; Schmitt et al., 2001).

Ideally, in SEC columns, analyte species should not interact with the gel material, such that the separation is based solely on size. However, in practice this is rarely the case as charge repulsion or attraction occurs between the analyte and gel phase and such interaction leads to variation in retention time (Chin and Gschwend, 1991; Jackson et al., 2005; Pelekani et al., 1999). This problem has been recognized when studying dissolved organic matter (DOM) in isolated soil solutions and is thought to arise because of their mixed polyelectrolytic and hydrophobic nature (Jackson et al., 2005; Laing, 2010b; Asakawa et al., 2011; Asakawa et al., 2008). It was concluded by Collins (2004) that the process of separation is actually a combination of size-based fractionation, ion exclusion and ion exchange effects. Attempts have been made to minimize specific interactions between HAs and SEC column gel phases by testing different buffer solutions as mobile phases. Many studies have adopted pH buffers such as TRIS (tris(hydroxymethyl)aminomethane; pKa 8.07), and phosphate solutions (pKa_2 7.2) or neutral salts such as ammonium nitrate. Asakawa et al. (2008) used sodium phosphate buffer (pH 7.0)/acetonitrile (3:1, v/v) and avoided non-size-exclusion effects, but the recovery of the HA under investigation was incomplete, implying irreversible sorption within the column. A further problem arises from the possible dissociation of organic complexed metal through interaction with both the gel and mobile phases. In general, only metal complexes whose dissociation rate is insignificant during the separation process are suitable for this form of speciation analysis. Recent studies on dissolved organic matter (DOM) produced variable results using SEC fractionation. For example, Jackson et al. (2005) used SEC for fractionation of Ni and U in natural waters. A low recovery (1.6-18.3% of total) was obtained for Ni suggesting labile complex formation with DOM; recovery was higher for U (11.9 -74.9%); Laborda et al. (2009) used 0.1 M ammonium nitrate at pH 7.4 as an eluent and achieved >93% recovery of Cu, Ni and Co complexed by dissolved organic matter from compost. Schmitt et al. (2001) fractionated DOM in lake water by SEC-ICP-MS and achieved good recovery for Al, Fe and Ni but only 40% recovery for Pb.

The intensity of specific HA-gel interactions probably depends on the chemical properties of the humic substances, such as functional group composition and aromaticity (Asakawa et al., 2008; Asakawa et al., 2011; Pelekani et al., 1999). Humic substances isolated from different sources have different chemical and structural properties which present difficulties when comparing SEC elution patterns and metal fractionation. In all cases, perhaps the greatest problem lies with the incomplete recovery of analyte metals and dissolved humus substances reported in the literature

(Jackson et al., 2005; Schmitt et al., 2001; Asakawa et al., 2011; Asakawa et al., 2008). Given the heterogeneous nature of HAs and FAs this amounts to a further form of fractionation which is effectively excluded from the audit of metal species determined by analysis of the column eluent.

1.2.5 Geochemical speciation model of WHAM-VI & VII

Sophisticated geochemical ‘assemblage’ models have been developed, to resolve chemical speciation in the environment, which can describe adsorption of elements to mineral and humic surfaces. Commonly used models include the extended version of WHAM (Tipping, 1994 & 1998), the NICA-Donnan Model (Benedetti et al., 1996), ORCHESTRA (Meeussen, 2003) and Visual Minteq (Gustafsson and Van Schaik, 2003). These models were initially developed for the aqueous environment but are increasingly used to describe speciation in soil pore water and fractionation within the soil solid phase. WHAM (Windermere Humic Aqueous Model), in particular, has been applied to the problem of predicting the effects of ‘whole soil’ chemistry on metal speciation and fractionation (Tipping et al., 2003; Shi et al., 2008; Almas et al., 2007; Buekers et al., 2008; Marzouk, 2012).

WHAM has been parameterized from a large number of data sets describing adsorption of metals on individual geocolloids (e.g. synthetic Fe oxides, purified HA and FA etc.) generated over the last few decades. It is a mechanistically based equilibrium model which effectively partitions the free energy of metal-geocolloid interaction into ‘chemical bonding’ and ‘electrostatic’ components (Tipping, 1994). It is internally consistent with respect to mass and charge balances and also incorporates speciation code to describe the formation of simple inorganic complexes in solution.

The current versions of WHAM (VI and VII) are both a combination of three sub-models. Metal binding to humic substances (HA and FA) is modelled by ‘Humic Ion Binding Model VI’ (or VII). In this (sub)model, HAs and FAs are assumed to be rigid hydrophobic spheres of uniform size with hydrophilic ion-binding groups on the surface. Two classes of discrete binding site (strong and weak acids) are assumed; these may be notionally identified as carboxyl and phenolic hydroxyl groups. Each ‘class’ of site has four different ‘types’ of group present in equal amounts, which differ in their ‘intrinsic’ pKa value (the pKa that applies with a macromolecular surface charge of zero). The model takes full account of ion competition for binding sites and different binding strength caused by formation of bidentate and tridentate complexes with metal ions (Tipping, 1998). It is assumed that the intrinsic stability constants for metal-proton exchange on humic ligands ($\log K_{MAH}$) for 1:1, 1:2 and 1:3 metal-ligand complexes follow a rigid progression in which $\log K_{MAH}$ for a 1:1 complex is simply multiplied by the stoichiometric value (1, 2 or 3) to derive the range of values for all three forms of complex. However, there is also allowance for a small proportion of very strong complexes where an additional factor is applied.

In the extended (assemblage) version of WHAM, the ‘SCAMP’ model is used to predict metal binding to mineral (hydr)oxides in the soils (Lofts and Tipping, 1998) and a simple cation exchange model, based on Donnan principles, is used to describe purely electrostatic binding of cations to aluminosilicate clays. Reactions between solutes and cations in soil pore water are also considered in the model and include cation complex formation with inorganic anions, FAs and HAs. The humus acids (HA and FA) and other geocolloids (Fe, Al, Mn (hydr)oxides and clay) are designated ‘particulate’ (flocculated in the solid phase) or ‘colloidal’ (in suspension, or dissolved, in the solution phase).

The main difference between the assemblage models WHAM-VI and VII lies in improvements to Model VI to produce Model VII. Tipping (2011) described Model VII as a simpler model than Model VI in its postulated multidentate metal binding sites, but one that incorporates more metals. It differs from Model VI with respect to metal cation binding, in that the arrangement of multidentate sites has been modified and some parameters have been eliminated. In addition, Model VII provides better descriptions of metal binding at higher pH as a result of (re-)parameterization on more extensive datasets. Although Model VII represents an improvement on Model VI, in practice its predictions do not differ greatly for many elements. A relative assessment has been undertaken for WHAM-VII on aqueous samples (Lofts and Tipping, 2011) and minespoil soils (Marzouk, 2012).

WHAM-VI and VII have been used in a very limited number of studies predicting trace metal solubility in soils. Reasonable predictions of Ni, Zn and Cd solubility have been obtained by both WHAM and alternative geochemical models. However, Cu and Pb solubility are generally poorly predicted (Marzouk, 2012; Buekers et al., 2008b). Overestimation of Cu and Pb solution concentrations (underestimation of solid surface binding) has been observed using default parameters in WHAM for soils with high pH values (Buekers et al., 2008b); more reliable predictions have been obtained for acidic soils and in humus-rich (peaty) soils (Tipping et al., 2003).

The operational requirements of geochemical models such as WHAM expose several weaknesses, all of which will contribute to failures in predicting metal speciation in solution, fractionation in the solid phase and overall solubility.

Defining the overall reactive (labile) metal fraction: A range of approaches have been used to measure the reactive metal pool in soils including, for example, extraction with a range of concentrations of HNO_3 (0.1 M, 0.43 M or 0.22 M) (Smith et al., 2004; Tipping et al., 2003; Almas et al., 2007) and E-values (Buekers et al., 2008b). Clearly a valid estimation of the reactive pool is critical to successful model predictions of solubility etc.; differences between chemical extraction and isotopic dilution have already been discussed (see Sections 1.2.3). In addition, WHAM operates on the basis that trace metals bound to dissolved humic substances are completely labile. However, there is increasing evidence of the existence of non-labile metal fractions adsorbed on DOM which implies that the model may lead to an overestimation of the free ion activity in soil solution if based on total concentration inputs in aqueous phases (soil pore water or stream water etc.).

Reactive surface area of geocolloids: WHAM does not take account of possible differences in surface morphology between pedogenic geocolloids and their synthesised counterparts from which WHAM was originally parameterised. For example, Buekers et al. (2008b) argued that the surface area of Fe-oxyhydoxides may be larger in soil than that of synthesized oxyhydroxides because of isomorphic impurities within the oxyhydroxide structure. They achieved better model fits to metal solubility data by increasing the assumed surface area of Fe oxides within the WHAM model.

Mixed adsorption phases: Current geochemical models do not consider possible interactions between binding phases, such as surface adsorption of HA or FA on mineral (hydr)oxides. Such assemblages undoubtedly exist in soils and the overall effects on surface charge and on availability of adsorption sites may be significant.

For example, Antelo et al. (2009) found enhancement in copper adsorption in the gibbsite/HA/Cu^{2+} ternary system compared to the corresponding binary systems.

Estimation of reactive geocolloid content in soils: WHAM requires an estimation of the concentration of all binding phases (Fe, Al, Mn (hydr)oxides, HA, FA and ‘generic’ aluminosilicate clay). However, the methods used to measure active Fe/Mn-(hydr)oxides vary among studies and include approaches such as oxalate and pyrophosphate extraction (Almas et al., 2007) or dithionite extraction (Buekers et al., 2008b). Similarly, there is no standard approach to determine the amount of active humic substances (particulate and colloidal HA and FA) in soils and various assumptions are used in practice. Concentrations of HA and FA, both in the solid (particulate) or solution (colloidal) phases, are often estimated from the total organic carbon content in soil or in soil solution. Weng et al. (2002) assessed two previous assumptions applied to the solution phase, in which DOM was assumed to comprise either (i) 30% active HA, 30% active FA and 40% inert carbon compounds or (ii) 65% FA and 35% inert material. For the solid phase, they assumed that humus had a carbon content of 50% and 50% of humus was active HA. Buekers et al. (2008) and Cheng et al. (2005) assumed that 50% of DOM is organic carbon, and 65% of this was active FA.

Inclusion of all possible binding phases: The current version of WHAM identifies a range of geocolloidal binding phases (listed above) but this is not comprehensive. For example it has been shown that hydroxy-interlayered clays are active in adsorbing metals at low pH values (Degryse et al., 2011) while sorption of metals on calcite surfaces has been widely studied (Ahmed et al., 2006).

The need for these assumptions must limit the accuracy of any geochemical model applied to whole soil metal fractionation where the model has been parameterised using data from single, ‘purified’ geocolloidal studies. There is clearly a need to address such limitations if models such as WHAM are to gain wider acceptance for predicting solubility, speciation and fractionation of trace metals in soil and become accepted in wider applications such as risk assessment.

1.3 Objectives and thesis structure

The primary aim of this work was to use stable isotopes to determine the lability of Ni, Cu, Zn, Cd and Pb in a wide range of soils and to i) predict the trace metal solubility in soil from the measured lability using the geochemical speciation models WHAM VI and VII; ii) study the effect of soil properties and sources of contamination on metal lability and iii) measure the lability of Cu, Zn and Pb on soil extracted humic acid.

Chapter 2 describes general materials and methods including measurement of soil properties and trace metal lability using different techniques. The development of a method incorporating SEC with ID method for measuring metal lability on humic acid is also presented.

Chapter 3 investigates the lability of Ni, Cu, Zn, Cd and Pb in two sets of soils: metal-amended (MA) soils and Urban soils from Nottingham and Wolverhampton (Urban). Metal lability was measured using isotopic dilution with or without chelex resin purification step (Section 1.2.3) and using 0.43 M HNO_3 extraction (Section 1.2.1). The measured labile fraction using different methods is compared as a function of soil properties. Two types of empirical model were then developed based on the set of metal amended soils and assessed for their ability to describe and predict metal lability (isotopic exchangeable fraction) from soil properties for both sets of soils. The results were used to investigate both the influence of soil properties and sources of contamination on metal lability. In addition, the lability of Zn and Cd was also compared with values of isotopic exchangeable fraction measured by radio-isotopic dilution 10 years previously to investigate the possibility of longer-term fixation.

In Chapter 4, the use of both empirical and mechanistic geochemical models (WHAM-VI and VII) were assessed by predicting Ni, Cu, Zn, Cd and Pb solubility in the metal-amended soils and Urban soils used in Chapter 3. The WHAM model requires an estimate of the total concentration of 'reactive metal' in soils, across all adsorbed phases. Therefore E-values (M_E) were used to represent labile metal and compared with 0.43 M HNO_3-extractable metal (M_{Ext}), which has both been measured and discussed in Chapter 3. To investigate the origins of errors and bias in the prediction of metal solubility the modelled fractionation of trace metals in the solid phase was also investigated. Attempts were made to optimize the phase parameters to improve model prediction. Finally, comparison was made between the results from WHAM-VI and WHAM-VII to assess the value of recent attempts to improve the model.

The lability of Pb in soil is further investigated in Chapter 5, as its isotopic ratio can be used to determine the origin of contamination. The lability of Pb determined by ID and isotopic ratio were measured in three sets of soils: rural roadside, sewage farm and urban soils from London. Each group of soils has a specific source of Pb input in addition to Pb from parent material. Sequential extraction was also applied to a sub set of these soils to investigate how different fractions may contribute to the lability and a binary model was used to estimate source affected Pb lability.

Chapter 6 represents work to quantify trace metal lability on humic acids. Dissolved humic acids were incubated with Cu, Zn and Pb and spiked with stable isotopes before fractionation and analysis using SEC-ICP-MS. The kinetics of isotopic exchange on humic acid molecules is presented and the lability of Cu, Zn and Pb calculated. The labile metal pool of metal on humic acids was also determined by single extractions with EDTA at a range of concentrations.

Finally, overall conclusions are presented in Chapter 7 alongside recommendations for future investigations.

2 CHAPTER 2: MATERIALS AND METHODS

Overview

This chapter presents general materials and methods used for soil characterization including determination of soil pH, organic matter content, mineral oxides content, clay content and major and trace metal concentrations. For comparison, some soil properties were measured by several methods. The chapter also describes the three forms of multi-element stable isotope dilution (ID) developed to determine (i) labile Ni, Cu, Zn, Cd and Pb in soils (E-values), (ii) the presence of fixed metal in suspended colloidal particles (SCP-metal) and (iii) the change in lability of Cu, Zn and Pb adsorbed by humic acids during equilibration using size exclusion chromatography (SEC) coupled with ICP-MS. General information of all soil and humic acid samples such as sampling and storage is summarized in Table 2-1. Detail of samples collection and preparation are provided in relevant chapters and sections below.

2.1 Soil characterization

2.1.1 Soil pH

Soil pH was measured on suspensions of air-dried soil (< 2 mm) in 0.01 M $CaCl_2$, with a soil: solution ratio of 1:2.5, shaken in Oak Ridge polycarbonate centrifuge tubes on an end-over-end shaker for 30 minutes. A pH meter and combined glass electrode (Ag/AgCl) (Model pH 209, HANNA Instruments, Bedford, UK), calibrated using buffers at pH 4.01 and 7.00 was used to measure the soil suspension pH. Soil pH was also measured in the 0.01 M $Ca(NO_3)_2$ suspensions used for ID measurements with a soil: solution ratio of 1:30.

Table 2-1: Summary of general information of all soil/humic acid samples

	Samples	No. of soils	Sampling place	Storage	Detail	References
	Soils					
Chpt. 3 & 4	Metal Amended (MA)	23	UK and Europe	natural soils incubated with metal salts for 2.5 years and stored for more than 10 years	section 3.2.1	Tye et al., 2003, 2004 Hough et al., 2005
	Urban	50	Nottingham and Wolverhampton	urban soils stored for more than 10 years		Hough et al., 2005 Thornton et al., 2008
Chpt. 5	Rural Roadside	42	Main roads in Nottinghamshire	N/A	section 5.2.1	N/A
	Sewage Farm	16	Sewage farm in Nottinghamshire	N/A		N/A
	London	50	Great London	'London Earth' archive from BGS		Johnson et al., 2005
	Humic Acid					
Chpt. 6	Peat humic acid		Extracted from Sphagnum moss peat	Stored dry in glassware for 23 years	section 6.2.1	Marshall et al., 1995
	Grassland humic acid		Extracted from grassland soils in University Farm			

2.1.2 Organic matter content

The concentrations of organic matter, humic acid and fulvic acid in soil were measured in three ways.

2.1.2.1 *Loss on ignition*

Loss on ignition (LOI) was used to estimate total organic matter content in soils (Rowell, 1994). Approximately 5 g of air-dried soil (< 2 mm) was ignited in a muffle furnace at 550°C for 7 hr. The LOI (%) was then determined gravimetrically.

2.1.2.2 *Alkaline extraction of humic and fulvic acids*

Humic and fulvic acids were extracted from sieved air-dried soil samples (< 4 mm) in 0.1 M NaOH using a solid:solution ratio of 1 g in 15 ml. The samples were shaken for 24 hours before separation of the supernatant solution, containing HA and FA, by centrifugation at 2200 g and filtration (< 0.22 μm). To separate HA and FA, 1 ml of 1.5 M HNO_3 was added to 10 ml of the filtered alkaline soil extract to lower the pH to < 2.0 and cause flocculation of HA. Total carbon content in the alkaline (HA + FA) and acidified (FA only) extractants were measured using a Shimadzu TOC-Vcp analyser and the total concentrations of HA and FA in the soils were calculated based on the assumption that the carbon content of both HA and FA is ~50% (Weng et al., 2002). It was considered possible that HA and FA may not be fully extracted by a single extraction on 2 mm sieved soils in peaty soils (Dr Steve Lofts (CEH), personal communication). Therefore, repeated (triple) extractions of soil in 0.1 M NaOH were undertaken, before HA and FA separation, for 6 selected soil samples with TOC contents ranging from high (31.3 g kg^{-1}) to low (2.2 g kg^{-1}). With one exception, there was no significant difference in the efficiency of single and multiple extractions (Fig. 2-1). However, the single soil which appeared to require repeated extractions to recover HA and FA showed clear visual evidence of coal particles which may explain the apparently slower solubilisation of organic matter. It was therefore decided to adopt a single extraction with 0.1 M NaOH to measure total HA and FA in all soils.

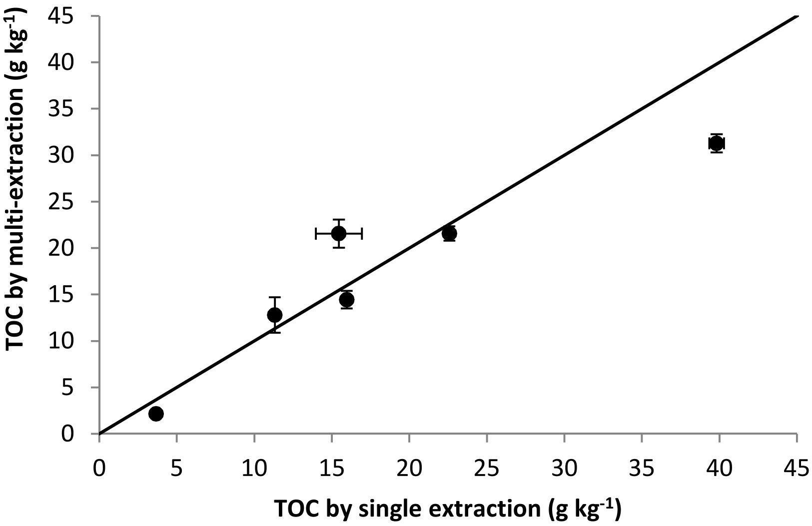


Figure 2-1: Comparison of total alkali-soluble organic carbon content (g kg^{-1}) in 6 selected soils (< 2 mm) using single (X) and multiple (Y) alkaline extractions. The solid line is the 1:1 relation; error bars represent the standard error of duplicates.

2.1.3 Soil particle size analysis by laser granulometry

Soil samples (< 2 mm) were gently crushed with a pestle and mortar to break up aggregates. Soil organic matter was oxidised by suspending 0.5 g soil in 25 mL of 30% H_2O_2 overnight then heating to 60°C for 1.5 hours and then 80°C for 2 hours. The sample was then washed twice in deionized water. A dispersing agent ‘Calgon’ (25 mL of 0.057 M of sodium hexametaphosphate in 0.66 M of sodium carbonate) was added to each sample and suspensions were homogenised in an ultrasonic bath. All samples were analysed by Beckham Coulter LS 230 particle size analyzer in the Department of Geography, the University of Nottingham.

2.1.4 Mineral oxides

Iron, Al and Mn oxides were determined by extraction with a mixture of sodium dithionite, sodium citrate and sodium bicarbonate (DCB extraction; modified from Anschutz et al. 1998). Sodium dithionite (0.07 M) was used to reduce Fe^{III} to Fe^{II} and 0.3 M sodium citrate to buffer pH, prevent decomposition of dithionite to hydrogen sulphite, and complex Fe^{2+} ions; 1 M of sodium bicarbonate was used to elevate the pH. Sample tubes were left in a shaking water bath at 20°C for 24 hours with lids left slightly open to let gas escape but retain water vapour by condensation. After centrifugation, concentrations of Fe, Al and Mn were determined by ICP-MS (Section 2.2.6.1).

2.1.5 Dissolved organic and inorganic carbon in solution

Soil was suspended in 0.01 M of $Ca(NO_3)_2$ (1 g: 30 mL) and equilibrated on an end-to-end shaker for 3 days. Following centrifugation (2200 g) and filtration (< 0.22 µm), the dissolved organic/inorganic carbon content was measured using a Shimadzu TOC-Vcp analyser. For estimation of FA concentration in solution it was assumed that dissolved organic matter contains 50% C and that 65% of this organic matter consists of active fulvic acids (Buekers et al., 2008b; Cheng et al., 2005).

2.2 Major and trace cations in soil

2.2.1 Exchangeable major cation concentration in soil

Concentrations of exchangeable Na, Mg, K and Ca were measured on soils (<4 mm). Samples were suspended in 1 M of NH_4NO_3 (1g: 5 mL) for 30 minutes (Rowell, 1994). Following centrifugation (2200 g) for 30 minutes and filtration (< 0.22 µm), the supernatant solutions was diluted 1-in-10 with 2% nitric acid before analysis by ICP-MS (Section 2.2.6.1).

2.2.2 Total trace metals in soil by acid digestion

Approximately 200 mg samples of finely ground soil were digested in PFA vials within a 48-place Teflon-coated graphite block digester (Model A3, *Analysco* Ltd, Chipping Norton, UK) with 2 mL of concentrated HNO_3 (69% AR) and 1 mL of $HClO_4$ at 80°C for 8 hr and then at 100°C for a further 2 hr. An aliquot (2.5 mL) of HF (40% AR) was then added and the samples were heated to 120°C for 8 hr. A further 2.5 mL of HNO_3 and 2.5 mL MilliQ water were then added to the dry residue and the vessels heated at 50°C for 30 min. The digested soil samples were kept in a 5% HNO_3 matrix and the total concentrations of Ni, Cu, Zn, Cd and Pb determined by ICP-MS (Section 2.2.6.1).

2.2.3 Sequential extraction procedure

The sequential extraction procedure used in this study (Table 2-2), modified from Tessier et al. (1979), is described in Li and Thornton (2001). The concentration of trace metals in the residual fraction was determined by difference between the total trace metal content (Section 2.2.2.1), measured independently, and the summation of F1 to F4 of the SEP.

Table 2-2: Summary of the modified Li and Thornton (2001) sequential extraction procedure (SEP)

Fraction	**Extraction reagents**	**Extraction time**	**Soil:solution ratio (g : mL)**
F1: exchangeable	0.5 M $MgCl_2$	0.33 h	1 : 8
F2: carbonate	1 M NaOAc* (adjusted to pH 5 with HOAc)*	5 h	1 : 8
F3: Fe/Mn oxide	0.04 M $NH_2OH.HCl$	6 h at 96°C	1 : 20
F4: OM and sulphide	0.02 M HNO_3 and H_2O_2, 3.2 M NH_4OAc*	5 h at 85°C; 0.5 h at 20°C	1 : 16
F5: residual	HNO_3, HF, $HClO_4$	19 h	1:12.5

*OAc is acetate

2.2.4 Single extraction of trace metals with 0.43 M HNO_3

The method of single extraction by dilute HNO_3 was adapted from Tipping et al. (2003). Soil samples (1.0 g; < 2 mm) were extracted in 0.43 M HNO_3 (10 mL) by shaking for 2 hr. Suspensions were then centrifuged (2200 g) and the supernatant filtered (< 0.2 μm) prior to analysis by ICP-MS (Section 2.2.6.1).

2.2.5 Concentration of major and trace cations in equilibrated 'soil solutions'.

Dissolved major and trace cation concentrations were measured in 0.01 M $Ca(NO_3)_2$ solutions (30 mL) equilibrated with soil (1.0 g; < 2 mm). The soil suspensions were equilibrated on an end-over-end shaker for 3 days as the pre-equilibrium time used for measuring E-values (mg kg^{-1}) (Section 2.3.2.1). After centrifugation (2200 g) and filtration (< 0.2 μm) the supernatant was acidified to 2% HNO_3 before analysis of major cations (Na, Mg, Ca and K) and trace metals (Ni, Cu, Zn, Cd and Pb) by ICP-MS (Section 2.2.6.1).

2.2.6 Chemical analysis

2.2.6.1 ICP-MS settings for multi-element analysis

Major and trace cation concentration extracted from soil were determined using ICP-MS (Thermo-Fisher Scientific X-Series^{II}) with a 'hexapole collision cell' (7% hydrogen in helium) to reduce polyatomic interferences. Samples were introduced by an autosampler (Cetac ASX-520 with 4 × 60-place sample racks) through a concentric glass venturi nebuliser (Thermo-Fisher Scientific; 1 mL min^{-1}). Internal standards were introduced to the sample stream via a T-piece, including Sc (100 µg L^{-1}), Ge (50 µg L^{-1}), Rh (20 µg L^{-1}) and Ir (10 µg L^{-1}) in 2% HNO_3. Two sets of external multi-element calibration standards were used for both major elements (concentrations ranged from 0 – 30 mg L^{-1}) and trace elements (concentrations ranged from 0 – 100 µg L^{-1}). Sample processing was undertaken using Plasmalab software with internal cross-calibration between pulse-counting and analogue detector modes where required. All elemental concentrations were converted to mg kg^{-1} for results and discussion.

2.2.6.2 Use of size exclusion chromatograph

Fractionation of humic acid (HA) based on molecular size was made by using High Performance Liquid Chromatography (HPLC; Dionex ICS-3000 chromatography system) coupled to a size exclusion column (SEC; GE Healthcare, Superose 12 10/300 GL). The SEC column comprises with a stationary phase of cross-linked agarose matrix with a separation range from 1000 to 300,000 Da (globular proteins). A guard column (Hamilton, Peek PRP-X100) was used prior to the SEC column to avoid SEC column blockage. The mobile phase was an eluent of 0.1 M of Tris buffer adjusted to pH 8.2 with nitric acid. Humic acid samples of 100 µl were injected with an auto sampler triggered by Chromeleon ® software at a flow rate of 1 ml min^{-1}. The column outflow was connected directly to the nebuliser of ICP-MS for isotopic analysis, with instrument settings as described in Table 2-3 (Section 2.3.1).

2.3 Measuring isotopically exchangeable Ni, Cu, Zn, Cd and Pb in soils

2.3.1 Stock isotope solution

Enriched stable isotopes of ^{62}Ni (IA ≥ 98.2%), ^{65}Cu (IA ≥ 99.0%), ^{70}Zn (IA ≥ 92.7%), ^{108}Cd (IA ≥ 69.7%) and ^{204}Pb (IA ≥ 98.8%) were purchased from ISOFLEX, San Francisco CA, USA, as metal foil samples. The foils were dissolved in concentrated HNO_3 (69%) and diluted with MilliQ water to 5% HNO_3 for preservation. The concentrations of the enriched isotope stock solutions were calculated from the reported sensitivity (counts per second per µmol^{-1} L^{-1}; CPS ppb^{-1}) measured for a series of isotopes for each element covering a range of atomic masses. To enable this, Ni, Cu, Zn, Cd and Pb (NIST 981) standards were made. The ICP-MS operating conditions are shown in Table 2-3. The five elements were measured separately to avoid too much plasma noise by measuring too many isotopes at the same time. The measured concentration of the stock isotopic spike solutions were: ^{62}Ni (603.3 mg L^{-1}), ^{65}Cu (840.5 mg L^{-1}), ^{70}Zn (142.2 mg L^{-1}), ^{108}Cd (122.5 mg L^{-1}) and ^{204}Pb (441.8 mg L^{-1}).

Table 2-3: ICP-MS operating and data acquisition parameters for multi-element analysis when determining the concentration of enriched isotope stock solutions.

Parameter	Value and/or description
ICP-MS instrument	Thermo-Fisher Scientific X-Series^{II} with a 'hexapole collision cell' (7% hydrogen in helium) upstream of the analytical quadrupole.
Nebuliser	Concentric glass venturi - Thermo-Fisher Scientific; 1 mL min^{-1}
Quadrupole parameters	Minimum and maximum settle times 1000 µs; safe resting mass: 205 amu
Autosampler	Cetac ASX-520
Forward power	1400 Watts
Reflected power	< 5 Watts
Data acquisition	
Dwell time for isotope ratio measurements	Soil supernatants: 10 ms for ^{62}Ni, ^{70}Zn, ^{108}Cd, ^{202}Hg and ^{204}Pb; 0.1 ms for ^{77}Ar Cl, ^{82}Se and ^{83}Kr; 2.5 mS for the other isotopes Humic acid samples: 40 ms for ^{63}Cu, ^{65}Cu, ^{66}Zn, ^{70}Zn, ^{204}Pb, ^{206}Pb, ^{207}Pb and ^{208}Pb
Detector dead time	35 ns
Scanning mode	Pulse counting
Measuring mode	CCT-KED mode
External standards (soil supernatants only)	NIST SRM 981 Pb and Certiprep calibration standard solutions of Ni, Cu, Zn and Cd in 2%HNO_3
Software	Plasmalab (version 2.5.4; Thermo-Fisher Scientific)

2.3.2 Measuring isotopically exchangeable Ni, Cu, Zn, Cd and Pb in soil

2.3.2.1 Experiment procedure

The ID method used in this study was adapted from Atkinson et al. (2011). Soils were pre-equilibrated in 0.01 M $Ca(NO_3)_2$ (1 g: 30 mL) on an end-over-end shaker for 3 days; 4 suspensions were made for each soil. An aliquot of isotopic spike solution (0.4 mL), enriched with ^{62}Ni, ^{65}Cu, ^{70}Zn, ^{108}Cd and ^{204}Pb, was added into two of the suspensions and the other two were left as controls. As the isotopes were kept in a 2%

nitric acid matrix, the addition of the acidified spike solutions may cause a decrease in pH. Therefore, the volume of the spike solution was as small as possible to minimize the possible mobilization of non-labile metal from the soil. Unacidified spike solutions are not recommended because trace levels of metal ions may adhere to containers. Gabler et al. (2007) suggested that the decrease in soil suspension pH caused by adding a spike could be minimized by separately determining the E-values of individual elements. However, in this experiment, the multi-element spike solution was diluted immediately before adding to the samples, to limit the decline in pH and enable simultaneous determination of all five elements. The spike isotopes used were chosen because they have relatively low natural isotopic abundance (Table 2-4). Therefore, only a small addition to the system was required to produce a significant increase in IA from the background level. To reduce the number of individual operations and to avoid adding different amount of isotopes each soil, the samples were classified into several groups according to their total metal content. The level of spike concentration was determined by the highest concentration of metal in each group to ensure there was a significant difference in isotopic ratio between the spiked and unspiked samples (> 20 % increase). After spiking, the suspensions were shaken for a further 3 days to attain isotopic equilibrium. The solution and solid phases were then separated by centrifugation (2200 g) and filtration (< 0.2 μm) and the isotopic ratios (^{62}Ni/^{60}Ni, ^{65}Cu/^{63}Cu, ^{70}Zn/^{66}Zn, ^{108}Cd/^{111}Cd, ^{204}Pb/^{206}Pb, ^{204}Pb/^{207}Pb, and ^{204}Pb/^{208}Pb) in the supernatant solution were measured by ICP-MS in ‘collision cell with kinetic energy discrimination’ (CCT-KED) mode to avoid interference from the chlorine dimer (^{35}Cl-^{35}Cl) on ^{70}Zn (Malinovsky et al., 2005; Stenberg et al., 2004). It is well known that the relative abundance of Pb isotopes vary according to the sources of Pb present in the soil. Therefore, instead of relying on relative abundances of naturally occurring isotopes, the isotopic ratio in unspiked samples (blanks) were also determined for all five elements. To avoid the instrument tripping to analogue detector mode, all the sample solutions were diluted to less than 100 µg L^{-1} ^{208}Pb, to produce ICP signals for ^{208}Pb of < 2×10^{6} CPS. Details of ICP-MS operating conditions are listed in Table 2-3.

Table 2-4: Characteristics of the spike isotopes (Debiever and Barnes, 1985)

Spike Isotope	Natural Isotopic Abundance (%)	Enriched Isotopic abundance (%)	Isotope ratio used for the calculations	Natural Isotopic Ratio
^{62}Ni	3.59	98.2	^{62}Ni:^{60}Ni	0.138
^{65}Cu	30.8	99.0	^{65}Cu:^{63}Cu	0.445
^{70}Zn	0.60	92.7	^{70}Zn:^{66}Zn	0.022
^{108}Cd	0.9	69.7	^{108}Cd:^{111}Cd	0.070
^{204}Pb	1.4	98.8	^{204}Pb:^{208}Pb	0.027

2.3.2.2 Calculation of E-values

A potential source of error when analysing stable isotopes using ICP-MS is mass discrimination, or mass bias. Isotopes with greater mass have a greater sensitivity and so the ratio of measured ICP-MS signals (CPS) for two isotopes is not equal to their true isotopic ratio. External mass discrimination correction was therefore applied using the certified isotopic standard reference material (NIST, SRM 981, Pb) for Pb and a mix of single ICP-MS calibration standards for Ni, Cu, Zn and Cd. The certified isotopic reference is required for Pb because of the variability in Pb isotope ratios arising from their radiogenic origin in natural environment. By contrast, the assumption behind using single calibration standards to correct mass bias for Ni, Cu, Cd and Zn is that a fixed ratio of isotopes applies universally (Debievre and Barnes, 1985). The mixed standard (25 µg L^{-1}) was used as a running correction factor during analysis and measured after each set of 8 samples.

A correction factor (K-Factor) was calculated (Eq. 2-1), where the K-Factor is the correction factor for mass bias, IR and CR are the true isotopic ratio and the measured count ratio for the isotopic standard. For individual samples, the K-factors for each isotopic ratio (eg $^{204}\mathrm{Pb}/^{208}\mathrm{Pb}$, $^{206}\mathrm{Pb}/^{208}\mathrm{Pb}$, $^{207}\mathrm{Pb}/^{208}\mathrm{Pb}$ etc) were estimated by linear extrapolation between the K-factors measured for standards at intervals of 8 samples.

$$\text{K-Factor}=\frac{\mathrm{IR}}{\mathrm{CR}} \qquad (2\text{-}1)$$

The isotopic abundance was then calculated from Eq. 2-1 and the concentrations of isotopically exchangeable Ni, Cu, Zn, Cd and Pb (E-value, mg kg^{-1}) were calculated from Eq. 2-2.

$$M_E=\left(\frac{Am_{Msoil}}{W}\right)\left(\frac{C_{spk}\ V_{spk}}{Am_{Mspk}}\right)\frac{\left({}^{spk}IA_{spike}-{}^{bg}IA_{spk}\ R_{ss}\right)}{\left({}^{bg}IA_{soil}R_{ss}-{}^{spk}IA_{soil}\right)} \qquad (2\text{-}2)$$

where Am_{Msoil} and Am_{Mspk} is the average atomic mass of Ni, Cu, Zn, Cd and Pb in soils and spikes respectively, W is the weight of the soil (kg), C_{spk} is the gravimetric concentration of the metal in the spike solution, V_{spk} is the volume of spike added (L), IA is the isotopic abundance (mole) and R_{ss} is the ratio of isotopic abundances, spiked (spk): background (bg), for the two isotopes in the spiked soil solution.

For comparative purposes, the lability is often expressed as a percentage of the total metal content of the soil (Eq. 2-3):

$$\%M_E=\frac{M_E}{\text{Total metal concentration}}\times 100 \qquad (2\text{-}3)$$

2.3.3 Measuring isotopically exchangeable Ni, Cu, Zn, Cd and Pb in soil with resin purification:

This method used to determine the presence of non-labile metal in suspended colloidal particles (SCP-metal; < 0.2 μm) in soil suspensions was described by Lombi et al. (2003). Analytical grade Na-Chelex-100 resin (Bio-Rad laboratories, UK) was converted to the Ca^{2+} form by equilibrating in 0.5 M $Ca(NO_3)_2$ for 2 hours. The resin was then washed twice with MilliQ water to remove remaining Na^+ and the pH measured (7.9). An aliquot of Ca-Chelex resin (approx. 100 mg) was introduced into 10 mL of filtered (< 0.2 μm) soil solution from both spiked and unspiked suspensions (Section 2.3.2.1). After equilibration for 2 hours, the resin was rinsed with MilliQ water three times to remove colloidal particles. Metals were then eluted from the resin with 0.5 M HNO_3 and the measured isotopic ratio used to calculate the labile pool (E-resin; M_{Er}). Results were then compared with E-values (M_E) measured directly on the filtered solution phase (Section 2.3.2.2) to determine the presence of non-labile SCP-metal.

2.4 Geochemical modelling solubility

The WHAM geochemical models (Versions VI; Tipping et al., 2003 and VII; Lofts and Tipping, 2011) were used to predict Ni, Cu, Zn, Cd and Pb speciation and concentration in the solution phase of soil suspensions (1 g in 30 mL 0.01 M $Ca(NO_3)_2$). The input variables included temperature, soil pH, total suspended particulate material SPM (g L^{-1}), total reactive trace metal concentration (mol L^{-1}); clay content, Al, Fe and Mn oxides, HA and FA (g L^{-1}) in both solid and solution phases, dissolved Na, Mg, Al, K and Ca solution concentration, bicarbonate concentration (mol L^{-1}) and Fe^{3+} and Al^{3+} activities (WHAM-VI only). When alternative input variables were available, the model was assessed using different sets of input variables. Model output includes total metal concentration in solution (mol L^{-1}) and the proportion of fractions associated with soil solid and colloidal phases. Detail of model settings and variables as well as the results are described and discussed in Chapter 4.

2.5 Statistical analysis

Minitab version 16.2.2 was used for various statistical analyse including T-tests, Pearson correlation coefficients and step-wise correlation, the default level of confidence was 95% ($P < 0.05$). Solver in Microsoft Excel was used for optimizing parameters in empirical models. The value of residual standard deviation (RSD) was used to assess the performance of empirical models (Eq. 2-4).

$$\mathrm{RSD}=\sqrt{\frac{1}{(n-c)}\sum_{i=1}^{n}(Mi-Pi)^2} \qquad (2\text{-}4)$$

where c equals to the number of the optimized constants; n is the number of the samples and M_i and P_i are the measured and predicted results respectively.

2.6 Quality control:

Standard reference material® 2711, Montana soil, was used on regular bases for quality assurance of trace element concentration analysis by ICP-MS. Quality assurance of isotope measurement was confirmed by measuring isotopic ratio of ICP-MS single element standards and Pb NIST (981) standards during the run (section 2.3.2.2). Inter-laboratory calibration was carried out by measuring Pb isotopic ratio in selective samples of 0.01 M $Ca(NO_3)_2$ soil suspension in both University of Nottingham (UoN) and British Geological Survey (BGS); the mean ratio of $^{206}Pb/^{207}Pb$ (UoN/BGS) was 1.001 with SD=0.002 (Chapter 5).

3 CHAPTER 3: MEASURING LABILE NICKEL, COPPER, ZINC, CADMIUM AND LEAD IN METAL-AMENDED AND URBAN SOILS

3.1 Overview

The lability of trace metals in soils depends on several factors, related to both soil and contaminant source (Daviescolley et al., 1984; Degryse et al., 2004; Ledin et al., 1996; Tack, 2010). These often include:

(i) intrinsic soil properties such as geo-colloidal adsorption phases;
(ii) variable soil conditions including soil pH and Eh;
(iii) the source, chemical form and deposition route of the metal contamination;
(iv) the contact time between the soil and contaminant.

The primary purpose of the work presented in this chapter was to use stable isotopes to measure the lability of Ni, Cu, Zn, Cd and Pb in soils, and investigate how the lability of these trace metals is affected by the factors listed above. Thus, the lability of Ni, Cu, Zn, Cd and Pb was investigated using two distinct sets of soils. These included (i) soils, chosen for their range of land uses and soil properties, which had been incubated with metal nitrate salts (Ni, Cu, Zn, Cd and Pb) for several years and (ii) urban contaminated soils, from Nottingham and Wolverhampton, chosen for their range of metal contaminant concentrations and sources. Metal lability was measured in three different ways: (i) as isotopically exchangeable metal using an equilibrated electrolyte phase to assay the degree of isotopic dilution of a stable isotopic spike (M_E); (ii) as for (i) but using a chelex resin purification step to avoid contamination from non-labile metal held within suspended colloidal particles (SCP-metal) (M_{Er}); (iii) a single chemical extraction step with 0.43 M HNO_3 (M_{Ext}). Two types of empirical models were then developed and assessed for their ability to describe and predict metal lability (%M_E) from soil properties. Values of M_E and M_{Er} for both sets of soils were used to investigate the influence of soil properties and sources of contamination on metal lability. In addition, comparison between M_E and M_{Er} was used to establish the presence of submicron colloidal particulates (SCP; <0.2 µm)-metal and how this is affected by soil properties. Comparison of M_E and M_{Ext} enabled assessment of a widely used soil metal extractant (0.43 M HNO_3) against the more mechanistically based ID approach. The lability of Zn and Cd in both sets of soils was also compared with values of M_E measured by radio-isotopic dilution more than 10 years ago to investigate the possibility of longer-term fixation.

3.2 Material and Methods

3.2.1 Soil samples

Twenty three soils were sampled, mainly from the UK, from sites with contrasting land uses and parent materials providing a wide range of soil properties including pH, soil organic matter, mineral (hydr)oxides content and texture. These soils were initially collected for use in an earlier study of trace metal fixation and solubility in soils (Tye et al., 2003; Tye et al., 2004). They had been amended with metal nitrate salts to the limits prescribed by the UK Sludge Regulations (MAFF, 1993) (Ni = 75, Cu = 135, Zn = 300, Cd = 3 and Pb = 300 mg kg^{-1}) and incubated at 80% field capacity (FC) and 16°C for nearly 3 years and then stored dry for about ten years before the current study was undertaken. This set of soils was designated as ‘Metal Amended’ (MA).

A second set of 50 surface soils collected in Wolverhampton (WV) and Nottingham (NG), cities which have quite different industrial legacies. Sample site selection was based on the distribution pattern of total soil metal concentrations relating to past and present industrial and domestic land uses including brownfield sites, rail transport sidings, urban roadsides, waste disposal facilities, recreational areas, gardens, urban nature reserves and woodland and parkland areas. Data relating to these soils has also been previously published in an investigation of metal speciation and bioavailability in risk assessment (Thornton et al., 2008). This set of soils is designated as 'Urban' soils in the following discussions.

3.2.2 Soil characterization

A range of soil properties were measured, and in some cases re-measured to compensate for changes during storage, to enable a complete input dataset for WHAM-VII modelling to be compiled (see Chapter 4). These included reactive Al, Fe and Mn oxides, HA and FA following alkaline extraction, soil pH in 0.01 M $Ca(NO_3)_2$, DOC in the solution phase of soil suspensions, total trace metal concentrations, M_E, M_{Er} and metal extractable with 0.43 M HNO_3 (M_{Ext}) (for details see Sections 2.1 to 2.3). Clay content was estimated for MA soils from the soil texture for a given soil series but was measured directly on Urban soils as described in Section 2.1.3. Soil carbon content was determined in the previous work using a LECO combustion analyser (Tye et al., 2003). A comparison of E-values (M_E) for Zn and Cd measured previously using radio-isotopes (Tye et al., 2003) and stable isotopes (this study) was also undertaken to determine whether further metal fixation had occurred during the ten year storage period, notwithstanding the air dry nature of the soil.

3.2.3 Measuring E-values (M_E) and E-resin (M_{Er})

Labile Ni, Cu, Zn, Cd and Pb (M_E and M_{Er}) were determined by isotopic dilution (see Section 2.3). However, for several Urban soils, the native and/or spike metal concentrations in the suspension were too low to be reliably measured by ICP-MS which may compromise the quality of the isotope analysis (Midwood, 2007). Therefore, two modifications of the standard method were investigated: (i) an increase in the soil:solution ration from 1g in 30 mL of 0.01 M $Ca(NO_3)_2$ to a ratio of 2g: 30 mL; and (ii) using a suspending solution of 1×10^{-5} M EDTA to shift the labile metal equilibrium in favour of the soil solution while attempting to minimize the risk of mobilizing non-labile metal. Test experiments were undertaken on two Urban soils (Wolverhampton, WV22 and Nottingham, NG13) as low trace metal solution concentration in 0.01 M $Ca(NO_3)_2$ were observed for these soils. The measured ICP-MS signal, as counts per second (CPS), of ^{204}Pb and ^{208}Pb in the soil supernatant for both spiked and unspiked samples were compared with those from the standard method.

3.2.4 Describing lability (%M_E) as a function of soil properties

Two types of empirical models were established and assessed for describing and predicting % M_E (M_E divided by total metal concentration, M_{total}) from measured soil properties. The first was a linear multi-regression model where the simplicity of the equation allows inclusion of a large number of variables. Correlation between available soil characteristics and %M_E was tested including pH, %SOC, %Clay, Fe, Al and Mn oxide contents (mg kg^{-1}) and total Ni /Cu /Zn /Cd /Pb concentration (mg kg^{-1}). The significance of each variable was determined using Minitab vs 16.2.2 and only those that were significant ($p < 0.05$) in predicting %M_E were included in the model (Eq. 3-1). The constants were optimized using the *Solver* function in Microsoft Excel.

$$\begin{aligned}\%M_E \;=\; & k_0 + k_1\,(pH) + k_2\,(\%SOC) + k_3\,(FeOx) + k_4\,(MnOx) \\ & + k_5\,(AlOx) + k_6\,(\%Clay) + k_7\,(M_{total}) \qquad (3\text{-}1)\end{aligned}$$

The second model developed was a logistic (sigmoid) model based on the assumption that pH is the main factor that controls $\%M_E$. The logistic function of pH, as a major controlling factor, related to trace metal adsorption on binding phases (e.g. mineral (hydr)oxides) has been generally accepted in the literature (e.g. Sinitsyn et al., 2000; Lamy et al., 1993). The spread of the data can then be described as an exponential function which is different for each metal. Therefore, metal lability was first predicted by assuming that $\%M_E$ was controlled only by pH and that the pH_{50} is metal specific (Eq. 3-2).

$$\%M_E = \frac{100}{1+\exp[k_M(pH-pH_{50})]} \qquad (3\text{-}2)$$

where k_M is a metal-specific constant that controls the spread of the data across the pH range and pH_{50} is the pH at which $\%M_E$ is 50% . In addition to soil pH, the presence of adsorption phases such as mineral (hydr)oxides, clay and soil organic matter are likely to affect the 'spread' of the data. However using this approach it is impossible to include all the available variables which might play a role in adsorption. For simplicity it was therefore assumed that for each metal, the spreading factor is dominated by one adsorption phase. A further simplification was to combine the oxide phases, by allowing differences in the molecular weight for $Fe_2O_3{\cdot}H_2O$, MnO_2 and Al_2O_3, resulting in three variables (%SOC, mineral (hydr)oxides and clay) one of which would control the spread of the data (Eq. 3-3). This is then accounted for by introducing a new metal-specific exponential constant (k_S) which is a function of an additional soil characteristic (S); a power function (n) was added because it achieved a better fit to the data in practice.

$$\%M_E = \frac{100}{1+\exp[k_S S^n(pH-pH_{50})]} \qquad (3\text{-}3)$$

where k_S is a factor that controls the spread of the data depending on a soil characteristic (S) other than pH. The constants in both Eq. 3-2 and 3-3 were optimized using the *Solver* function in Microsoft Excel to assess the performance of each model.

3.3 Results and Discussion

3.3.1 Soil characteristics of MA and Urban soils

Soil properties (pH, %SOC, %Clay, Fe, Mn and Al oxides) for MA soils are summarized in Fig. 3-1 and details listed in Table 3-1. Soil pH in 0.01 M $Ca(NO_3)_2$ covered a wide range (pH 3.57 - 7.12), with 4 soils being very acidic ($pH \leq 4$) and 7 soils with pH close to 7. Organic matter (%SOC) ranged from 1.0% to 7.1%. Iron oxide content (FeOx) ranged from 5 - 30 g kg^{-1} and is likely to strongly affect trace metal lability as it has a high affinity and sorption capacity for cationic trace elements (Tack, 2010). Mn oxides content (MnOx) was significantly lower than FeOx but MnOx can strongly adsorb trace metals (Dong et al., 2003; Trivedi and Axe, 2001). Clay content ranged from 5% (in very sandy soils) to a maximum of 70%. Total concentrations of Ni, Cu, Zn, Cd and Pb after incubation are shown in Table 3-1. Although the main source of metal in the MA soils was addition as metal salts prior to incubation (see Section 3.2.1), some of the soils also had a considerable amount of native soil metal (Tye et al., 2003; Tye et al., 2004).

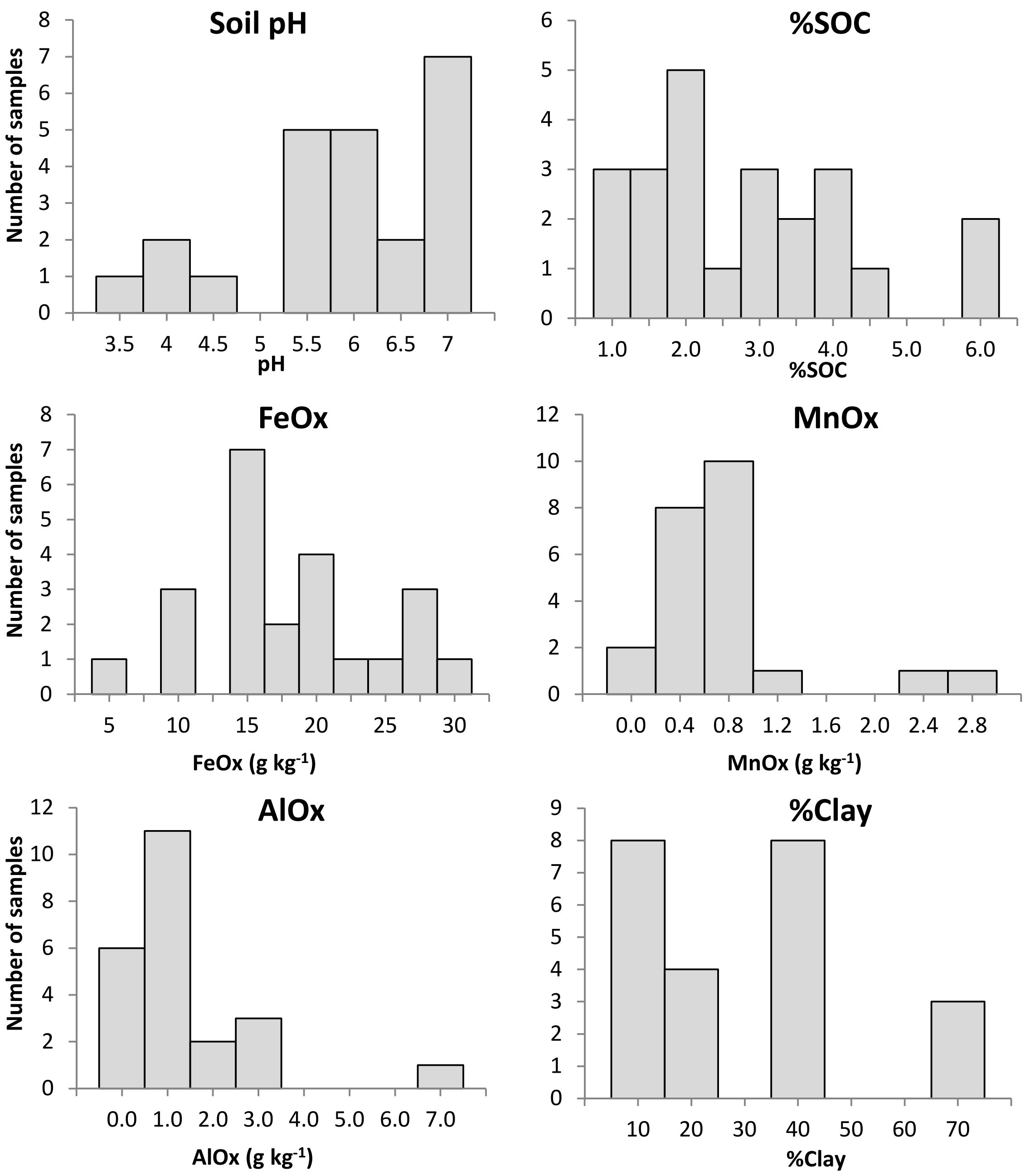


Figure 3-1: Soil characteristics of MA soils shown as histograms for soil pH, %SOC, FeOx (g kg^{-1}), MnOx (g kg^{-1}), AlOx (g kg^{-1}) and %Clay.

Table 3-1: Summary of soil properties and total Ni, Cu, Zn, Cd and Pb concentration following metal amendment in MA soils

Soil series	**pH**	**SOC**	**FeOx**	**MnOx**	**AlOx**	**Clay**	**Total Ni**	**Total Cu**	**Total Zn**	**Total Cd**	**Total Pb**
		%	**g kg^{-1}**	**g kg^{-1}**	**g kg^{-1}**	**%**	**mg kg^{-1}**	**mg kg^{-1}**	**mg kg^{-1}**	**mg kg^{-1}**	**mg kg^{-1}**
Bardsey	3.57	2.91	19.9	0.16	1.44	35.0	69.6	134	273	2.5	343
Wick	3.76	4.25	17.9	0.22	1.26	10.0	82.1	158	327	2.7	346
Iveshead	3.79	5.88	16.3	0.20	2.99	5.0	73.8	147	328	3.1	345
Pwelliperian	4.69	2.70	14.6	0.47	3.44	35.0	97.7	161	498	3.1	341
Rivington	5.27	2.15	10.4	0.40	0.65	5.0	71.1	133	324	2.9	311
Braunshweig	5.36	2.01	5.4	0.61	0.10	15.0	73.3	138	318	2.8	292
Woburn	5.61	0.86	20.1	0.24	0.24	5.0	76.5	136	287	2.7	294
Fladbury	5.67	4.14	24.8	0.86	0.94	70.0	111	155	385	3.3	328
Ticknall	5.72	4.18	20.9	0.48	1.76	15.0	86.9	150	437	3.0	572
Cottam	5.76	2.07	15.6	0.61	0.67	15.0	92.1	161	365	3.5	332
Rosemaund	5.84	1.70	10.8	0.74	0.56	35.0	124	153	351	2.9	297
Arrow	6.06	1.72	15.9	0.57	0.64	10.0	83.1	156	347	3.2	323
Watlington	6.11	1.21	18.2	0.81	0.22	10.0	88.5	146	335	3.1	304
Insch	6.20	3.30	27.4	0.78	7.38	17.5	79.2	140	293	2.8	268
Newport	6.31	0.92	15.8	0.51	0.55	10.0	83.3	157	350	2.9	314
Gleadthorpe	6.45	1.50	8.8	0.35	0.41	10.0	75.2	145	341	3.0	306
Denchworth	6.78	4.21	18.9	0.77	1.07	70.0	98.0	155	316	2.8	300
Ragdale	6.78	3.41	29.8	0.79	1.60	35.0	107	154	404	3.3	321
Bridgets	6.91	2.00	28.7	2.68	3.01	35.0	108	142	377	4.3	306
Marian	6.95	5.98	15.8	2.54	0.15	35.0	93.6	172	417	3.2	377
Worcester	7.08	2.77	15.4	0.79	0.00	35.0	124	166	436	3.3	339
Hanslope	7.08	3.10	27.6	0.77	1.41	35.0	93.6	139	360	2.9	285
Evesham	7.12	2.08	22.0	0.98	1.11	70.0	97.6	155	337	3.0	312

Correlations between soil properties are given in Table 3-2, only weak correlation was found between soil pH and MnOx; also between AlOx and FeOx. Total concentration of Ni was weakly positively correlated with pH, MnOx and %Clay a positive correlation between total Cd and MnOx ($P<0.001$) was also observed. This may be attributed to strong binding on these solid sorption phases at neutral – high pH (Rieuwerts et al., 2006; Zimdahl and Skogerboe, 1977). As the majority of metal was added to this set of soils, the effect of soil properties on total metal concentration has however been reduced.

Table 3-2: Correlation matrix between measured soil properties, MA soils (r-values and P-values in brackets); significant correlations are in bold italic type.

MA soil properties	**pH**	**SOC (%)**	**FeOx (g kg^{-1})**	**MnOx (g kg^{-1})**	**AlOx (g kg^{-1})**	**Clay (%)**
SOC (%)	-0.20 (0.36)					
FeOx (g kg^{-1})	0.28 (0.19)	0.22 (0.31)				
MnOx (g kg^{-1})	***0.56*** ***(<0.05)***	0.19 (0.37)	0.31 (0.15)			
AlOx (g kg^{-1})	-0.14 (0.54)	0.25 (0.25)	***0.48*** ***(<0.05)***	0.05 (0.81)		
Clay (%)	0.37 (0.08)	0.26 (0.24)	0.37 (0.08)	0.34 (0.11)	-0.01 (0.97)	
Total Ni (mg kg^{-1})	***0.51*** ***(<0.05)***	0.04 (0.85)	0.27 (0.21)	***0.43*** ***(<0.05)***	-0.11 0.63	***0.60*** ***(<0.05)***
Total Cu (mg kg^{-1})	0.25 (0.25)	0.33 (0.13)	-0.07 (0.74)	0.29 (0.18)	-0.20 0.36	0.30 (0.16)
Total Zn (mg kg^{-1})	0.25 (0.25)	0.23 (0.29)	0.05 (0.81)	0.31 (0.16)	-0.02 (0.93)	0.21 (0.34)
Total Cd (mg kg^{-1})	0.40 (0.06)	-0.02 (0.92)	0.32 (0.14)	***0.68*** ***(<0.001)***	0.09 (0.67)	0.14 (0.53)
Total Pb (mg kg^{-1})	-0.16 (0.46)	0.41 (0.05)	0.01 (0.95)	-0.02 (0.92)	-0.06 (0.77)	-0.09 (0.68)

The Urban soils also covered a wide range of soil properties and land uses (Fig. 3-2, Table 3-3). Soil pH ranged from 3.9 to 8.1 with 60% pH >6.0. As brownfield site soils most are also low in organic matter content (%SOC). Fe oxide concentration ranged from 7.08 to 43.0 g kg^{-1} and was strongly correlated with Mn oxide (Table 3-4).

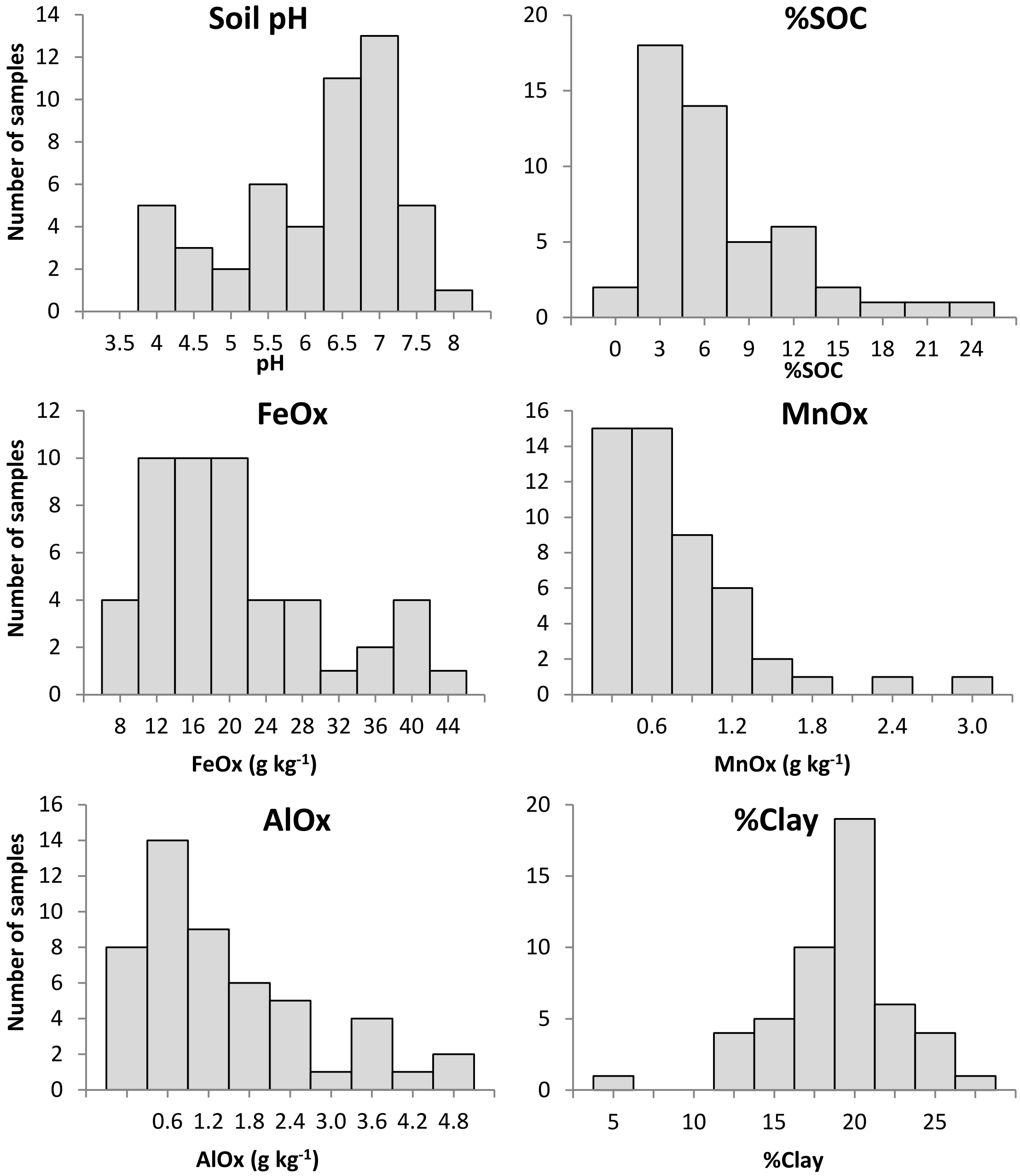


Figure 3-2: Soil characteristics of Urban soils shown as histograms for soil pH, %SOC, FeOx (g kg^{-1}), MnOx (g kg^{-1}), AlOx (g kg^{-1}) and %Clay.

Table 3-3 a): Summary of soil properties, sampling location, landuses and total Ni, Cu, Zn, Cd and Pb concentration in Urban soils (Wolverhampton)

Sample	pH	SOC %	FeOx g kg^{-1}	MnOx g kg^{-1}	AlOx g kg^{-1}	Clay %	Total Ni mg kg^{-1}	Total Cu mg kg^{-1}	Total Zn mg kg^{-1}	Total Cd mg kg^{-1}	Total Pb mg kg^{-1}	Grid Ref.	Landuse
WV1	4.63	3.43	8.4	0.24	0.81	20.4	18.3	35.4	134	0.4	56.7	391030 301170	Grassland (Golfcourse)
WV2	3.92	2.81	9.7	0.44	1.08	22.5	12.1	19.4	61.5	0.3	53.2	388450 300860	Grassland (Golfcourse)
WV3	6.11	11.0	18.2	1.26	1.59	15.7	55.3	403	1050	3.7	266	393690 297700	Park
WV4	5.87	3.35	13.8	0.46	1.07	19.8	22.9	69.1	313	0.6	109	392180 297280	Grassland (open land)
WV5	6.46	13.4	26.3	0.84	1.16	14.6	77.9	233	588	1.3	227	393730 296530	Old industrial (gas work)
WV6	6.84	10.3	38.5	1.52	2.50	18.6	300	2200	4170	26.1	746	394490 295160	Wasteland (old landfill)
WV7	7.41	6.45	23.1	0.67	0.88	16.9	83.3	493	1170	3.6	590	394960 295960	Grassland
WV8	5.64	10.1	19.7	0.92	4.47	16.4	57.4	158	553	2.1	358	397060 296980	Woodland
WV9	5.56	4.39	16.8	0.62	1.66	18.4	32.3	134	300	0.9	273	390170 301290	Grassland
WV10	7.07	8.49	19.9	0.84	0.94	18.3	66.9	298	1100	4.1	1050	391330 301090	Old industrial (tyre factory)
WV11	4.46	3.68	14.9	0.38	1.80	22.9	25.3	87.7	514	1.9	173	390810 296710	Domestic garden
WV12	5.79	2.73	11.1	0.51	0.75	18.9	15.0	30.4	270	0.7	199	388240 295750	Grassland
WV13	5.54	3.83	12.6	0.64	0.71	24.4	25.4	51.1	308	1.0	131	392760 295760	Park
WV14	5.22	25.1	20.3	0.55	0.68	5.1	58.4	265	1190	5.1	332	392830 299710	Nature reserce (railway)
WV15	7.12	12.1	19.2	3.01	0.55	11.8	47.6	101	290	1.1	140	388910 299580	Nature reserce (railway)
WV16	4.01	14.5	7.1	0.27	5.10	16.8	29.1	89.1	135	0.7	88.4	392380 295460	Vegetated colliery spoil heap
WV17	5.64	2.76	11.1	0.52	0.87	19.4	26.9	73.8	239	0.6	141	391240 297580	Old industrial (disused factory)
WV18	4.63	5.49	16.7	0.51	2.49	22.6	39.8	204	524	1.7	150	395120 299290	Grassland (not maintained)
WV19	5.37	5.19	12.6	0.41	0.01	19.1	45.8	58.7	192	0.5	48.7	395150 299140	Grassland
WV20	4.14	4.54	10.8	0.25	2.36	15.3	15.1	77.6	105	0.5	85.7	396720 301130	Grassland
WV21	6.41	2.89	13.5	0.48	0.00	24.0	54.5	48.7	102	0.2	31.9	393960 299860	Brownfield
WV22	8.08	3.00	12.3	0.35	0.00	21.3	43.1	90.0	154	0.3	45.9	394390 299860	Brownfield
WV23	7.63	1.70	12.8	0.29	0.02	19.0	65.6	79.0	267	0.7	78.8	395400 299670	Brownfield
WV24	7.22	3.21	14.1	0.45	0.43	21.1	33.6	82.3	605	1.2	351	394680 299830	Brownfield
WV25	4.05	11.4	20.1	0.70	3.84	13.8	55.2	188	356	1.8	217	394210 298970	Grassland (park)
WV26	6.60	2.78	9.9	0.28	0.00	20.6	20.0	52.6	133	0.5	63.3	394600 298750	Grassland
WV27	7.67	3.47	28.7	0.54	0.29	20.5	39.1	160	279	0.6	177	392800 298520	Brownfield
WV28	7.25	6.51	21.1	0.44	0.66	18.8	44.8	150	281	0.6	156	391700 310630	Brownfield
WV29	6.87	20.6	27.4	1.03	0.64	20.0	94.1	338	739	3.3	429	391680 300080	Deciduous woodland
WV30	6.32	3.63	13.5	0.34	0.26	18.6	24.0	82.3	158	0.6	109	391780 300120	Grassland (park)

Table 3-3 b): Summary of soil properties, sampling location, landuses and total Ni, Cu, Zn, Cd and Pb concentration in Urban soils (Nottingham)

Sample	pH	SOC %	FeOx g kg^{-1}	MnOx g kg^{-1}	AlOx g kg^{-1}	Clay %	Total Ni mg kg^{-1}	Total Cu mg kg^{-1}	Total Zn mg kg^{-1}	Total Cd mg kg^{-1}	Total Pb mg kg^{-1}	Grid Ref.	Landuse
NG1	4.75	5.00	40.6	1.67	3.81	24.6	49.5	51.4	340	1.8	194	455470 336660	Grassland (football field)
NG2	5.52	4.77	14.9	0.86	2.33	28.7	24.4	50.0	229	1.1	412	455280 338770	Churchyard
NG3	6.47	5.11	34.4	1.14	1.60	21.4	44.1	26.5	111	0.3	60.9	453760 341550	School
NG4	6.29	5.30	18.8	0.49	1.24	18.3	37.1	224	881	2.5	1300	454700 343720	Domestic garden
NG5	6.99	3.60	14.6	0.47	0.38	20.1	31.6	57.9	439	1.1	353	457360 342370	Domestic garden
NG6	6.78	10.9	39.5	0.81	1.36	19.8	60.1	161	459	2.0	406	458720 339210	Railway
NG7	7.00	5.41	21.4	0.85	0.88	18.8	59.0	129	359	1.5	385	458500 339110	Grassland (imported)
NG8	4.15	7.90	38.2	1.32	4.52	18.0	62.6	65.2	374	1.6	352	459710 339170	Grassland (racecourse)
NG9	6.27	4.81	14.6	0.38	1.50	20.5	28.2	47.6	169	0.6	205	453570 344310	Vegetated colliery spoil heap
NG10	6.53	5.29	18.5	0.79	1.38	21.7	35.2	54.4	301	0.8	257	455170 342480	Vegetated colliery spoil heap
NG11	7.25	9.34	25.3	1.30	1.83	14.8	228	1260	1690	2.1	1110	455770 338590	Disused factory
NG12	7.38	14.3	43.0	2.31	3.32	12.1	380	3190	2520	5.6	1240	455760 338550	Disused factory
NG13	7.02	1.29	15.6	0.35	0.03	20.7	20.3	26.0	26.0	0.1	18.5	453570 344210	Grassland (amenity)
NG14	6.54	11.4	16.7	0.59	3.59	13.0	370	644	1740	38.2	600	464120 340530	Sewage Farm
NG15	5.99	6.86	15.5	0.30	2.79	20.7	199	382	1020	19.2	337	464130 340810	Sewage Farm
NG16	7.57	5.31	25.0	0.93	0.61	18.9	44.0	92.9	223	0.9	121	462830 340800	Brownfield
NG17	6.34	18.3	27.7	1.06	1.33	11.6	99.0	274	634	1.1	1410	462770 340730	Brownfield
NG18	6.78	3.35	22.2	1.13	0.60	20.1	43.7	35.7	216	1.4	79.2	462900 340700	Brownfield
NG19	6.93	2.98	34.8	1.43	2.19	18.7	32.3	31.7	132	0.4	65.9	446800 339730	Brownfield
NG20	6.68	3.00	31.0	0.70	1.01	24.0	32.2	52.9	299	0.7	90.0	447040 339240	Brownfield

Table 3-4: Correlation matrix between measured soil properties, Urban soils (r-values and P-values in brackets); significant correlations are in bold italic type.

Urban soil properties	**pH**	**SOC %**	**FeOx (g kg^{-1})**	**MnOx (g kg^{-1})**	**AlOx (g kg^{-1})**	**Clay %**	**Total Ni (mg kg-1^{1})**	**Total Cu (mg kg^{-1})**	**Total Zn (mg kg^{-1})**	**Total Cd (mg kg^{-1})**
SOC (%)	-0.06 (0.68)									
FeOx (g kg^{-1})	0.26 (0.07)	***0.33*** ***(0.05)***								
MnOx (g kg^{-1})	0.22 (0.13)	***0.36*** ***(0.05)***	***0.65*** ***(0.001)***							
AlOx (g kg^{-1})	***-0.52*** ***(0.001)***	***0.31*** ***(0.05)***	***0.31*** ***(0.05)***	0.25 (0.08)						
Clay (%)	-0.05 (0.75)	***-0.72*** ***(0.001)***	-0.14 (0.33)	***-0.30*** ***(0.05)***	-0.17 (0.24)					
Total Ni (mg kg^{-1})	0.26 (0.06)	***0.40*** ***(0.05)***	***0.41*** ***(0.05)***	***0.39*** ***(0.05)***	***0.36*** ***(0.05)***	***-0.39*** ***(0.05)***				
Total Cu (mg kg^{-1})	0.25 (0.08)	***0.36*** ***(0.05)***	***0.47*** ***(0.001)***	***0.48*** ***(0.001)***	0.28 (0.05)	***-0.35*** ***(0.05)***	***0.85*** ***(0.001)***			
Total Zn (mg kg^{-1})	0.22 (0.12)	***0.43*** ***(0.05)***	***0.43*** ***(0.05)***	***0.38*** ***(0.05)***	0.27 (0.06)	***-0.36*** ***(0.05)***	***0.83*** ***(0.001)***	***0.86*** ***(0.001)***		
Total Cd (mg kg^{-1})	0.09 (0.55)	0.26 (0.06)	0.14 (0.32)	0.09 (0.55)	***0.34*** ***(0.05)***	-0.23 (0.10)	***0.78*** ***(0.001)***	***0.45*** ***(0.001)***	***0.69*** ***(0.001)***	
Total Pb (mg kg^{-1})	0.24 (0.10)	***0.45*** ***(0.001)***	***0.37*** ***(0.05)***	***0.32*** ***(0.05)***	0.20 (0.16)	***-0.39*** ***(0.05)***	***0.58*** ***(0.001)***	***0.60*** ***(0.001)***	***0.63*** ***(0.001)***	***0.29*** ***(0.05)***

In comparison to the MA soils, total metal concentrations in the Urban soils covered a wider range (Ni, 12.1−380; Cu, 19.4−3190; Zn, 26−4170; Cd, 6.1−38.2; Pb, 18.5−1410, mg kg^{-1}) as a consequence of historical contamination (Table 3-3). Individual metal concentrations exhibited strong correlations implying contamination from the same source (Table 3-4). Nickel, Cu, Zn and Pb were positively correlated with %SOC, FeOx and MnOx which, as it has been discussed above, may be because of their accumulation in soils with strong binding phases (Rieuwerts et al., 2006; Zimdahl and Skogerboe, 1977). Cadmium was less affected by soil properties only showing a correlation with AlOx.

3.3.2 E-values (M_E and M_{Er})

3.3.2.1 Modified method for determining M_E in soils with low metal solubility

Increasing the soil:solution ratio resulted in a small increase in the Pb solution concentration (Table 3-5), this was insufficient to improve the %RSD measured (n = 6) and was therefore not a useful was of increasing suspension metal concentration. In contrast, using 1×10^{-5} M EDTA as the suspending solution in systems with a soil: solution ration of 1 g : 30 mL resulted in an increase in solution metal concentration and a reduction in %RSD compared to values measured in 0.01 M $Ca(NO_3)_2$, especially for ^{204}Pb (Table 3-6). There was also a significant difference in ^{204}Pb between unspiked and spiked samples, but not for the ^{208}Pb isotope, which suggests that the non-labile pool was not being mobilized by the acidified spike (see discussion in Section 2.3.2.1). The results suggest that suspension in $Ca(NO_3)_2$ produces a trace metal concentration in the solution which is not analytically robust, a low concentration of EDTA may be used as an alternative suspending solution.

Table 3-5: Comparison of ICP-MS signal for ^{204}Pb and ^{208}Pb (CPS) in spiked and unspiked suspensions for two Urban soils with soil: solution ratios of 1 g in 30 ml and 2 g in 30 ml. Results are an average of 6 measurements; %RSD is the relative standard deviation. Background counts in 0.01 M $Ca(NO_3)_2$ (blank) are listed for comparison.

soil:solution ratio	**^{204}Pb (CPS)**				**^{208}Pb (CPS)**			
	1 g : 30 ml		2 g : 30 ml		1 g : 30 ml		2 g : 30 ml	
	Mean	RSD%	Mean	RSD%	Mean	RSD%	Mean	RSD%
WV22 (pH 8.2) unspiked	56.1	11.7	71.2	54.5	1444	3.5	1252	59.4
spiked	60.4	6.20	78.2	59.3	700	2.9	947	81.1
NG13 (pH 7.6) unspiked	39.8	9.40	60.7	13.7	744	4.0	297	14.7
spiked	74.9	6.50	62.2	8.30	558	3.6	226	18.7
Blank	40.3				150			

Table 3-6: Comparison of ICP-MS signal of ^{204}Pb and ^{208}Pb (CPS) for spiked and unspiked samples of two Urban soils suspended in 0.01 M $Ca(NO_3)_2$ or 1×10^{-5} M EDTA (soil:solution 1 g : 30 mL). Values are an average of 6 measurements; %RSD is the relative standard deviation. Background counts in 0.01 M $Ca(NO_3)_2$ (blank) are listed for comparison.

Suspending solution	**^{204}Pb (CPS)**				**^{208}Pb (CPS)**			
	0.01M $Ca(NO_3)_2$		1×10^{-5} M EDTA		0.01M $Ca(NO_3)_2$		1×10^{-5} M EDTA	
	Mean	RSD%	Mean	RSD%	Mean	RSD%	Mean	RSD%
WV22								
(pH 8.2)								
unspiked	56.1	11.7	17995	2.80	1444	3.50	686675	2.90
spiked	60.4	6.20	155067	1.10	700	2.90	620508	1.00
NG13								
(pH 7.6)								
unspiked	39.8	9.40	5534	2.10	744	4.00	209771	2.80
spiked	74.9	6.50	307009	4.2	558	3.60	377738	4.70
Blank	40.3				150			

3.3.2.2 Labile Ni, Cu, Zn, Cd and Pb in soils

Values of %M_E for Ni, Cu, Zn, Cd and Pb in MA and Urban soils are shown in Fig. 3-3. With the exception of Cd in MA soils, average labilities were all below 50%, which emphasises that the use of total metal concentration in risk assessment is likely to overestimate risk. The metal lability of all five metals was greater in the MA soils than in the historically contaminated Urban soils (c. 25% greater for Cu, Zn and Cd and c. 30% greater for Ni and Pb), even after almost three years incubation at 80% field capacity and 10 years of dry storage.

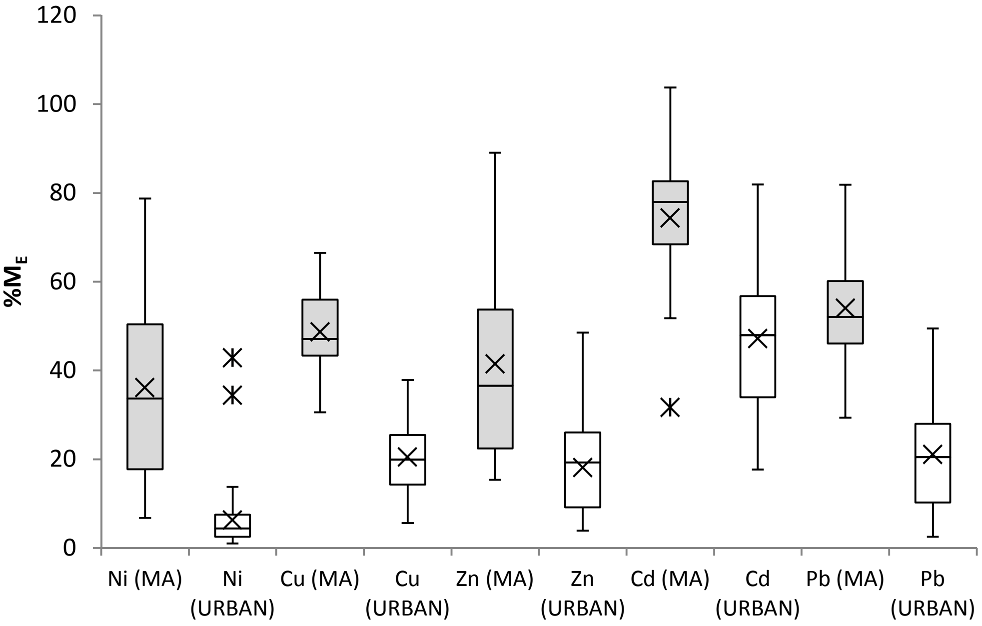


Figure 3-3: Box and whisker plot showing lability of Ni, Cu, Zn, Cd and Pb (% M_E) for soils from MA soils (23) and Urban soils (50); the mean value (×) is also shown and outliers are marked as an asterix.

Despite the difference in average %M_E the relative lability of the five metals followed the sequence: Cd > Pb ≥ Cu > Zn > Ni in both sets of soils, which agrees with the findings of Gabler et al., (2007) who measured the lability of these five metals on 115 unpolluted soil samples using a stable isotope dilution approach. The range of lability in MA soils is a product of the 'ageing' or 'fixation' process that occurred during 3 years of incubation and possibly during the following 10-year period of dry storage (see Section 3.3.5). The difference between the five metals reflects the variation in mechanism or rates of metal fixation into soil constituents. The main fixation mechanisms operating may be solid-phase diffusion on goethite (Bruemmer et al., 1988) and on Fe, Al and Mn (hydr)oxides (Trivedi and Axe, 2001). In calcareous soils, diffusion in to carbonate (Hamon et al., 2002; Collins et al., 2003; Buekers et al., 2007), or (surface) precipitation in high pH soils is important (Ma et al., 2006b). The relatively larger proportion of Zn and Ni fixed may have been due to substitution for Fe in oxyhydroxides (Buekers et al., 2008a; Manceau et al., 2000). Such reactions may be less likely for larger ions such as Cd and Pb (Buekers et al., 2008a; Xu et al., 2006). In general, fixation reactions are correlated with the ionic radius of the metals: the larger the radius, the slower the reaction i.e. Ni ≥ Cu > Zn > Cd ~ Pb (Degryse et al., 2009; Degryse et al. 2007). It has also been shown that the reaction is affected by soil properties such as pH and total metal concentration (Tye et al., 2003; Ma et al., 2006b). For example, the relatively lower lability (greater fixation) of Pb than Cd may be because Pb is precipitated as insoluble minerals such as pyromorphite in phosphate rich soils (Dermatas et al., 2008); Cu however, is predominantly bound with organic matter which leads to a slightly higher lability than Zn. Metal salt incubation studies of the fixation process have also recorded changes in soil characteristics during incubation. Thus, for example, soil pH may decrease during incubation which suppresses Pb fixation (Degryse et al., 2007) or increase causing enhanced fixation of Cd and Zn (Tye et al., 2003).

For the Urban soils, the range of labilities must be determined not only by past fixation processes but also by the various sources of contamination that made up the soil metal loading. The lower metal lability in the Urban soils may because of longer contact time between metal and soil and/or because the metals were added to soils in forms which are less soluble than the metal salts added to MA soils. The former may have relatively small contribution as both sets had metal-soil contact time for more than 13 years. Fixation process, though depends on soil properties such as pH (Tye et al., 2003), is believed to be much slower over a time period of years (Crout et al., 2006). The latter, however, is in line with expectation. For example, low Pb lability is generally found in minespoils soils where the origin of the metal is relatively insoluble metal sulphides or carbonate minerals (Degryse et al., 2004; Marzouk et al., 2013). Similarly Cd and Zn in soils surrounding smelters are typically contaminated by atmospheric deposition or smelter wastes which give rise to relatively low metal lability (Degryse et al., 2004). The relatively high lability of Cd in Urban soils, relative to other metals does not necessarily indicate that Cd was added in soils in a more soluble source; it has been shown that Cd and Zn added to soil in identical (isomorphically substituted) forms nevertheless have quite different labilities due to the action of soil processes (Marzouk, 2012; Degryse et al., 2004). The average $\%M_E$ for Ni was only 6.3% (includes outliers) and $\%M_E$ values fell in a restricted range (with 75% of the soils having less than 10% Ni lability). A smaller range of lability was also observed for Zn in the Urban soils than in the MA soils. This was not expected as (i) the Urban soils had a larger variation of metal contaminant sources and (ii) the ranges of soil pH values (a major determinant of $\%M_E$) in the MA and Urban soils were broadly similar (Figs 3-1 and 3-2) . A possible explanation is that the Urban soils had relatively low levels of Ni loading with 90% having Ni concentrations ≤ 100 mg kg^{-1}. Thus it is likely that a large proportion of Ni in these soils is geogenic, bound within primary minerals, and relatively non-reactive. Similar results were found by (Ma and Rao, 1997) as they studied nine contaminated soils with relatively low Ni concentrations and found that 57 - 94% of Ni was in the residual fraction measured by sequential extraction.

3.3.2.3 Presence of non-labile metal in soil colloidal phases

The presence of a non-labile fraction of metal in suspended colloidal particles (SCP-metal) in the filtered supernatant solutions from the soil suspensions used to measure E-values can be investigated by comparing M_E and M_{Er}. For most of the soils, the SCP-metal had only a very small effect on measured E-values and statistical analysis showed that a significant difference was only observed for Ni, Cu and Pb in the soils investigated (Table 3-7).

Table 3-7: P values for a paired T-test comparing M_E and M_{Er}.

	Ni	Cu	Zn	Cd	Pb
MA	***0.014***	***0.011***	0.055	0.233	***0.032***
Urban	0.909	***0.000***	0.277	0.766	0.083

Within the MA soils Ni, Cu and Pb exhibited evidence of SCP-metal but only Cu showed evidence of this in the Urban soils; good correlations between $\%M_E$ and $\%M_{Er}$ for these metals were observed with r^2 values of 0.999, 0.951, 0.992 and 0.950, respectively (Fig. 3-4) because the majority of soils provided very little evidence of SCP-metal. The average difference between $\%M_E$ and $\%M_{Er}$ is shown in Table 3-8, and except for Zn, Cd and Pb in MA soils, all other values of $\Delta\%M_E$ is less than 1%. Negative values, i.e. $\%M_{Er}$ is higher than $\%M_E$, have been observed for Ni in MA soils (-0.57) and Zn and Pb in Urban soils (-0.69 and -0.94, respectively), which can be explained by experimental error, attributed to i) the chelex resin mobilizing the non-labile fraction; and ii) lack of accuracy in measuring samples with very low metal concentrations. The average standard error for (% total metal content) calculated for M_E and M_{Er} are shown in Table 3-8.

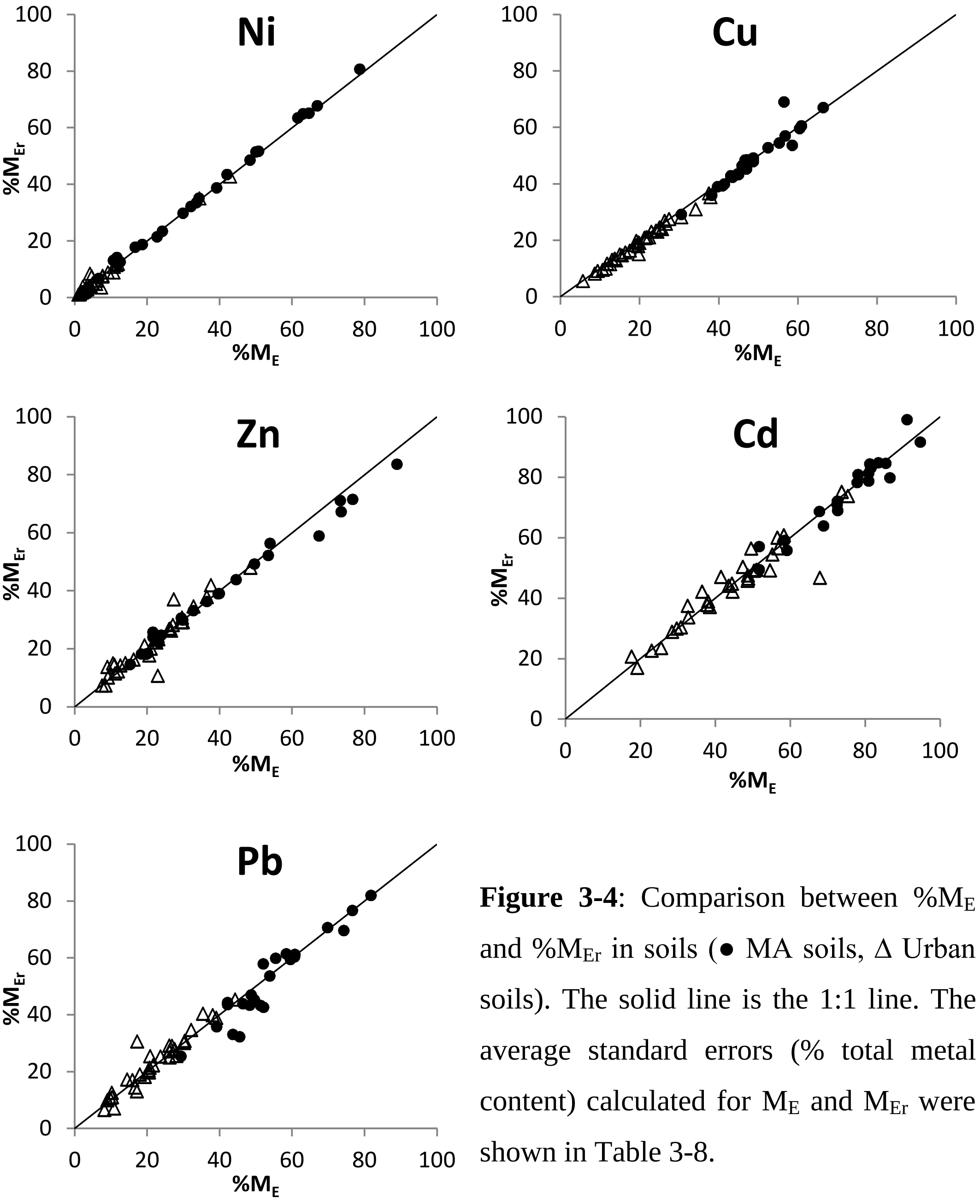


Figure 3-4: Comparison between $\%M_E$ and $\%M_{Er}$ in soils (● MA soils, Δ Urban soils). The solid line is the 1:1 line. The average standard errors (% total metal content) calculated for M_E and M_{Er} were shown in Table 3-8.

Table 3-8: The average difference between $\%M_E$ and $\%M_{Er}$ (italic), and average standard errors (% total metal content) calculated for M_E and M_{Er}

		Ni	**Cu**	**Zn**	**Cd**	**Pb**
MA	$\Delta\%M_E$	*-0.57*	*0.25*	*1.24*	*4.40*	*2.29*
	SE ($\%M_E$)	0.56	0.99	0.98	1.55	1.41
	SE ($\%M_{Er}$)	0.85	1.35	1.50	2.33	1.91
Urban	$\Delta\%M_E$	*0.02*	*0.84*	*-0.69*	*0.25*	*-0.94*
	SE ($\%M_E$)	0.28	0.52	0.47	1.02	1.13
	SE ($\%M_{Er}$)	0.20	0.58	0.99	2.06	1.81

The ratio of M_E : M_{Er} is shown in Figs. 3-5 and 3-6 as a function of $\%M_E$ and soil pH, respectively, for all metals and soils. Despite experimental errors that result in relative scattering of the data along the ratio = 1 line, the results observed are generally larger and more scattered at low levels of $\%M_E$ as might be expected because as metal lability increases there is less scope for a substantial effect from non-labile SCP-metal and M_{Er} will gradually approach the value of M_E. The deviation of M_{Er} from M_E values is also generally greater with increasing soil pH. This result agrees with the findings of others (e.g. Lombi et al., 2003; Marzouk, 2012) that in high pH soils, there is a larger proportion of non-labile SCP-metal which is likely to be more mobile. It has been suggested that the presence of colloidal carbonate or Fe oxide particles in the soil solution of high pH soils may contribute to this fraction (Lombi et al., 2003; Nolan et al., 2009). The presence of non-labile SCP-Cu and Pb has also been suggested to be a function of strong adsorption of Cu and Pb on DOM while Ni, Zn and Cd more likely to be related with colloidal Fe, Al and Mn (hydr)oxides (Lombi et al., 2003; Ma et al., 2006b; Nolan et al., 2009; Marzouk, 2012). However, in this work no significant correlation ($p>0.05$) was observed between the ratio of M_E : M_{Er} and solution concentration of Fe, Al, Mn or DOC measured in 0.01 M $Ca(NO_3)_2$ (solid: solution ratio 1 g: 30 mL). Marzouk (2012) only found a very weak correlation between this ratio and mineral oxides in solution, suggesting that for Pb the relatively high pH may enhance adsorption onto all colloidal mineral oxides. For Cu, the small difference between M_E and M_{Er} may be a consequence of the presence of a salt (0.01 M $Ca(NO_3)_2$) as this will help flocculate DOC from the soil solution (Romkens et al., 1996) and thus $\%M_{Er}$ may be less than that measured in a solution with lower ionic strength.

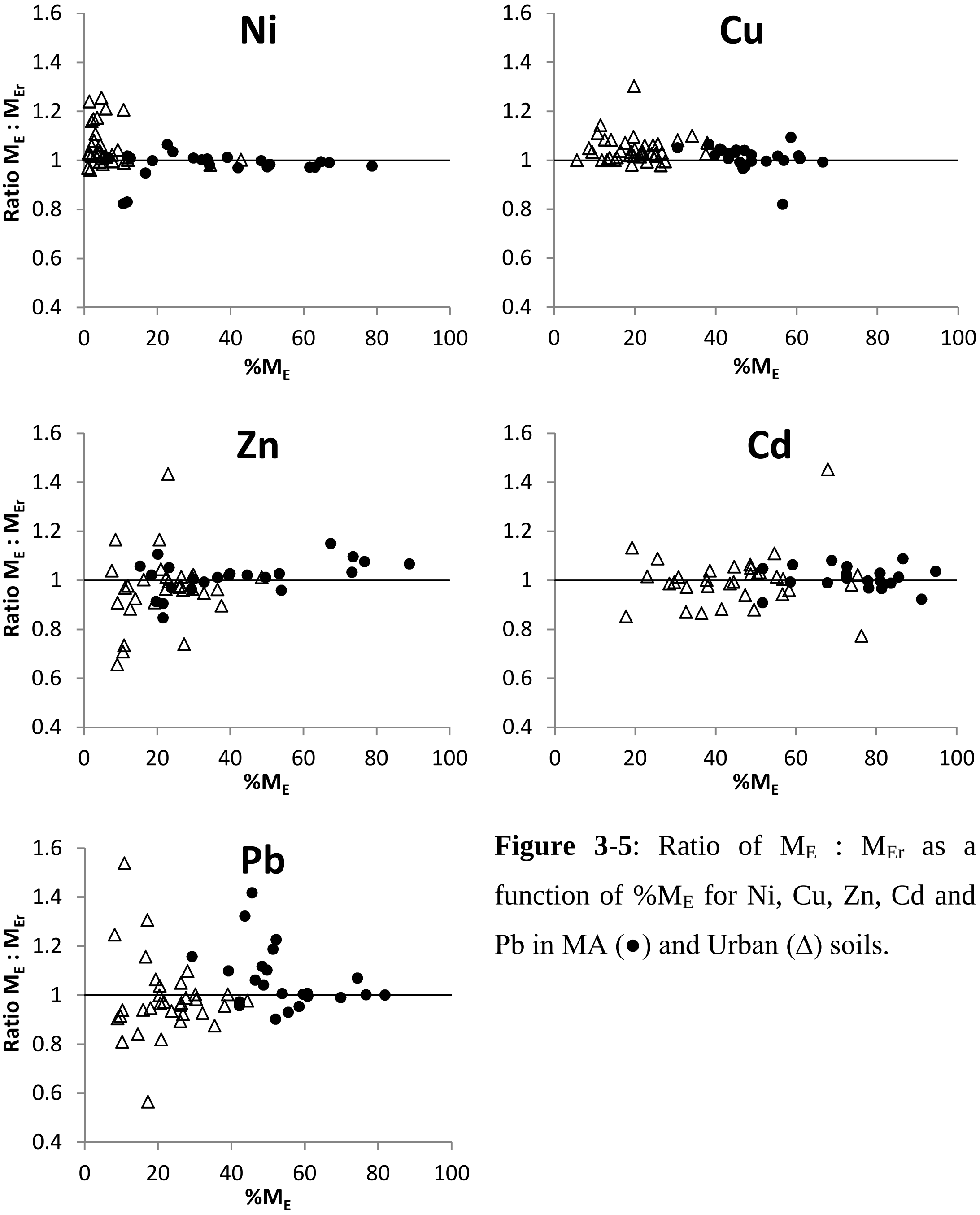


Figure 3-5: Ratio of M_E : M_{Er} as a function of %M_E for Ni, Cu, Zn, Cd and Pb in MA (●) and Urban (Δ) soils.

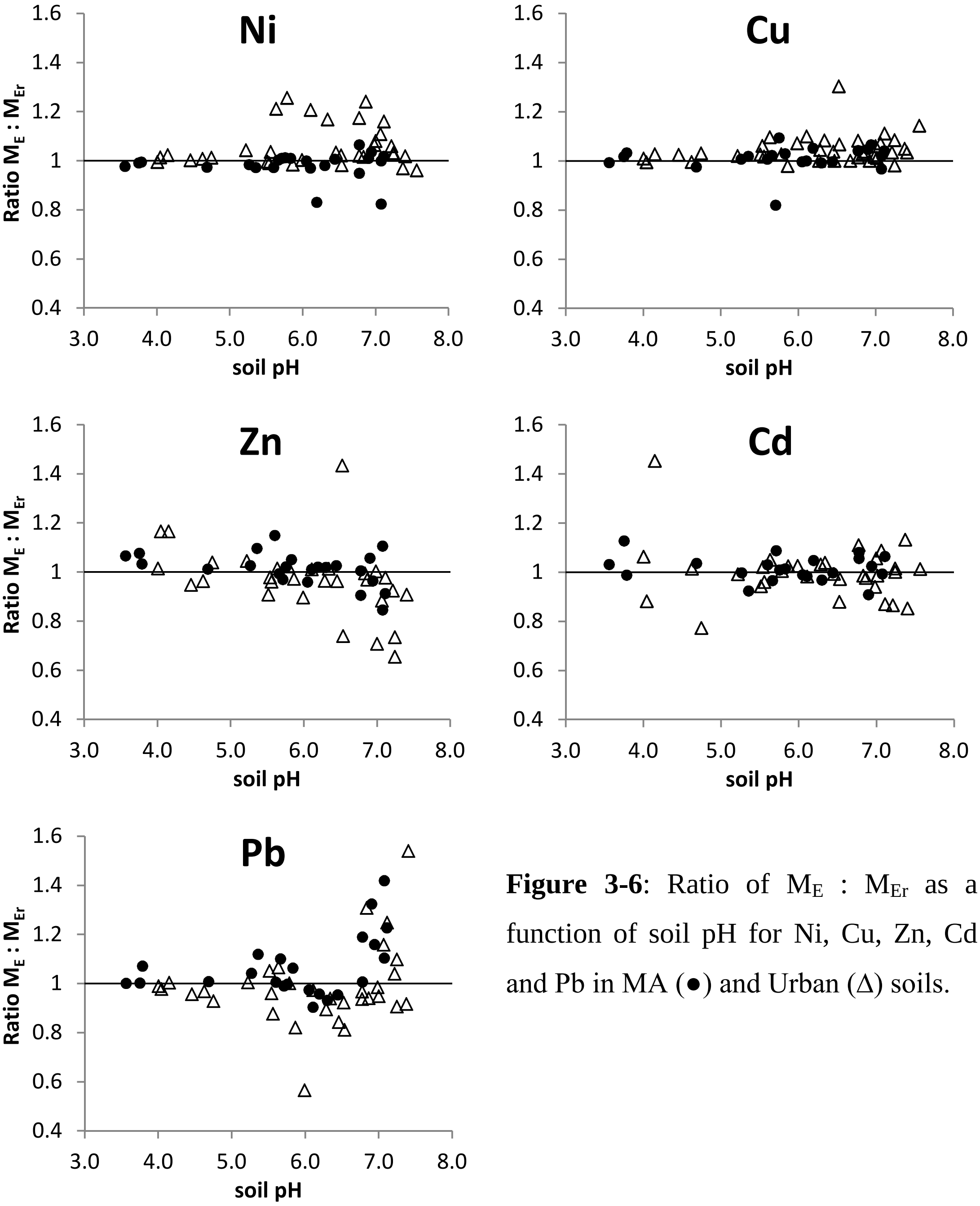


Figure 3-6: Ratio of M_E : M_{Er} as a function of soil pH for Ni, Cu, Zn, Cd and Pb in MA (●) and Urban (Δ) soils.

3.3.3 Modelling of E-values using soil properties

3.3.3.1 Effect of soil properties on metal lability (%M_E)

An advantage of using a multiple linear-regression model is the ability to include many variables. In the current study, seven variables were available to describe metal lability (Eq. 3-1). Prior to parameterization, correlations between individual soil properties and %M_E were assessed (Table 3-9).

For both sets of soils, pH was inversely related to lability (%M_E) and was the most important characteristic for all five metals ($p<0.05$) in agreement with the findings of others including Martinez and Motto, (2000), Rieuwerts, et al. (2006), Schulin et al., (2010) and Buekers et al., (2007). Stronger adsorption of metals is certainly expected with increasing pH due to the increase in negative charge on all adsorption surfaces (Crout et al., 2006). Metal fixation is not necessarily linked directly to strength of adsorption, but it seems reasonable to assume that factors that increase adsorption strength will also increase fixation. Other processes may also explain the influence of pH on lability, including metal hydrolysis at high pH followed by strong, preferential adsorption of the resulting metal hydroxide complexes (Basta and Tabatabai, 1992) and precipitation as carbonate phases, (e.g. Gambrell, 1994; Charlatchka and Cambier, 2000).

Apart from pH, other properties were found to be important for specific metals or for a specific set of soils. In MA soils, MnOx was strongly correlated with metal lability ($p<0.05$) and, although it is a strong binding phase, especially for Pb ($p<0.001$) (Bonten et al., 2008; Weng et al., 2001), it also had an intercorrelation with soil pH (Table 3-2). Of the five metals, Cu lability was most poorly related to measured soil properties. Copper fixation has previously been shown to be poorly related to soil properties (Buekers et al., 2007; Buekers et al., 2008b). It is widely recognised that adsorption on organic matter dominates Cu dynamics in soils (Weng et al., 2001) but no significant correlation was found between Cu lability and %SOC in this study ($p>0.05$). In contrast, the lability of Ni and Zn were significantly correlated with several soil properties, including Fe and Mn oxides and clay content. The underlying mechanism for metal fixation on Fe-oxide is trace metals diffusion into micropores and/or incorporation into the mineral structure (Jacquat et al., 2009; Manceau et al., 2000; Degryse et al., 2011). A relatively weak and negative correlation was found between %Clay and $\%M_E$ for Ni and Zn ($k=-0.484$ and -0.482, respectively). Clay minerals are a source of negative surface charges in soils and are generally recognised as important adsorption surfaces for trace cations (Tack, 2010). However, only a very small proportion of labile metals are normally truly ‘exchangeable’ (Nakhone and Young, 1993) and it may be that the correlation with clay content reflects a more general association with soil geocolloid concentration rather than implying that labile Zn and Ni are held as exchangeable cations on alumina-silicate clay minerals. Specific adsorption of Zn on hydroxyl-interlayered clays in acidic soils has also been demonstrated by Degryse et al. (2011). In the MA soils, there is a negative correlation between total Ni, Zn or Cd content and lability ($\%M_E$), which is likely to be a result of the native metal (before incubation) being less labile than those added as metal salts. Thus, Sterckemen et al. (2009) also found more isotopically exchangeable Cd ($\%M_E$ = 46%) in cultivated soils than in geochemically enriched soils ($\%M_E$ = 20%) in which there was likely to be a higher proportion of Cd from parent materials held within primary mineral matrices.

Table 3-9: Correlations between soil properties and %M_E for MA soils. Significant values are shown in bold where P is <0.001 or <0.05.

MA soils	**pH**	**SOC**	**FeO_x**	**MnOx**	**AlOx**	**Clay**	**Total metal content**
%M_E values		(%)	(g kg^{-1})	(g kg^{-1})	(g kg^{-1})	(%)	(mg kg^{-1})
Ni	-0.855	0.008	-0.419	-0.576	-0.179	-0.484	-0.716
	<0.001	>0.05	***<0.05***	***<0.05***	>0.05	***<0.05***	***<0.001***
Cu	-0.505	-0.256	-0.269	-0.483	-0.456	-0.271	-0.305
	<0.05	>0.05	>0.05	***<0.05***	***<0.05***	>0.05	>0.05
Zn	-0.813	-0.022	-0.427	-0.533	-0.227	-0.482	-0.522
	<0.001	>0.05	***<0.05***	***<0.05***	>0.05	***<0.05***	***<0.05***
Cd	-0.795	0.102	-0.403	-0.545	-0.268	-0.221	-0.575
	<0.001	>0.05	>0.05	***<0.05***	>0.05	>0.05	***<0.05***
Pb	-0.73	0.071	-0.067	-0.649	0.056	-0.311	0.331
	<0.001	>0.05	>0.05	***<0.001***	>0.05	>0.05	>0.05

For Urban soils, in general, there is less effect of soil properties on metal lability, seen by comparing P-values evaluating the significance of correlation (Table 3-10). This implies that lability may be more affected by other factors including the source of contamination and contact time. Soil pH is the principal control over %M_E for all five metals, but was less strongly for Ni, Zn and Cd (k= -0.282, -0.567 and -0.633 respectively) than in the MA soils (k = -0.855, -0.813, and -0.795). Iron, Mn and Al oxides also have a weak influence on lability. Total Ni content is weakly correlated with lability and for Cu, there is a correlation between %M_E and Fe and Mn oxides which was also observed by Thornton et al. (2008), who used sequential extraction on a subset of the Urban soils used here and found a significant amount of Cu associated with the Fe and Mn oxides fraction. Only Pb correlated strongly with soil pH with a very small contribution from Fe oxide content (k= -0.291).

Table 3-10: Correlations between soil properties and %M_E for Urban soils. Significant values are shown in bold where P is <0.001 or <0.05

Urban soils	**pH**	**SOC**	**FeOx**	**MnOx**	**AlOx**	**Clay**	**Total metal content**
%M_E values		(%)	(g kg^{-1})	(g kg^{-1})	(g kg^{-1})	(%)	(mg kg^{-1})
Ni	-0.282	0.051	-0.164	-0.178	0.33	-0.024	0.371
	<0.05	>0.05	>0.05	>0.05	***<0.05***	>0.05	***<0.05***
Cu	-0.513	-0.162	-0.363	-0.32	0.116	0.183	-0.191
	<0.001	>0.05	***<0.05***	***<0.05***	>0.05	>0.05	>0.05
Zn	-0.517	0.151	-0.406	-0.262	0.301	-0.108	0.128
	<0.001	>0.05	***<0.05***	>0.05	***<0.05***	>0.05	>0.05
Cd	-0.633	-0.336	-0.248	-0.2	0.156	0.39	-0.231
	<0.001	***<0.05***	>0.05	>0.05	>0.05	***<0.05***	>0.05
Pb	-0.802	-0.041	-0.291	-0.273	0.271	0.064	-0.166
	<0.001	>0.05	***<0.05***	>0.05	>0.05	>0.05	>0.05

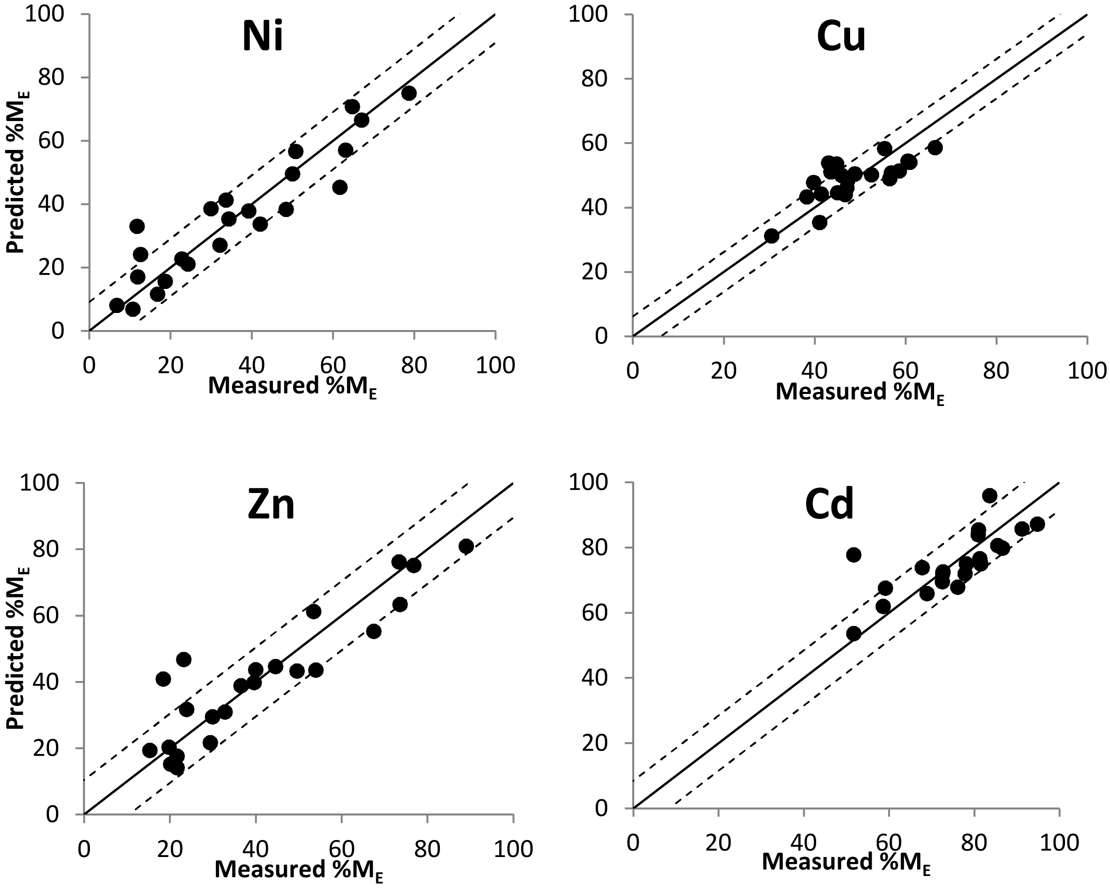
3.3.3.2 Multi-regression model:

As metal lability in the Urban soils are likely to have been affected by factors other than soil properties, especially metal source characteristics, the model constants (for Eq. 3-1) were parameterized on MA soils alone. The optimized constants, RSD and correlation co-efficient (R) are summarized in Table 3-11. Figure 3-7 compares modelled and measured %M_E values and shows a good prediction was achieved for Ni, Zn and Cd. Copper and Pb were less successfully modelled. For Cu, where lability is least affected by soil properties, this was expected. Another possible explanation is the absence of factors giving rise to strong Cu binding phases in the model, such as sulphide content for example (Du Laing et al., 2009). Lead is more likely to precipitate in non-labile mineral form in soils such as the widely reported chloropyromorphite ($Pb_5(PO_4)_3Cl$) (Lang and Kaupenjohann, 2003) and, again, its behaviour in soils is generally less well described by speciation models (Gustafsson et al., 2011). Previous studies have used linear regression in modelling metal solubility and often fail to adequately describe Pb behaviour in soils (e.g. Groenenberg et al., 2012).

Soil pH was negatively correlated with %M_E for all five metals and accounted for the largest proportion of the total variance in %M_E in the MA soils: 73.1%, 25.5%, 66.1%, 63.2% and 53.3% for Ni, Cu, Zn, Cd and Pb respectively (Table 3-11). Thus, soil pH appears to be the most important predictor of lability in agreement with the findings of other studies (e.g. Rieuwerts et al., 2006; Marzouk, 2012). Apart from soil pH, the significance of soil properties on predicting %M_E is metal specific. For example, the addition of FeOx improves the prediction for Ni and Zn. Although MnOx is correlated with the lability of all five metals, it only explains $\leq 0.4\%$ of total variance in predicting %M_E and the improvement observed for Ni, Zn and Cd by including MnOx in the model was therefore small. For Pb however, MnOx accounted for 8.2% of the total variance. The contribution from clay content was statistically significant ($p<0.05$) for Ni and Zn in the MA soils, however it only explained a very small proportion of the total variance in %M_E.

Table 3-11: Linear regression model coefficients, RSD and R-values for model prediction of %M_E. The regression (see Eq. 3-1) was parameterized from the %M_E (mg kg^{-1}) measured on MA soils. Values in brackets are the % of total variance. *N.S: not significant*

	Constant	**pH**	**FeOx (g kg^{-1})**	**MnOx (g kg^{-1})**	**AlOx (g kg^{-1})**	**Clay (%)**	**Total metal content (mg kg^{-1})**	**RSD**	**R**
Ni	161	-12.3 (73.1)	-0.49 (10.8)	-1.10 (<0.10)	*N.S*	0.04 (<0.10)	-0.49 (1.90)	9.05	0.93
Cu	76.3	-3.73 (25.5)	*N.S*	-2.61 (4.80)	-2.70 (24.7)	*N.S*	*N.S*	6.13	0.75
Zn	183	-13.4 (66.1)	-0.67 (5.5)	1.27 (<0.10)	*N.S*	-0.12 (<0.10)	-0.14 (10.9)	10.4	0.91
Cd	177	-9.62 (63.2)	*N.S*	2.22 (0.40)	*N.S*	*N.S*	-14.7 (7.70)	8.44	0.84
Pb	96.3	-6.30 (53.3)	*N.S*	-6.98 (8.20)	*N.S*	*N.S*	*N.S*	8.23	0.78



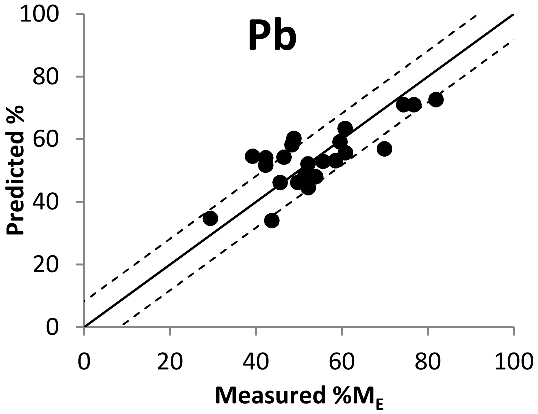


Figure 3-7: Comparison of measured and modelled %M$_E$ using a linear regression model (see Eq. 3-1) for MA soils. The solid line is the 1:1 line. Dashed lines represent ±1 RSD for the model fit. Optimized parameters are given in Table 3-11.

3.3.3.3 Logistic model

Although linear regression modelling can provide reasonable predictions of metal lability where $\%M_E$ is largely a function of soil characteristics and a single contaminant source predominates, as in the MA soils, the application of this model is limited to the range of data used to parameterise the model. Extending 'prediction' of lability outside this range can result in physically impossible outcomes such as $\%M_E$ being negative, exceeding 100% or having a positive value when the metal content is zero. A sigmoidal model based on a similar logistic function to that used to describe metal adsorption on variable charge surfaces against pH (Eq. 3-2) offers more realistic boundaries to model outcomes (i.e. values within 0 – 100%). The optimized values of pH_{50} and the exponential constants k_M are shown in Table 3-12. The sequence of optimized pH_{50} values was Cd > Pb > Cu > Zn > Ni, in agreement with previous general observations on the 'order' of metal lability (see Section 3.3.2.2). Thus, the results suggest that Cd is likely to be more labile than the other four metals as at a pH = 7.89, 50% of the Cd remains labile. The exponential factor k_M reveals the range of predicted $\%M_E$ with change in pH. Copper produced the lowest k_M value (0.16) so that Cu undergoes transition from 100% labile to a fixed form over a more restricted range of pH values, whereas Ni and Zn are labile over a broader range of pH values with k_M values equal to 0.76 and 0.72 respectively.

Inclusion of each of the adsorption phases (OM, Oxides, and clay) was then used to predict $\%M_E$ according to Eq. 3-3; optimized parameters are also summarized in Table 3-12, together with the values of RSD, correlation coefficient and $\Delta\%M_E$ evaluating the model prediction. For Ni, Zn and Cd, the best prediction, lowest RSD and highest correlation coefficients, was achieved by including total oxide content within Eq. 3.3 (S value) to control the spread of predicted $\%M_E$ values as a function of pH. For Cu and Pb, clay and organic matter content respectively improved the prediction. The values of optimized constants for these combinations are highlighted in the table. The prediction of $\%M_E$ is improved by including the effect of an additional adsorption phase for each metal with the most significant improvements in model fit for Ni and least for Cu. However, the improvement was relatively small when the RSD values were compared with those for the model solely based on pH implying that pH was still the most important property controlling lability in the MA soils. **Table 3-12:** Values of model parameters (pH_{50}, k_M/k_S, n), RSD, R values and $\Delta\%M_E$ (average deviation of predicted $\%M_E$ from measured) from sigmoidal model prediction of %E-value using Eq. 3-2 and Eq. 3-3 applied to MA soils only. The values in bold italic are the best fit of the measured $\%M_E$.

		Equation 3-2				
		Ni	**Cu**	**Zn**	**Cd**	**Pb**
	pH_{50}	5.06	5.55	5.36	7.89	6.33
	k_M	0.76	0.16	0.72	0.66	0.35
	RSD	11.5	7.59	13.2	8.73	8.97
	R	0.85	0.51	0.81	0.77	0.72
	$\Delta\%M_E$	-0.39	-0.01	0.01	0.13	0.12

		Equation 3-3				
		Ni	**Cu**	**Zn**	**Cd**	**Pb**
pH+%SOC	**pH_{50}**	4.94	5.17	5.02	7.59	***6.27***
	k_S	0.54	0.07	0.23	0.38	***0.12***
	n	0.28	0.57	0.95	0.86	***0.86***
	RSD	11.2	7.66	13.3	11.5	***8.53***
	R	0.86	0.50	0.81	0.65	***0.76***
	$\Delta\%M_E$	0.18	0.66	1.43	0.37	***0.74***
pH+Ox	**pH_{50}**	***4.99***	5.47	***5.29***	***8.03***	6.43
	k_S	***0.16***	0.06	***0.12***	***1.19***	0.08
	n	***0.83***	0.57	***0.95***	***-0.40***	0.86
	RSD	***10.4***	7.45	***12.3***	***8.06***	8.84
	R	***0.88***	0.53	***0.84***	***0.81***	0.73
	$\Delta\%M_E$	***0.15***	0.40	***1.08***	***0.06***	0.03
pH+%Clay	**pH_{50}**	5.01	***5.70***	5.16	7.22	6.46
	k_S	0.20	***0.03***	0.03	0.10	0.02
	n	0.41	***0.57***	0.95	0.86	0.86
	RSD	10.0	***7.17***	14.5	11.8	11.4
	R	0.89	***0.58***	0.77	0.65	0.51
	$\Delta\%M_E$	1.05	***0.12***	2.75	0.19	2.04

Measured values of $\%M_E$ for Ni, Cu, Zn, Cd and Pb as a function of soil pH in both MA and Urban soils are shown in Figs. 3-8 to 3-12 alongside the range of the predicted values presented using either the mean plus or minus one standard deviation values of the adsorption phases measured for the MA soils (i.e. oxide content (g kg^{-1})) for Ni, Zn and Cd; %Clay content for Cu and %SOC content for Pb. Predicted values of $\%M_E$ for the individual MA soils are also shown for direct comparison between predicted and measured values of $\%M_E$. The measured (a) and predicted (b) values for the Urban soils are shown for comparison. The latter were predicted from the model parameterised using the MA soils; no attempt was made to parameterise the model (Equations 3-2 and 3-3) using the Urban soils.

In MA soils, for all five metals, the model generally underestimated lability ($\%\Delta M_E > 0$) but most measured values were within the range of predicted values. Underestimation was observed in the low to mid pH range, and overestimation in the high pH range for Zn. Both measured and predicted Cu labilities were weakly correlated with pH over a very restricted range and the difference in $\%M_E$ predicted between pH 3.6 and 7.12 was <25%. Lead lability was predicted well for low pH soils, but the measured values were more scattered in the high and neutral pH soils.

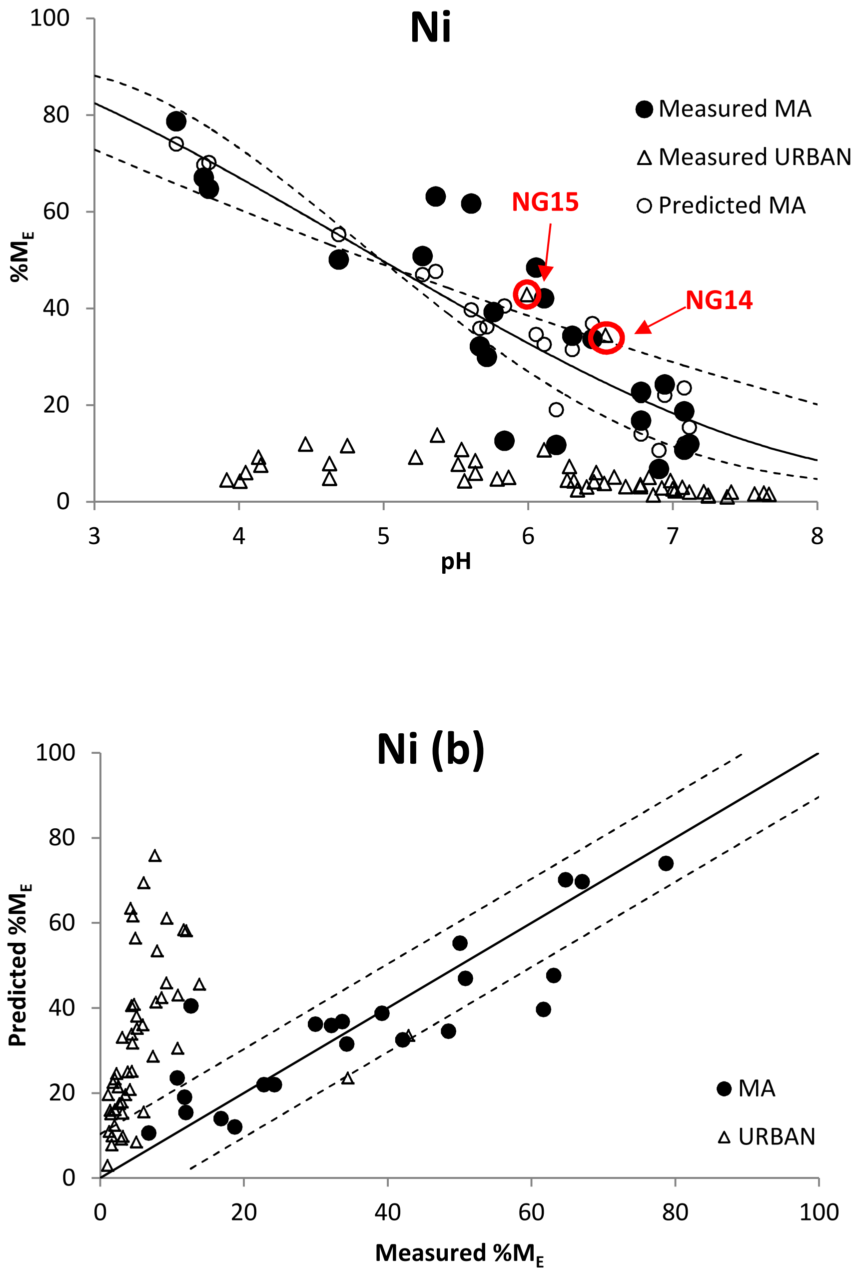
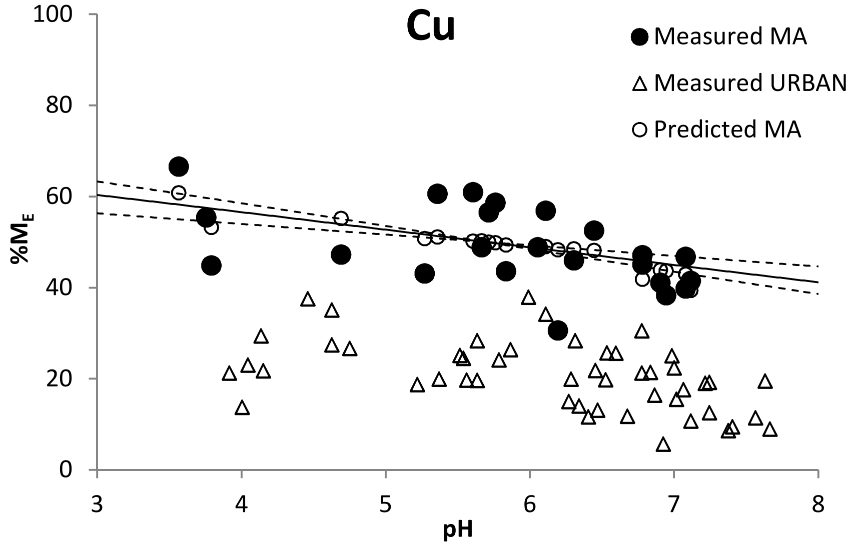


Figure 3-8 (a): Measured values of $\%M_E$ (Ni) for MA (●) and Urban (Δ) soils as a function of soil pH. The solid line represents the predicted $\%M_E$ using mean oxides contents (g kg^{-1}); while the dashed lines represent the range of predicted $\%M_E$, using mean ± SD oxides contents (g kg^{-1}); **(b)** Comparison between measured and modelled $\%M_E$ (Ni) from Eq. 3-3 for MA and Urban soils. The solid line is the 1:1 line. The dashed line represents ±1 RSD for the model fit.



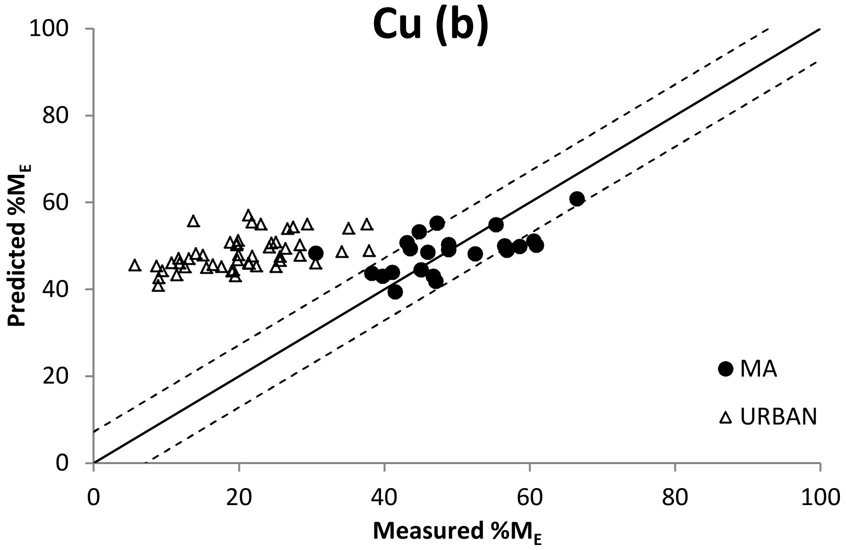


Figure 3-9 (a): Measured values of %M_E (Cu) for MA (●) and Urban (Δ) soils as a function of soil pH. The solid line represents the predicted %M_E using mean oxides contents (g kg^{-1}); while the dashed lines represent the range of predicted %M_E, using mean ± SD oxides contents (g kg^{-1}); **(b)** Comparison between measured and modelled %M_E (Cu) from Eq. 3-3 for MA and Urban soils. The solid line is the 1:1 line. The dashed line represents ±1 RSD for the model fit.

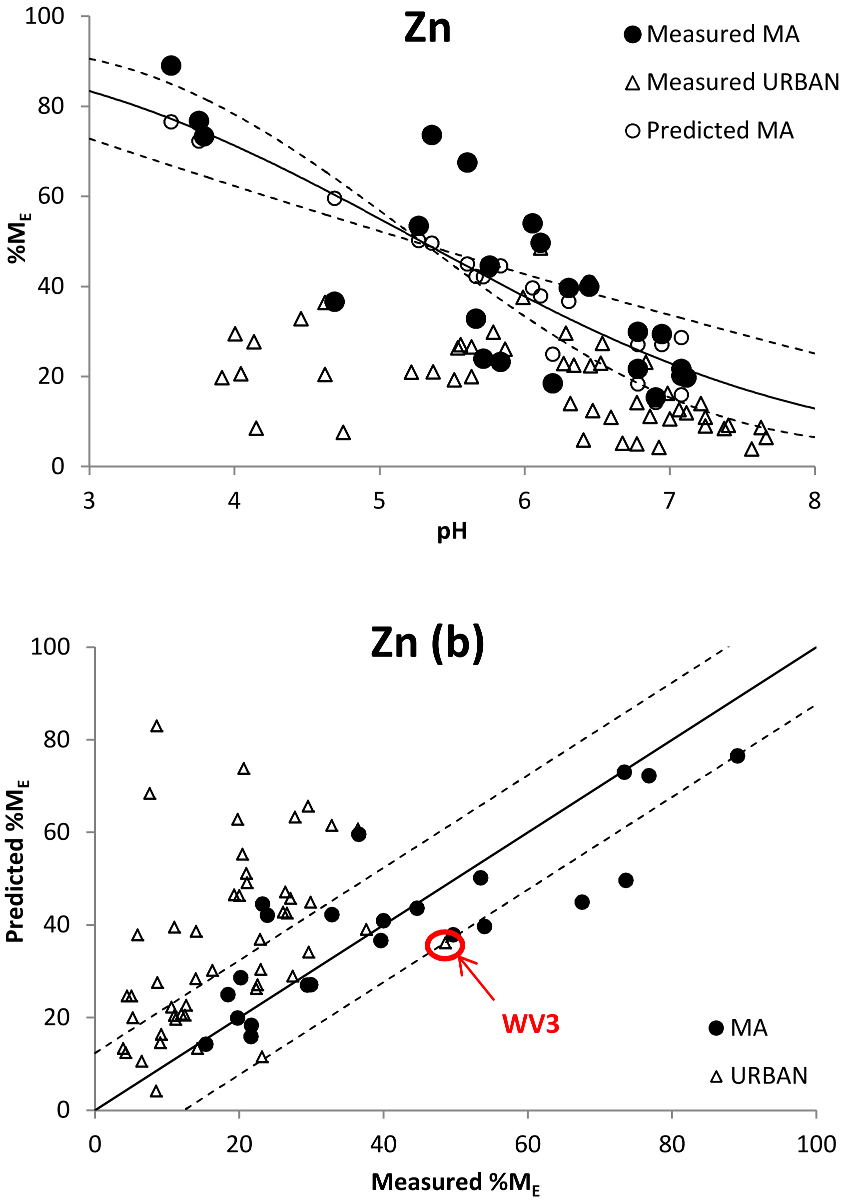
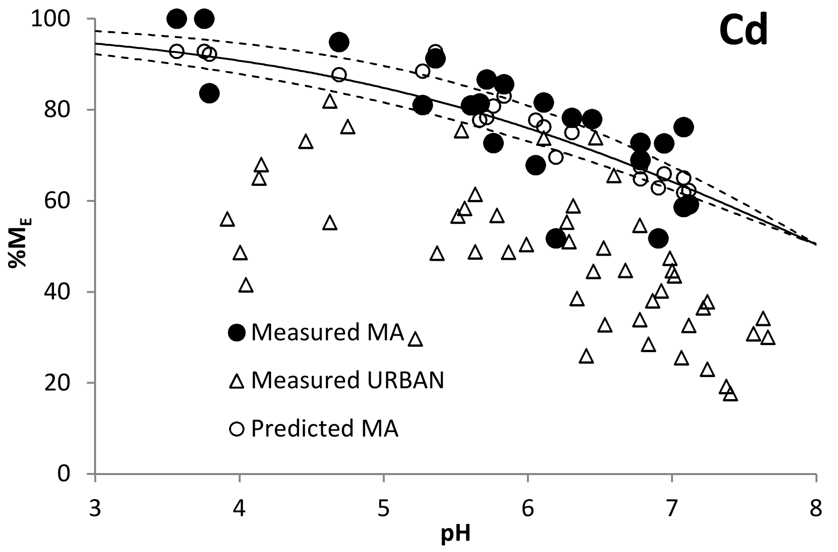


Figure 3-10 (a): Measured values of %M_E (Zn) for MA (●) and Urban (Δ) soils as a function of soil pH. The solid line represents the predicted %M_E using mean oxides contents (g kg^{-1}); while the dashed lines represent the range of predicted %M_E, using mean ± SD oxides contents (g kg^{-1}); **(b)** Comparison between measured and modelled %M_E (Zn) from Eq. 3-3 for MA and Urban soils. The solid line is the 1:1 line. The dashed line represents ±1 RSD for the model fit.



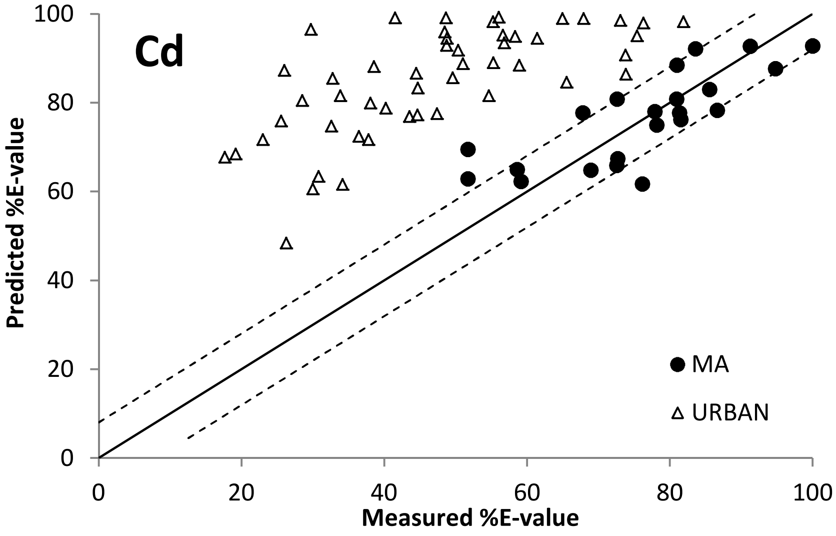


Figure 3-11 (a): Measured values of %M_E (Cd) for MA (●) and Urban (Δ) soils as a function of soil pH. The solid line represents the predicted %M_E using mean oxides contents (g kg^{-1}); while the dashed lines represent the range of predicted %M_E, using mean ± SD oxides contents (g kg^{-1}); **(b)** Comparison between measured and modelled %M_E (Cd) from Eq. 3-3 for MA and Urban soils. The solid line is the 1:1 line. The dashed line represents ±1 RSD for the model fit.

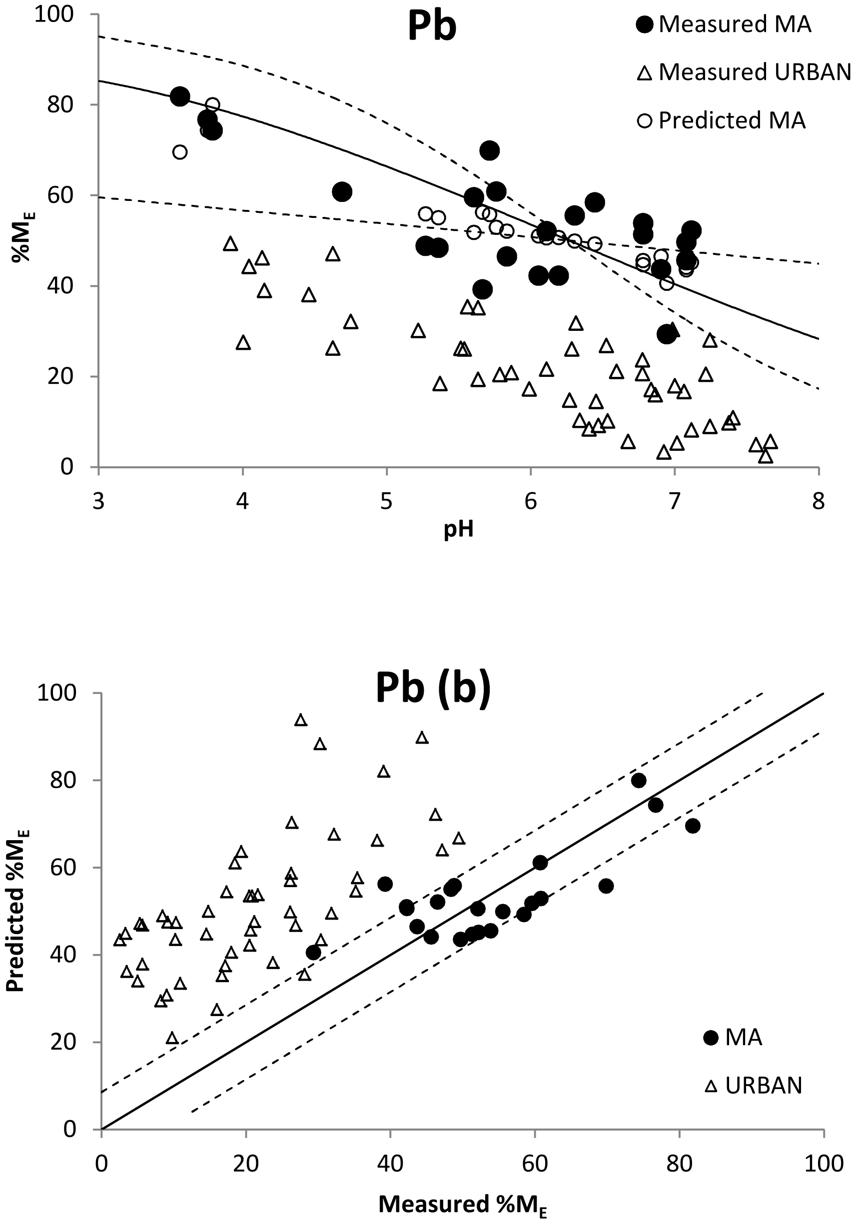


Figure 3-12 (a): Measured values of %M$_E$ (Pb) for MA (●) and Urban (Δ) soils as a function of soil pH. The solid line represents the predicted %M$_E$ using mean oxides contents (g kg^{-1}); while the dashed lines represent the range of predicted %M$_E$, using mean ± SD oxides contents (g kg^{-1}); **(b)** Comparison between measured and modelled %M$_E$ (Pb) from Eq. 3-3 for MA and Urban soils. The solid line is the 1:1 line. The dashed line represents ±1 RSD for the model fit.

The values of pH_{50} optimized using Eq.3-3 were similar to those for Eq.3-2 and followed the observed sequence of relative metal lability in soils (Table 3-12). The involvement of oxides in controlling exponential factors for Ni, Zn and Cd is in line with observations reported in previous studies co-precipitation (Zwolsman et al., 1993; Du Laing et al., 2009) or fixed into mineral oxides as discussed in Section 3.3.3.1. Inclusion of %SOC or oxides as the S-variable (Eq. 3-3) can both improve the prediction of $\%Pb_{E,}$ as the RSD value decreased from 8.97 using Eq. 3-2 to 8.53 for %SOC and 8.84 for oxides. Strong adsorption of Pb on organic matter in soils was often found in modelling practices for Pb solubility (Gustafsson et al., 2011; Guo et al., 2006). However, the effect of oxides on Pb may be diminished as a consequence of combining the three oxides phases (Fe, Mn and Al), as Pb tends to bind more to MnOx in the low and mid pH range with increasing binding on FeOx at high pH. The very low value of k_S for %Clay in predicting $\%M_E$ for Cu (0.03) implies the effect of clay is relatively insignificant, as expected.

Overall, the logistic model gives a reasonable prediction of $\%M_E$ for metals in MA soils from a very limited number of soil properties and fitted constants. Although the model performance is poorer than the fit achieved with multiple linear regression (Eq. 3-1) (except for Cd (lower RSD values) the increase in RSD values are 1.27, 1.05, 1.90 and 0.30 for Ni, Cu, Zn and Pb respectively) it produced results that are understandable in terms of the mechanisms known to control trace metal lability and is perhaps a more robust model for predicting metal lability from soil properties for reasons discussed above.

Attempts were also made to predict metal lability in the Urban soils using the logistic model after parameterization using the MA soils (Eq. 3-3; Table 3-12). As expected, the model grossly over-predicted values of $\%M_E$ for all five metals (Figs. 3-8 to 3-12).

As discussed previously, this is a consequence of both longer soil-contaminant contact times (Tye et al., 2003; Buekers et al., 2007) and the form of the trace metals originally added to the soils. Soil pH barely affected the lability of Ni (Fig. 3-8a), Cu (Fig. 3-9a) and Zn (Fig. 3-10a) in the Urban soils and the lability was greatly overestimated in soils with low pH values.

Nickel

For Ni (Fig. 3-8), soils with high pH values produced reasonable predictions of %M_E but this may be a result of low predicted lability coincidently agreeing with measured values. The most significant overestimation of %M_E occurred in soils with low pH values (< 5.0). Low total Ni concentrations were found in these soils (mean = 38.5 mg kg^{-1}; SD = 18.4 mg kg^{-1}), which indicated that Ni in these soils may be attributable to parent material (see Section 3.3.2.2). Two soils, both from a sewage farm (NG14 and NG15; Table 3-3b), with the highest %M_E (34.4% and 42.9%, respectively), fell within the range of predicted values (Fig. 3-8a). This may suggest that Ni lability is more affected by soil properties in those two soils than in the other Urban soils. A similar contrast was found by Degryse et al. (2004) studying Cd and Zn lability in sludge-amended soils where a negative correlation was found between pH and %M_E. However, they also suggested that the correlation between metal lability and other soil properties, such as organic matter content, may only be a coincidence, as the organic matter was a co-contaminant with metals in sewage sludges. Thus, a negative correlation was observed between organic carbon and metal lability which was initially low in sewage sludges. It can therefore be concluded that metal lability may be more related to characteristics of the sludge than properties of the soil (Stacey et al., 2001), but often coincidental correlations are observed between lability and soil properties.

Copper

Predicted Cu lability values (Fig. 3-9) (%M_E, mean = 48.3) were much larger than measured (%M_E, mean=20.5) but with a very restricted range (SD=3.9%). The measured lability also tended to be more variable than the range predicted by the MA-parameterised model. In contrast to the results for Ni, there was a systematic displacement in the predicted compared to the measured values across the whole pH range (Fig. 3-9a), rather than a convergence at higher pH values. The model overestimation of %M_E was more obvious for soils with low metal lability but this applied across the full pH range studied. However, it was difficult to identify any specific effects of landuse (or historical landuse) on Cu lability as, for example, soils from different brownfield sites produced values of %M_E from 5.6% (NG19) to 30.5% (NG18).

Zinc

For Zn, the model gave a better prediction than seen for Cu and Ni, with about half of the samples (n=22) within the range delineated by the ± RSD limits (12.3%) for the model fit to the MA soils. Generally, Zn lability was overestimated (Fig. 3-10), especially for soils with low pH values. The average deviation from the model for the 9 soils with pH < 5.0 was 44% between measured and predicted (SD = 16.3%). However the reasons that lead to the bias may be different from those for Ni, as total Zn concentrations (mean = 283 mg kg^{-1}; SD = 178 mg kg^{-1}) in these soils were generally higher than the background level of Zn in soils from England and Wales (Rawlins et al., 2012). Therefore it is expected that the urban soils had received Zn from sources other than parent material. Again, it is difficult to summarize the effect of landuse on metal lability for the whole dataset because of the complex range of Zn sources; for example, from field notes, metals in soil WV20 were probably associated with canal dredgings whereas soil WV25 was contaminated from its location in an industrial estate and proximity to a railway, both had similar Zn lability of 27.7% and 20.6% respectively. There was only one sample where Zn lability was, unexpectedly, underestimated by the MA-parameterised model and fell slightly outside of the RSD range: soil WV3 is an acidic woodland soil (pH = 6.1) with high DOC concentration (180 mg L^{-1} in 0.01 M $CaCl_2$, 1g: 30 mL).

Cadmium

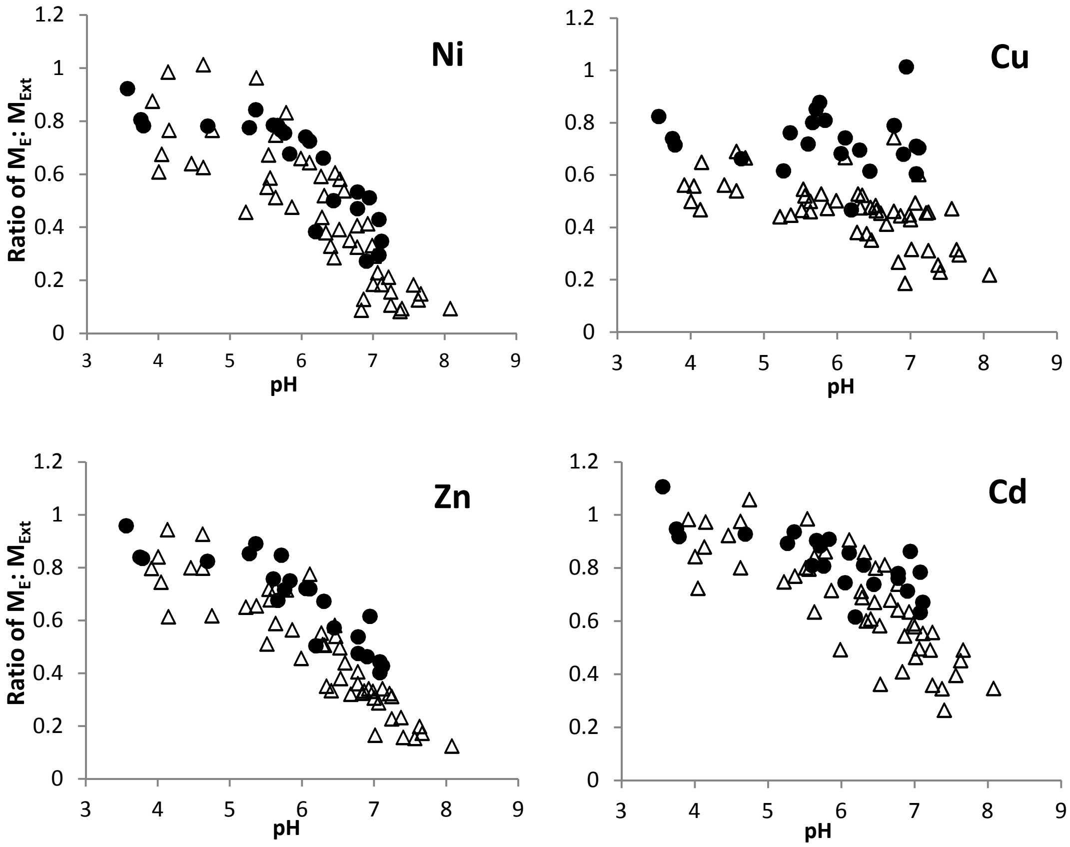
Cadmium lability for the Urban soils (Fig. 3-11a) generally followed the same trend with pH as the MA soils but, like Cu, is displaced fairly uniformly across the pH range. The smallest bias was observed in a small groups of soils (n=5, 10% of Urban soils dataset) with high measured Cd lability (Mean = 74%, SD = 5.8%). Although Cd is likely to remain more labile in contaminated soils compared to the other four metals (Izquierdo et al., 2013; Esnaola and Millan, 1998), there was a clear difference between the Urban and MA soils (for 90% of the Urban soils). For example soil WV14 was collected from a site adjacent to a tyre factory with a high total Cd concentration (4.1 mg kg^{-1}) and with a measured lability of 50.4%, less than the predicted value. A similar conclusion regarding Cd lability in field contaminated soils was also made by Degryse et al. (2004) comparing radio-labile Cd in ‘metal salt incubated soils’ and soils sampled adjacent to a smelter.

Lead

Figure 3-12b shows a reasonably strong correlation between measured and predicted Pb lability (R=0.67) which suggests that the lability of Pb in Urban soils may be more affected by soil properties than the other four metals. However, there was also a large average displacement of 30.1%, from the model trend which suggests that the Pb added to these soils was originally in a relatively non-soluble form. An alternative explanation for the substantial displacement of %M_E from the model is the secondary formation of poorly soluble Pb minerals (e.g. cerussite, $PbCO_3$; chloropyromorphite, Pb phosphate minerals) that lower the overall lability in the soil. For example, the average measured lability of Pb in the phosphate-rich sewage farms soils (13.8%) was much less than that predicted by the MA soil parameterised model (50.0%) suggesting precipitation of Pb phosphate minerals either in the soil or during the processing of the sewage sludge.

3.3.4 Comparison between E-values and 0.43M HNO_3 extractable metal

A single extraction using 0.43 M HNO_3 (M_{Ext}) has been widely used to determine the *reactive* metal fraction in soils (Tipping et al., 2003; Groenenberg et al., 2010). However, it has also been criticized in that the capacity and efficiency of a single chemical extraction procedure can be strongly influenced by soil characteristics (Ayoub et al., 2003). Differences between M_E and M_{Ext} are shown in Fig. 3-13 as a function of pH and demonstrate that the labile metal fraction determined by 0.43 M HNO_3 extraction is often larger than that using the ID method (ratio < 1). For all five metals, the deviation between M_E and M_{Ext} was strongly related to pH with divergence greatest in high pH calcareous soils when the acid is likely to mobilize the non-labile metal fraction or dissolve carbonate binding phases (Degryse et al., 2009).



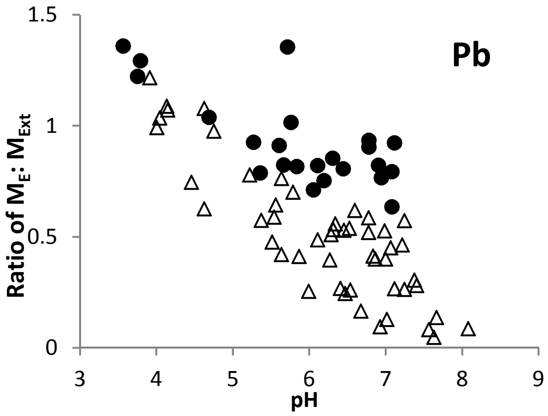


Figure 3-13: Ratio of E-values (M_E): HNO_3 extractable (M_{Ext}) as a function of pH in MA (●) and Urban (Δ) soils.

The general trend with pH, for both MA and Urban soils, was similar for Ni, Zn and Cd. The MA soils were generally closer to a ratio of 1.0 across the pH range and the displacement from the Urban soils was again similar for Ni, Zn and Cd in this regard. This is the expected pattern as the Urban soils present a greater opportunity for mobilization of non-labile metal by acid extraction and the MA soils have a clearer distinction between labile and non-labile with the soil metal loading consisting of recently added metal salt and metal bound into primary minerals. For Cu and Pb the trend was less distinct and the displacement of MA soils from Urban soils was greater and, for the MA soils, ratios were closer to 1.0 than seen for Ni, Zn and Cd. All three observations are consistent with a greater range of non-labile Cu and Pb forms in the Urban soil. For Cu, the difference in reactive pool measured by the two methods was less strongly correlated with pH compared with the other four elements. As discussed in Section 3.3.3.1, %M_E for Cu was least affected by soil pH. For Ni, Zn and Cd in the MA soils, the difference between the M_E and M_{Ext} was less significant at soil pH values < pH 6. Marzouk et al. (2013) also found that there was a good agreement between M_E and M_{Ext} for Zn and Cd for soils with pH < 6.4. For Pb in some of the very acid soils (pH < 4), %M_E was actually larger than %M_{Ext} as also observed by Marzouk et al. (2013). This may be explained by the fact that the relatively short time for equilibration when using the nitric acid extraction is not long enough for all labile Pb to be extracted from the soil binding phases.

The ‘error’ produced by using M_{Ext} as an estimate of M_E was substantial – especially at high pH values. For some high pH soils, isotopically exchangeable Ni and Pb was less than 10% of M_{Ext}. In general, the error should be greatest when there is a relatively low labile metal concentration as such conditions provide the greatest potential for mobilization of non-labile metal. Results confirm that a single extraction using 0.43 M HNO_3 is suitable for use only in acidic organic soils and highly misleading when applied to calcareous soils (Rodrigues et al., 2010).

3.3.5 Change in E-values over time

The lability of Zn and Cd in MA and Urban soils was previously measured by isotopic dilution using the radio-isotopes ^{65}Zn and ^{109}Cd (Tye et al., 2003). Subsequently the soils were stored dry for about 10 years, although it should be noted that the soils were maintained in a moist condition for several months following the final measurement of E-value by Tye and co-workers. A paired t-test showed that for MA soils, the difference between M_{Rad} (lability measured using radio-isotopes) and M_E was significant for Zn ($p = 0.007$), but not for Cd ($p=0.416$). For the Urban soils, however, there was a significant difference in both Zn and Cd ($p<0.001$) with generally $\%M_{Rad} > \%M_E$ (Fig. 3-14a). This difference may be a consequence of processes occurring within the soils or it may be the product of artefacts within the two methods. Only a limited number of studies have compared the measurement of metal lability using both radio-isotopes and stable isotopes. Sterckeman et al. (2009) compared Cd lability measured using ^{111}Cd (stable; M_E) and ^{109}Cd (radioactive; M_{Rad}) and found that the results were equivalent but that M_E data were more repeatable. Considering the history of the MA soils, continuing fixation of Zn may have occurred. It is notable, for example, that the difference in M_E and M_{Rad} was more pronounced in high pH soils than in acidic soils (Fig. 3-14b). Even in air dried soils, particles may be covered in thin films of water which could mean that the soil remains sufficiently chemically reactive to allow solid phase matrix and surface diffusion processes. A relatively small difference between M_E and M_{Rad} was observed in MA soils (c. $M_E : M_{Rad} = 0.93$ and 1.00 for Zn and Cd respectively) compared with Urban soils (c. $M_E : M_{Rad} = 0.71$ and 0.69 for Zn and Cd). This was surprising as the contact time of metal in historically contaminated Urban soils was certainly much longer than metal incubated MA soils, therefore less fixation would be expected in Urban soils during

10 years of storage afterwards. Potential explanation include the existence of non-labile Zn and Cd present in the colloidal phase > 0.2 µm; as in Tye et al. (2003), direct measurement of M_{Rad} was made on samples after solid and solution were separated but without 0.2 µm filtration. Therefore, overestimation of Zn and Cd lability could have occurred especially for soils with low metal lability i.e. high pH MA soils and the majority of Urban soils (see Section 3.2.2.3). Also the relatively low metal solution concentrations, close to the detection limit of flame atomic absorption spectrophotometry (FAAS) or graphite furnace atomic absorption spectrophotometry (GFAAS), in most Urban soils and high pH MA soils may result in analytical problems in measuring Zn and Cd concentration in soil suspension when measuring M_{Rad}.

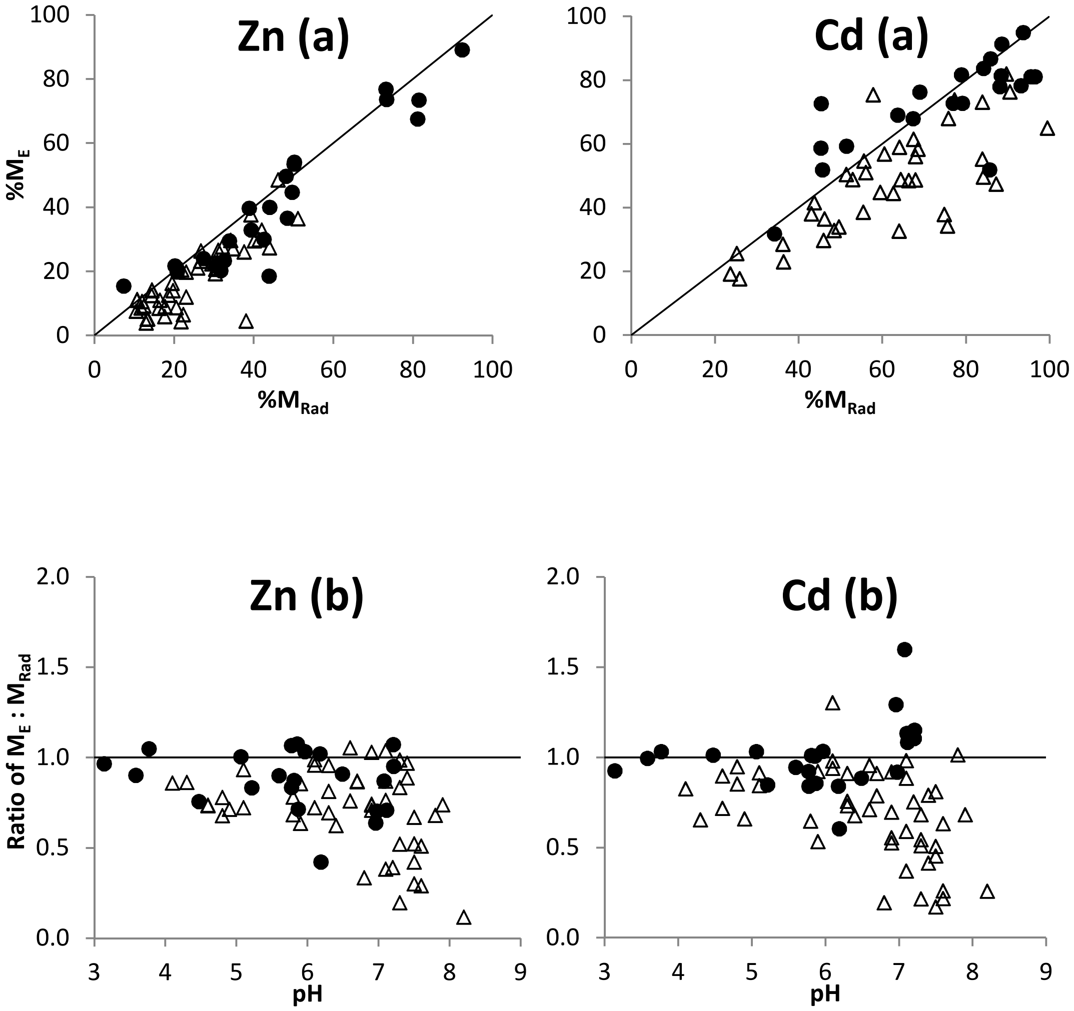


Figure 3-14: **(a)** Comparison of %M_E (this study) and %M_{Rad} (Tye et al., 2003) for Zn and Cd in MA (●) and URBAN (Δ) soils. The solid line is the 1: 1 line; **(b)** The ratio of M_E : M_{Rad} as a function of pH.

3.4 Conclusion

The results from experiments on metal salt amended and urban contaminated soils suggests that both soil properties and sources of contamination can affect the trace metal lability and the effect is metal specific. In MA soils, soil pH was the most important factor in controlling the lability for all five metals, and the effect was least significant for Cu. The logistic model can well describe the relationship between metal lability and soil properties explaining underlying mechanisms. The model indicates that mineral oxides tend to be important fixation phases for Ni, Zn and Cd; while Pb may also be strongly affected by organic matter in soils. However, such a model is not applicable for Urban soils as the source of contamination determines the lability and it is likely that metal in those soils was much less soluble than metal salts and resulted in much lower lability than in MA soils.

Single extraction by 0.43 M HNO_3 generally overestimates the lability, and the bias is pH dependent. In high pH calcareous soils, $\%M_{Ext}$ can be 10 times higher than the $\%M_E$ for Ni and Pb. This confirms that acid extraction method is only justifiable for acid organic rich soils. Difference in E-values measured in this study and that of Tye et al. (2003) was observed for Zn and Cd. Although soils were stored dry during these years, pH dependent fixation is still likely to taken place in these soils.

4 CHAPTER 4: PREDICTING THE SOLUBILITY AND SPECIATION OF TRACE METALS IN METAL-AMENDED AND URBAN SOILS

4.1 Overview:

It is generally accepted that solid↔solution partitioning, and speciation in the solution phase, largely determine the bioavailability and mobility of trace metals in soils. Various studies have modelled trace metal solubility, including solution concentration, free ion activity and speciation in soils, and used the outcomes to predict fate in the environment (e.g. Tipping et al., 2006; Bonten et al., 2008; Thakali et al., 2006). Free ion activity in solution may represent the most toxicologically relevant parameter for metal contaminated soils (Di Toro et al., 2001; Thakali et al., 2006) and has been successfully applied to predict the variation in trace metal concentration in plants (e.g. grass) in a range of soils (Hough et al., 2005). Total concentration in solution, however, will provide a better estimation of metal mobility in soils.

In modelling trace metal solubility in soils, both empirical equations and mechanistic geochemical speciation models have been developed. The empirical approach tries to relate the partitioning of metals to soil properties like soil pH and SOM. It has frequently been found to provide a simple but robust means of predicting solution concentrations of most of the divalent trace metal cations, including Ni, Cu, Zn, Cd and Pb, and is often found to be reasonably consistent across a range of soil types and conditions (Groenenberg et al., 2012; De Vries et al., 2008; Bonten et al., 2008). However, the limitation of such an approach has also been recognized. For example, Elzinga et al. (1999) established an extended Freundlich type model based on a large number of datasets from published laboratory incubation experiments. They found that, although a consistent model could be produced from the combined datasets, there were nevertheless problems when applying the parameterised model to field-derived metal solubility data. By contrast, mechanistic geochemical speciation models should be more reliable across a wider range of conditions and should not require re-parameterisation for individual datasets. Although more demanding in terms of input variables, mechanistic geochemical models also provide more comprehensive output information, such as speciation (solution phase) and fractionation (solid phase) which may help in assessing biological hazards and metal mobility. However, these models require some soil variables which are difficult to measure and even to define in some cases. For example, to predict metal solubility, the 'reactive' metal fraction in soil must be determined (Buekers et al., 2008b); 'active' adsorption phases must be estimated including the fraction of organic matter content and mineral (hydr)oxides involved in metal adsorption reactions (Buekers et al., 2008b; Weng et al., 2001). Poor estimates of these ill-defined variables may lead to large errors in model predictions of solubility.

In this chapter, the use of both empirical and mechanistic geochemical models were assessed by predicting Ni, Cu, Zn, Cd and Pb solubility in metal-amended (MA) soils and Urban soils from Nottingham and Wolverhampton (Urban). The origins of these two sets of soils, and their characteristics, have been described in Chapters 2 and 3. The empirical model used was based upon an extended Freundlich equation, adapted to predict free ion activity (Tye et al., 2003; Tye et al., 2004; Tipping et al., 2003; Groenenberg et al., 2010; Ivezic et al., 2012; Elzinga et al., 1999, etc). The geochemical speciation model, WHAM-VI, has been applied in a limited number of investigations of metal fractionation and solubility in whole soils with promising results (Tipping et al., 2003; Smith et al., 2004; Shi et al., 2008 etc). To investigate the origins of errors and bias in the prediction of metal solubility the modelled fractionation of trace metals in the solid phase was also investigated. The WHAM model requires an estimate of the total concentration of 'reactive metal' in soils, across all adsorbed phases. Therefore E-values (M_E) were used to represent labile metal and compared with 0.43 M HNO_3-extractable metal (M_{Ext}), which has also been widely used in this context. Finally, comparison was made between the results from WHAM-VI and WHAM-VII to assess the value of recent attempts to improve the model (Tipping et al., 2011).

4.2 Modelling Free Ion Activity (FIA) in solution using an extended Freundlich equation

The empirical model was based on an extended Freundlich equation (Eq. 4-1a) to predict the free ion activity of Ni^{2+}, Cu^{2+}, Zn^{2+}, Cd^{2+} and Pb^{2+} in the soil solution (Eq. 4-1b), adapted from Tye et al. (2003). It takes into account the labile adsorbed trace metal concentration in soil, the effect of soil pH (Section 2.1.1), and organic matter content (%SOC, Section 2.1.2) as one of the solid phases that adsorbs trace metals in a labile form. The predicted free ion activities of metal ions in the whole soil suspensions were compared with 'observed' values which were calculated using WHAM-VII solely from solution concentrations (Section 2.2.5). Details of the (WHAM) model settings and variables are given in the following section.

$$\mathrm{Log}_{10}\left(\frac{M_{Lab}}{(M^{2+})^{n}}\right) = k_o + k_{pH}(pH) + k_C\,(\%SOC) \qquad (4\text{-}1a)$$

$$p(M^{2+}) = \frac{k_0 + k_{pH}(pH) + k_c(\%SOC) - \log\,[M_{Lab}]}{n_F} \qquad (4\text{-}1b)$$

In Eq. 4-1b, $p(M^{2+})$ is the free ion activity in $-\log_{10}$ scale; $[M_{Lab}]$ is labile trace metal concentration measured by either ID with the chelex resin purification step (M_{Er}, Section 2.3.3) or 0.43 M HNO_3 extraction (M_{Ext}, Section 2.2.4), (mol kg^{-1}); (M^{2+}) is the free metal ion activity (mol L^{-1}); n_F is the power term constant and the constants k_o,

k_{pH} and k_C are empirical coefficients. Optimization was undertaken using the data analysis tool 'Solver' in Microsoft Excel to find the lowest residual standard deviation (RSD) comparing measured and predicted values of free ion activity.

4.3 WHAM modelling

Predicting trace metal solubility and speciation

WHAM-VII was used to predict Ni, Cu, Zn, Cd and Pb concentration in the solution phase of soil suspensions. Soil was suspended and shaken for 3 days in 0.01 M $Ca(NO_3)_2$ (solid: solution ratio 1 g in 30 mL) to reach equilibrium. The measured E-values without resin purification (M_E) were used as the reactive trace metal fraction, as the non-labile SCP-metal also contributes to the total solution concentration (see Section 3.3.2.3). The modelled metal concentration in solution was compared with measured values (Section 2.2.5) to assess the model performance. Speciation and fractionation information was given by the model output. The metal solubility data predicted by WHAM-VII and WHAM-VI were compared. The model variables and settings are listed in Table 4-1.

Modelling free ion activity

The 'observed' free ion activities of Ni^{2+}, Cu^{2+}, Zn^{2+}, Cd^{2+} and Pb^{2+} (mol L^{-1}) were calculated from WHAM-VII to compare with the results predicted by the empirical model (Eqs. 4-1a & b). Instead of using 'total reactive trace metal concentration' as an input, only measured solution concentrations were used for modelling metal speciation in solution. All the other model set-up options were the same as for modelling solubility and speciation in whole soil suspensions (Table 4-1).

Table 4-1: Summary of the variables, parameters and settings of WHAM-VII and VI in modelling trace metal solution concentration, speciation and fractionation.

Modelling trace metal solubility and speciation

Variables	Settings
Suspension condition	
Suspended particulate matter (SPM)	33.33 g L^{-1}; converted from 1g solid in 30 mL of electrolyte
Temperature (K)	288.15 K; 15°C
PCO_2 (atm)	Measured bicarbonate concentration (DIC)
Soil pH	Measured in 0.01 M $Ca(NO_3)_2$ soil suspensions.
Charge balance options	No charge balance imposed, i.e. a fixed (measured) pH value used
Adsorption phase (g L^{-1}):	
Clay content	MA soils: estimated from soil texture. Urban soils: measured by laser granulometry (Section 2.1.3)
Iron, Al and Mn oxides	DCB extraction (Section 2.1.4); converted to $Fe_2O_3.H_2O$, Al_2O_3 and MnO_2
Humic and fulvic acid	Measured by alkaline extraction (Section 2.1.2.2)
Colloidal fulvic acid	Estimated from measured DOC (Section 2.1.5)
Major cation and anion concentration (mol L^{-1})	
Dissolved major cations (Na, Mg, Al, K and Ca)	Concentration measured in filtered supernatant solutions of 0.01 M $Ca(NO_3)_2$ soil suspension (Section 2.2.5); *Precipitation option for Al (WHAM-VII only):* One mol of precipitated $Al(OH)_3$ forms 87 g of the binding phase within the particulate soil phase (S. Lofts., personal communication).
Fe^{3+} activity	*WHAM-VII: Precipitation option for Fe* assumes that Fe^{3+} activity is controlled by $Fe(III)(OH)_3$ solubility; calculated within the model. *WHAM VI:* The Fe^{3+} activity is estimated from $Log(Fe^{3+})=3pH+logK_{sp}$; Log K_{sp}=2.7 calculated externally to the model (e.g. Van Laer et al., 2006).
Dissolved nitrate (NO_3^-)	Solution concentration estimated as 0.02 M in the suspension
Total dissolved carbonate (all species)	Estimated from total inorganic carbon measured in solution (Section 2.1.5)
Trace reactive metal concentration in soil suspension (mol L^{-1})	
Total concentration Ni, Cu, Zn, Cd and Pb	Estimated from E-values (M_E) or 0.43 M HNO_3 extractable (M_{Ext})
Other settings:	
Activity coefficient correction	Debye-Hückel method
WHAM parameter data sets	Default master, solute and binding phase data bases were used in both WHAM VI and VII.

Modelling free ion activity ('observed FIA')*

Variables	Settings
Trace metal concentration (mol L^{-1})	
Solution concentration Ni, Cu, Zn, Cd and Pb	Measured in the filtered supernatant solution of soil suspensions in 0.01 M $Ca(NO_3)_2$; solid solution ratio was 1g: 30mL

*Other variable and settings were the same as above but with the omission of particulate binding phases.

4.4 Results and discussion

4.4.1 Empirical model of Free Ion Activity (FIA) in solution

The free ion activity for both sets of soils predicted from Eq. 4-1, were compared with the 'observed' FIA from WHAM-VII speciation of solution concentrations. The optimised coefficients (nF, k_o, k_{pH}, k_C) are listed in Table 4-2. Close correlations were found for all five metals (R values > 0.90), implying that the FIA is strongly controlled by the reactive metal fraction and soil pH. The coefficient for pH (k_{pH}) indicated a positive effect on $p(M^{2+})$, i.e. a higher pH leads to a decrease of trace metal FIA in solution. A negative effect from %SOC was only found for Pb^{2+} free ion activity, when soil organic matter apparently limited Pb^{2+} solubility. However, the effect of %SOC is likely to be very small (k_c = -0.002). Small positive k_c values for the other 4 metals indicated that inclusion of %SOC did not significantly improve the prediction of $p(M^{2+})$. The lack of significance of %SOC may be the result of relatively low and small range of organic matter content in both sets of soils (average %SOC for MA = 2.4%, SD= 1.7% and Urban = 7.9%, SD= 4.8), which was also found in a study for lead by Sauve et al. (1997).

Table 4-2: Coefficients for the best-fit solubility models (Eq. 4-1a, b) using, pH, %SOC and M_{Er} or M_{Ext} as determining variables. Model residual standard deviation values (RSD) and correlation coefficients (R; predicted vs observed) are included. Values in brackets are the % of the total variance.

		Ni	**Cu**	**Zn**	**Cd**	**Pb**
E-resin (M_{Er})	n_F	0.878	0.505	0.652	0.927	0.778
	k_o	-0.150	-1.314	-1.891	-0.472	-0.633
	k_{pH}	0.332	0.425	0.446	0.451	0.749
		(14.5)	*(38.5)*	*(56.2)*	*(55.5)*	*(63.7)*
	k_C	0.003	0.001	-	0.016	-0.002
		-	-	-	*(0.8)*	-
	M_{Er}	*(74.5)*	*(42.9)*	*(34.4)*	*(34.9)*	*(19.4)*
	RSD	*0.334*	*0.658*	*0.394*	*0.259*	*0.704*
	R	*0.948*	*0.902*	*0.950*	*0.955*	*0.903*
HNO_3 (M_{Ext})	nF	0.820	0.455	0.639	0.933	0.540
	k_o	-1.346	-1.701	-2.379	-0.855	-2.084
	k_{pH}	0.509	0.452	0.584	0.542	0.651
	k_C	0.019	0.011	0.013	0.023	-0.002
	RSD	*0.326*	*0.641*	*0.406*	*0.269*	*0.642*
	R	*0.951*	*0.909*	*0.941*	*0.952*	*0.913*

Overall the values of metal free ion activity were accurately predicted from Eq. 4-1 using either reactive metal pool (M_{Er} or M_{Ext}), with only a limited number of additional input variables relating to soil characteristics: soil pH, M_{Er} and %SOC. The difference between the predicted from the measured was less than 1 order of magnitude except for a few outliers amongst the Urban soils (Fig. 4-1 for M_{Er}). There was a clear division between the MA and Urban soils in terms of the range of free ion concentration in soils: generally metal solubility was higher in the MA soils. After 3 years of incubation at 80% field capacity, followed by 10 years storage dry, the metals added to the MA soils remain considerably more soluble than in the Urban soils – especially for Pb and possibly Ni. Although solubility must also be affected by metal loading, the average k_d values (distribution coefficient; -log L kg^{-1}) for the MA soils were greater than found in the Urban soils by factors of 0.56, 0.77, 0.74, 1.07 and 0.37

for Cd, Zn, Pb, Ni, and Cu respectively. The predicted values of p(M^{2+}) for the Urban soils also showed more scatter from model fits than MA soils (Fig. 4-1), which again indicates that the original source of contamination continues to affects the M^{2+} following extended contact with the soil.

Statistical analysis to determine the relative contribution of each soil variable to predicting p(M^{2+}) was made by stepwise regression (Minitab version 16.2.2). Soil pH was the most important variable for Zn, Cd and Pb, explaining 56.2%, 55.5% and 63.7% of the total variance respectively. This is consistent with many published studies (El-Falaky et al., 1991; Ma and Lindsay, 1993; Salam and Helmke, 1996; Sauve et al., 2000; Hough et al., 2005). By contrast, the reactive fraction (M_{Er}) was the dominant variable for determining Ni^{2+} and Cu^{2+} activity, accounting for 74.5% and 42.9% of the variance in p(Ni^{2+}) and p(Cu^{2+}) respectively. Previous studies suggest that including soil organic carbon content as a controlling influence may improve prediction of p(M^{2+}) as shown for Zn and Cd (Tye et al., 2003) and for Ni, Cu and Pb (Tye et al., 2004) and the importance of organic matter for metal sorption has been demonstrated in numerous adsorption isotherm experiments (Gerritse and Van Driel, 1984; Hooda and Aloway, 1994). However, in relative terms, %SOC is the least important variable compared with soil pH and reactive metal concentration (Tipping et al., 2003; McBride et al., 1997), which has also been demonstrated in this study. In fact, for both MA and Urban soils, %SOC was the least important variable and contributed only 0.8% of variance in p(M^{2+}) for Cd.

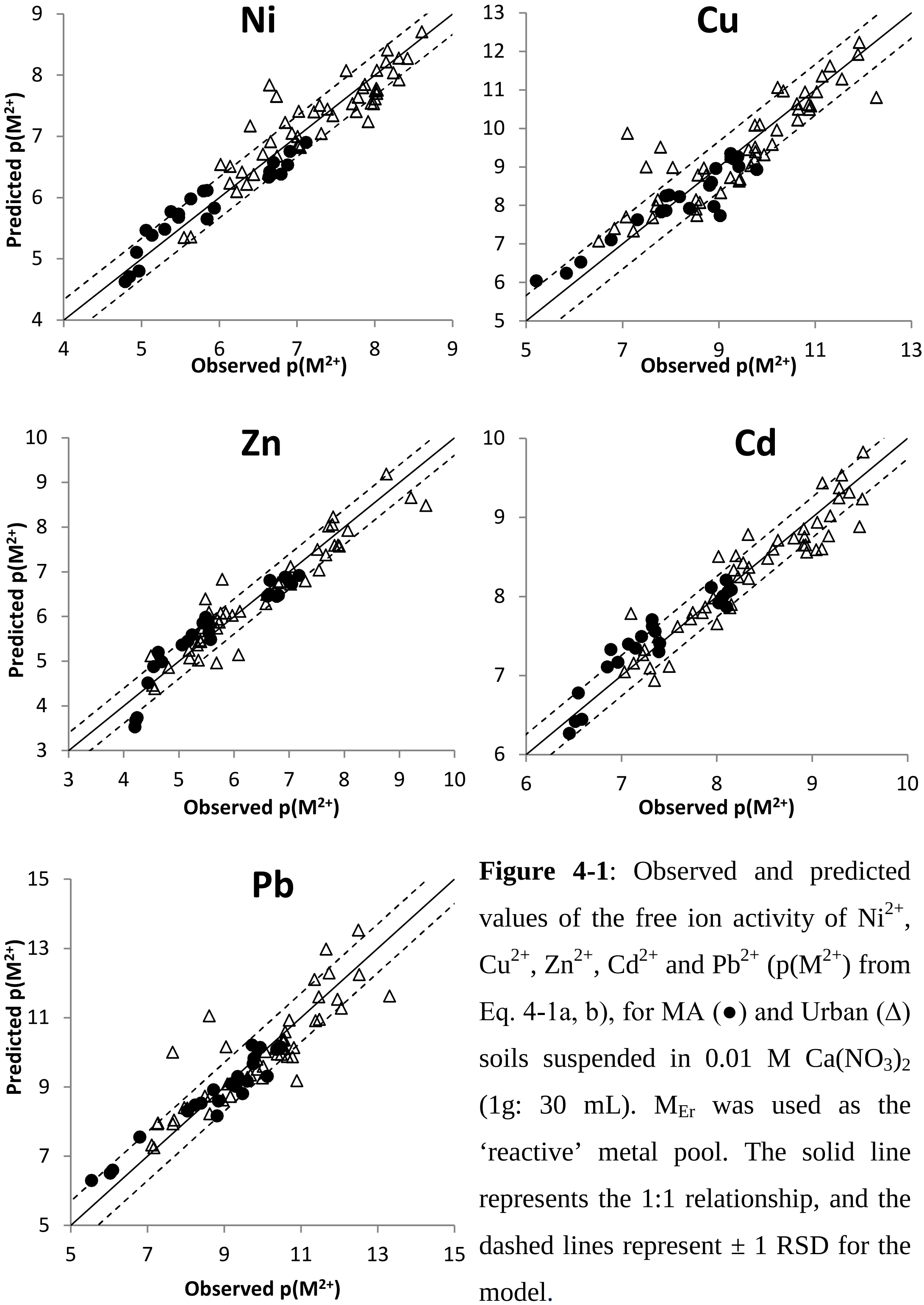


Figure 4-1: Observed and predicted values of the free ion activity of Ni^{2+}, Cu^{2+}, Zn^{2+}, Cd^{2+} and Pb^{2+} (p(M^{2+}) from Eq. 4-1a, b), for MA (●) and Urban (Δ) soils suspended in 0.01 M $Ca(NO_3)_2$ (1g: 30 mL). M_{Er} was used as the 'reactive' metal pool. The solid line represents the 1:1 relationship, and the dashed lines represent ± 1 RSD for the model.

It seems reasonable to expect that free ion activity (M^{2+}) should be estimated from the labile metal pool; Groenenberg et al. (2010) suggested that using total metal concentration provided a poorer prediction of metal solubility. However, the improvement in prediction provided by labile metal instead of total metal concentration is sometimes quite small (Degryse et al., 2003; Tye et al., 2003). In the current study, M_{Er} probably gives the closest estimation of the reactive metal fraction that controls the free ion activity in solution, while M_{Ext} is an estimation that is likely to overestimate the metal lability, especially at high pH (Section 3.3.4). However, for the extended Freundlich equation used here, there was no obvious advantage in using M_{Er} instead of a single chemical extraction with nitric acid (M_{Ext}). There was a marginally better prediction for Zn^{2+} and Cd^{2+} in solution, but not for Ni^{2+}, Cu^{2+} and Pb^{2+}, judging from values of RSD and R (Table 4-2). This may seem surprising but the lack of any clear advantage in using the labile metal fraction to predict M^{2+} in solution probably arises from compensating factors included within the regression parameters in Eq. 4-1a,b. The ratio M_E/M_{Ext} is highly pH-dependent (Section 3.3.4) and so changes in metal lability in modelling exercises using M_{Ext} will be subsumed within the coefficient k_{pH} in Eq. 4-1 (Tye et al., 2003). Thus optimised empirical models can give reasonably good predictions of FIA in soil solution.

Degryse et al. (2009) suggested that the differences in optimized constants derived from specific studies demonstrate that empirical regression models should not be extrapolated beyond the range of soil properties used in the original parameter-optimisation exercise. Ivezic et al. (2012) tried to develop a more widely applicable model by optimizing the predictive equation based on 4 independent datasets, and suggested that the model may still fail when apply on soils with extreme conditions such as very contaminated soils. Elzinga et al. (1999) collected a large number of published datasets with a total of 2000, 700 and 1500 data points for Cd, Cu and Zn sorption on soils respectively. They achieved a reasonably good fit for predicting Cd and Zn free ion activity, with a 95% prediction interval within 1 log unit of the 1:1 line for Cd (adjusted R^2 value, $R_{adj}^2 = 0.79$) and Zn (R_{adj}^2=0.83); results for Cu were less well predicted ($R_{adj}^2 = 0.69$). Furthermore, the prediction accuracy was poorer for model application to field (extracted soil pore water) data.

4.4.2 Predicting metal concentration in soil solution using WHAM-VII

4.4.2.1 Predicting total metal solution concentration from M_E

The performance of WHAM-VII in predicting Ni, Cu, Zn, Cd and Pb concentration in solution is illustrated in Fig. 4-2 as modelled against measured solubility on a $-\log_{10}$ scale (pM_{soln}). E-values (M_E) were used as the reactive metal pool in the soil. Overall WHAM-VII predicts trace metal solubility reasonably well for both sets of soils. The values of RSD were less than 1 ($\log_{10}$ unit) for all five metals. The average bias ($\Delta p(M)$) for Ni, Cu, Zn, Cd and Pb was 0.40, 0.23, 0.58, 0.56 and -0.02 respectively. Except for a few outliers in the Urban soils for Zn and Pb, the predicted solution concentrations were within 1 order of magnitude of the measured values.

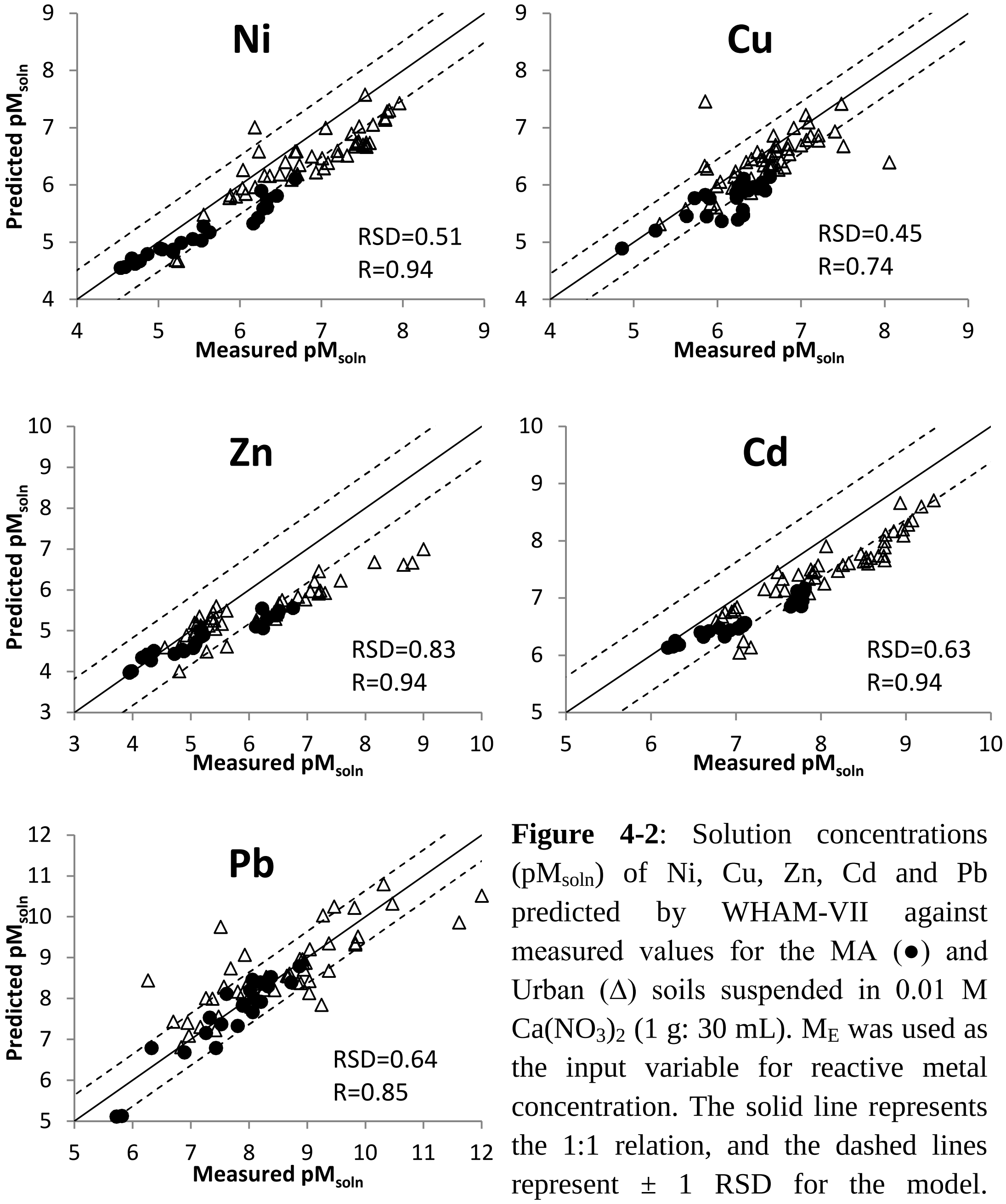


Figure 4-2: Solution concentrations (pM$_{soln}$) of Ni, Cu, Zn, Cd and Pb predicted by WHAM-VII against measured values for the MA (●) and Urban (Δ) soils suspended in 0.01 M Ca(NO$_3$)$_2$ (1 g: 30 mL). M$_E$ was used as the input variable for reactive metal concentration. The solid line represents the 1:1 relation, and the dashed lines represent ± 1 RSD for the model. Values of RSD and correlation coefficients (R) are also shown.

Generally, the WHAM-VII model gave a better prediction of metal solubility for MA soils than for the Urban soils. The most obvious difference was for Pb: the R values were 0.96 and 0.78 for MA and Urban soils respectively. Again this difference probably reflects the more diverse sources of contamination in the Urban soils. Among the five metals, the model gave a better prediction for Ni and Cd with relatively low deviation from the 1:1 line (RSD = 0.51 and 0.63 respectively) and high correlation coefficient (R = 0.94 for both metals). The high RSD value for Zn (0.83) arose because the model substantially overestimated the solution concentration for several high pH (6.93 to 8.08) Urban soils with very low Zn solution concentration. Prediction of Cu solubility was reasonably good (RSD = 0.45; R=0.74). Lead overall, was most poorly predicted by WHAM-VII (RSD=0.64; R=0.85). However, Figure 4-2 indicates there was a better prediction for MA soils than for Urban soils. Splitting the dataset, it was clear that, though there is less average bias, low correlation and large deviation arose from the contribution from the Urban soils (RSD=0.73, R=0.78, ΔpPb_{soln}= -0.08); whereas solubility for the MA soils was well predicted (RSD= 0.35, R= 0.96, ΔpPb_{soln}= 0.11).

For all five metals WHAM-VII, using the default parameters, generally overestimated solubility especially at low levels of measured solubility (high pM_{soln} values). These were likely to be the high pH calcareous soils, as the increasing pH leads to less metal competition with hydrogen ions for pH-dependent cation exchange sites on adsorption surfaces (McBridge et al., 1997; Knight et al., 1998; Meers et al., 2006; Meers et al., 2007; Martinez and Motto, 2000). The overestimation of the solubility was not found to be a problem for soils with high organic matter contents (Tipping et al., 2003), mainly peat soils. The default constants in WHAM were originally parameterized on ‘purified’ single geocolloidal systems and the most comprehensive datasets employed were from studies of metal binding by humic and fulvic acids. Therefore it is likely that soils in which humus is the dominant adsorption surface are likely to perform better than those with complex assemblages of organic and mineral geocolloids. As suggested by Smith et al. (2004) the properties of the cation binding sites of humic substances in peat appear very similar to those of isolated humic substances. It has been suggested in other studies that prediction of the trace metal solubility with multi-surface geochemical models is more successful when solid↔solution partitioning is controlled mainly by soil organic matter (Weng et al., 2002; Cances et al., 2003). In pH-neutral and calcareous (mineral) soils, predicting solubility is less successful without adjustment of the model default parameters (Schroder et al., 2005; Buekers et al., 2008b) and it is difficult to justify this process except as a fitting exercise.

The literature on modelling trace metal in whole soils using WHAM is very limited. However, it has been reported that prediction of Cu and Pb solubility is often less successful than modelling Cd and Zn solubility (Tipping et al., 2003; Bueckers et al., 2008b; Almas et al., 2007; Lofts and Tipping, 2011). Over- or under-estimation of Cu and, more often, Pb solubility by other geochemical models has also been reported; studies have used the NICA-Donnan model, Stockholm humic model and ORCHESTRA (MacDonald & Hendershot, 2006; Weng et al., 2002; Schroder et al., 2005). It has also been suggested that the default model parameters need to be adjusted before model application to whole soils. Targets for such adjustments have included the adsorption affinity to organic matter (MacDonald & Hendershot, 2006) and (hydr)oxides - especially for Pb (Degryse et al., 2009). This is a debatable practice however, given that constants in the original model have been derived for single adsorbent studies under controlled conditions and there is no basis for adjustment other than ‘goodness of fit’ to the whole soil data. It effectively reduces the otherwise mechanistic model to an empirical fitting exercise.

4.4.2.2 Comparing WHAM-predicted solubility using M_E or M_{Ext} as inputs

When applying geochemical speciation models to natural soils, choices have to be made regarding the variables required as model inputs. Relevant examples include the concentration of reactive elements, mineral (hydr)oxide content and estimation of the active fraction of solid and dissolved organic matter. Therefore, it is important to test the model sensitivity to these different choices thereby assessing the model uncertainty (Groenenberg et al., 2012). Several studies have been undertaken comparing the ‘reactive’ metal content, measured by chemical extraction (Almas et al., 2007) or E-values (Buekers et al., 2008b), to ‘total’ soil metal content (M_{Total}) as the principal input variable. As expected, it is usually found that using M_{Total} overestimates the solution concentration. In this study, using WHAM-VII, the prediction of solubility using M_E as the reactive metal concentration was compared to the use of M_{Ext}. Predicted solubility data using M_{Ext} are plotted against measured values in Fig. 4-3 and RSD and correlation coefficients were compared with the model performance using M_E in Table 4-3.

In marked contrast to the (empirical) extended Freundlich models (Section 4.4.1), the difference of model prediction between using M_E and M_{Ext} is very clear. Overall, using M_{Ext} as a variable gives an inferior prediction for all five metals with a much higher RSD and bias from the 1:1 line (i.e. larger average ΔpM values; Table 4-3). The overestimation was more pronounced in the low solubility range, dominated by the high pH Urban soils. As discussed in Section 3.3.4, the nitric acid extractant mobilizes non-labile metal in soils which, in turn, causes an overestimation of the solution concentration predicted using M_{Ext}. The difference between M_E and M_{Ext}

predicted solubility was much less at higher levels of solubility in acidic soils. Differences were more pronounced for Ni, Zn and Cd and less so for Cu and Pb. The use of M_{Ext} also causes greater scattering of results for all metals except Cu. This may have arisen from the relatively weak influence of pH on Cu lability in soils (Section 3.3.4).

Table 4-3: Summary of modelling outcomes predicting solution concentration of Ni, Cu, Zn, Cd and Pb using either E-values (M_E) or 0.43 M HNO_3 extractable metal content (M_{Ext}), as input variables to WHAM-VII.

		Ni	**Cu**	**Zn**	**Cd**	**Pb**
Using *E-value* (M_E)	RSD	0.51	0.45	0.83	0.63	0.64
	R	0.94	0.74	0.94	0.94	0.85
	$\Delta p(M)$	0.40	0.23	0.58	0.56	-0.02
	slope	0.82	0.73	0.54	0.78	0.80
	intercept	0.77	1.53	2.08	1.11	1.70
Using *HNO_3 extractable* metal content (M_{Ext})	RSD	0.95	0.64	1.23	0.89	0.76
	R	0.87	0.77	0.82	0.87	0.82
	$\Delta p(M)$	0.82	0.54	0.95	0.79	0.31
	slope	0.64	0.70	0.40	0.67	0.61
	intercept	1.48	1.42	2.51	1.79	2.94

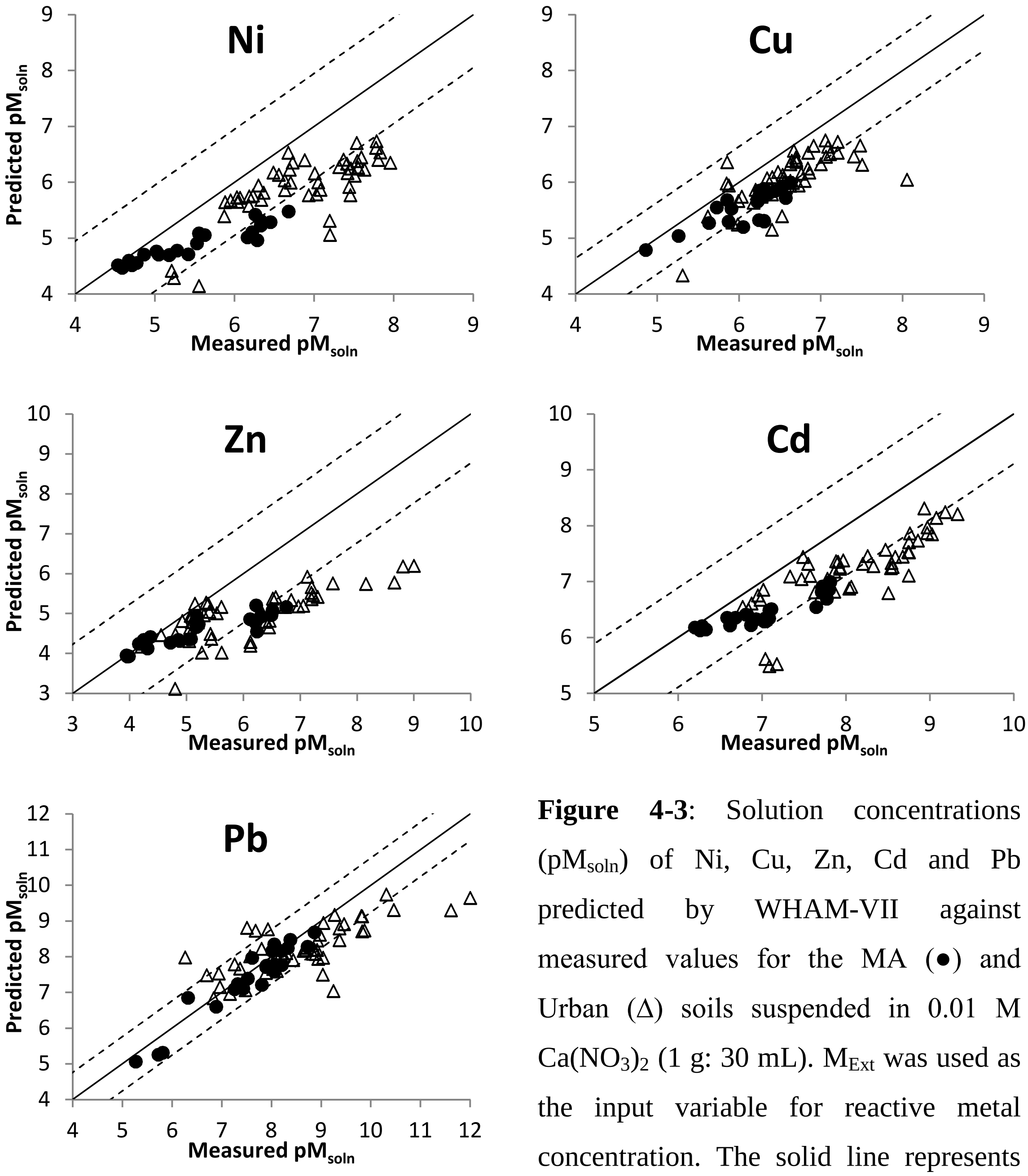


Figure 4-3: Solution concentrations (pM_{soln}) of Ni, Cu, Zn, Cd and Pb predicted by WHAM-VII against measured values for the MA (●) and Urban (Δ) soils suspended in 0.01 M $Ca(NO_3)_2$ (1 g: 30 mL). M_{Ext} was used as the input variable for reactive metal concentration. The solid line represents the 1:1 relation, and the dashed lines represent ± 1 RSD for the model.

4.4.3 Modelling trace metal binding on solid phases

Soils contain a wide range of geocolloids, each presenting different intrinsic affinities for trace metals and degrees of pH-dependent metal affinity, or dependence on surface charge. The resulting variation in the solid phase fractionation of metals must, in turn, have a significant impact on metal concentration in the soil solution (Almas et al., 2007; Shuman, 1991). Trace metals exhibit markedly different affinities for the various binding phases present. For example, Cu binds particularly strongly with humus and therefore its solubility is affected by changes in the solid↔solution partitioning of humus acids (Tye et al., 2004; McBride et al., 1997). Nickel and Zn, however, do not bind so strongly with humus and so their solubility is more affected by changes in pH (McBride, 1994); Pb binding is dominated by Mn oxide at low pH and Fe oxides at higher pH values.

Trace metal fractionation in the solid phase is provided by WHAM-VII as a results output. As discussed above, even using reactive trace metal concentrations based on M_E values, the model still underestimates solid phase binding (over-estimates solubility). Therefore, metal adsorption on different binding phases was investigated by interrogation of the WHAM-VII output data to determine whether there were any particular binding phases that caused the deviation between measured and predicted solubility observed.

4.4.3.1 Trace metal fractionation in soil as a function of pH

Soil pH is a major determinant of metal dynamics because it has a major impact on chemical speciation in soil solution (McLaughlin et al., 1998; Rieuwerts et al., 2005; Shuman, 1991) and also on strength of binding to particulate phases. Figure 4-4 illustrates the variation in Ni, Cu, Zn, Cd and Pb bound to different soil fractions as a function of soil pH intervals for both the MA and Urban soils. The fractionation was modelled using WHAM-VII and includes a complete breakdown of all particulate geocolloidal fractions and a single pool for the solution phase, including colloidal FA. Overall, although there is a large variation in the composition of soil binding phases (Section 3.3.1), metal fractionation is mainly controlled by soil pH. Figure 4-4 suggests that the relative importance of Fe-oxides for metal binding increases with pH, whilst that of organic matter (HA and FA) decreases. This is because the pH dependency of metal adsorption to mineral (hydr)oxides is much stronger (Weng et al., 2004) than to organic matter and thus becomes dominant in metal binding at high pH (> pH 7).

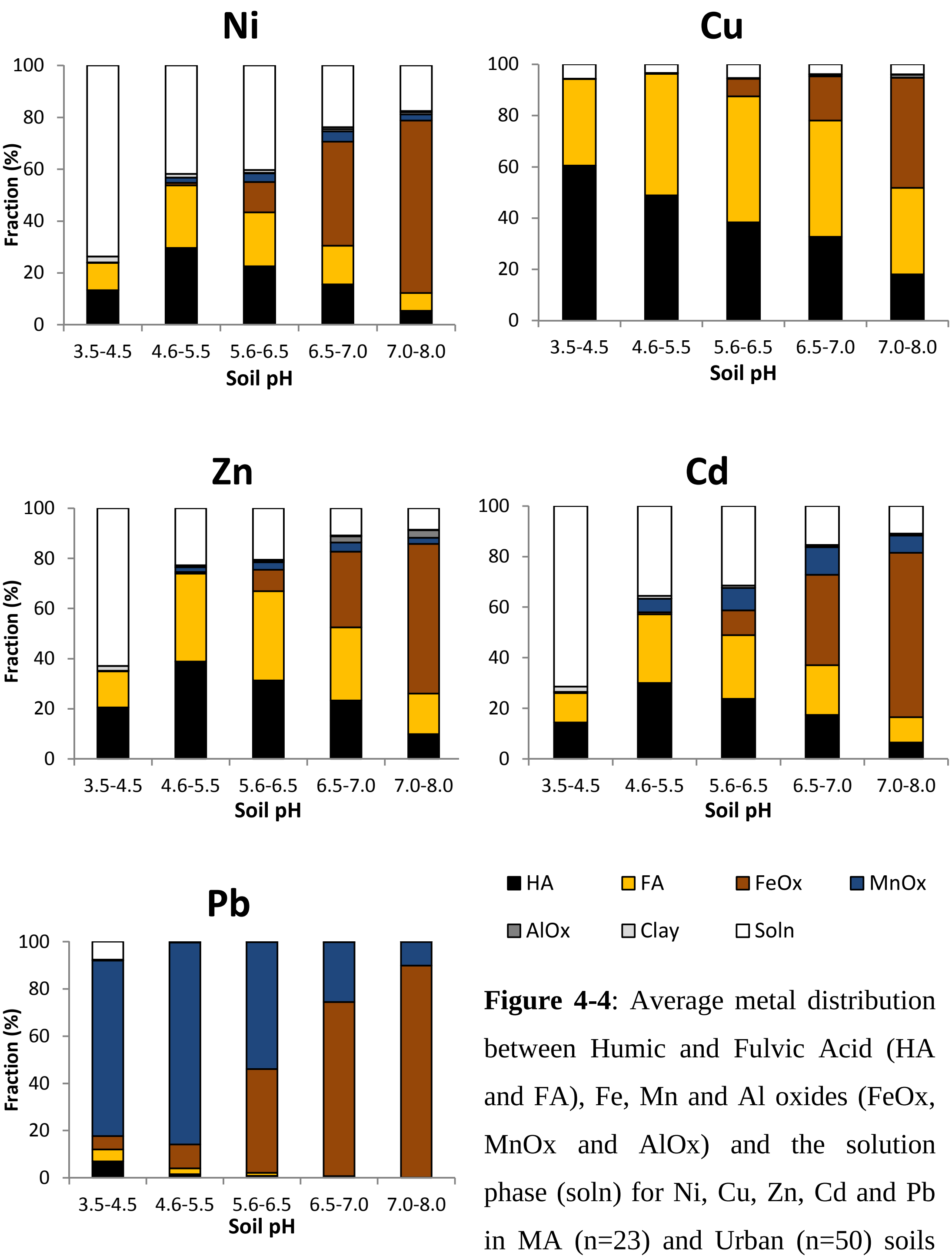


Figure 4-4: Average metal distribution between Humic and Fulvic Acid (HA and FA), Fe, Mn and Al oxides (FeOx, MnOx and AlOx) and the solution phase (soln) for Ni, Cu, Zn, Cd and Pb in MA (n=23) and Urban (n=50) soils grouped by pH class. Fractionation was predicted using WHAM-VII parameterized using M_E as input.

Most of the Cu (> 94.4%) and Pb (> 92.3%) were absorbed by the soil across the pH range, even at low pH values, although there were substantial differences in their affinity for particular binding phases. At low and medium pH, Mn oxides appeared to be an important binding phase for Pb whereas organic matter was more important for Cu. The increase of binding to whole soil with pH is clear for Ni, Zn and Cd and the binding to different soil fractions follows a similar pattern, but organic matter is more important for Zn binding than for Ni and Cd adsorption. This is supported by other studies. Buekers et al. (2008b) showed that between pH 3.4 and 4.5, the fractions of Ni and Zn sorbed on organic matter were 51% and 69% on average. When pH was raised to the circumneutral range, between 6.6 and 7.7, iron (hydr)oxide becomes the most important adsorbent. Non-specific adsorption of Ni, Zn and Cd on clay is only significant in soils with very low pH values (pH 3.5 – 4.5) and low organic matter contents, less than 2.2%, 1.9% and 2.2% respectively.

4.4.3.2 Metal binding to solid phase and bias in prediction of solubility

The absolute difference between measured and predicted metal solubility (ΔpM_{soln}) is plotted as a function of (i) soil pH and (ii) the fractions bound to geocolloidal phases (HA, FA, FeOx, MnOx, AlOx and clay) in Figs. 4-5 – 4-9. For all five metals, there was a strong relationship between model performance and soil pH, especially in the MA soils. For Ni, Cu, Zn and Cd, the extent of overestimation of the metal solubility increases with increasing soil pH. By contrast, prediction for Pb in MA soils seems relatively unaffected by soil pH. In the Urban soils, the model both underestimated the solubility in low pH soils and overestimated solubility in high pH soils.

Although HA and FA are important for binding of Ni, Zn and Cd, the bias observed seems to be mainly related to metal adsorption on Fe, Al and Mn oxides in the high pH range (pH > 6). Prediction for Cu is expected to be especially sensitive to the soil humic and fulvic acid content. Figure 4-6 (c) shows a decrease in bias with increase in HA concentration and a corresponding increase in bias with oxides content is also clear (Fig. 4-6e, f and g). However, as Cu is more dominantly bound with HA and FA (Fig. 4-4), it is less likely that the predicted solution concentration is affected by adsorption on oxides. In contrast with Cu, organic matter content is not the main factor controlling Pb solubility (Degryse et al., 2009). On average, WHAM predicts about 96% of the Pb is adsorbed on Fe and Mn oxides for the MA and Urban soils. Several studies have suggested that sorption of Pb on hydrous oxides is significantly under-estimated by geochemical speciation models (Morin et al., 1990; Tongtavee et al., 2005; Degryse et al., 2007). However, it is difficult to conclude from Fig. 4-9 whether failure to adequately predict Pb solubility is simply the result of poor parameterisation of Pb adsorption equations on particulate binding phases. Alternative explanations offered within the literature include a failure to account for (i) synergistic effects associated with adsorption on mixed oxide-humic surfaces (Heidmann et al., 2005; Pedrot, et al., 2009); (ii) the existence of highly specific sites on Fe (hydr)oxides with much greater intrinsic stability constants than used in the general surface complexation model (Buekers et al., 2008; Linde et al., 2007) or (iii) formation of discrete solid phases such as chloropyromorphite ($Pb_5(PO_4)_3Cl$) (Degryse et al., 2009), which is likely to affect Pb dynamics in some of the Urban and sewage sludged soils.

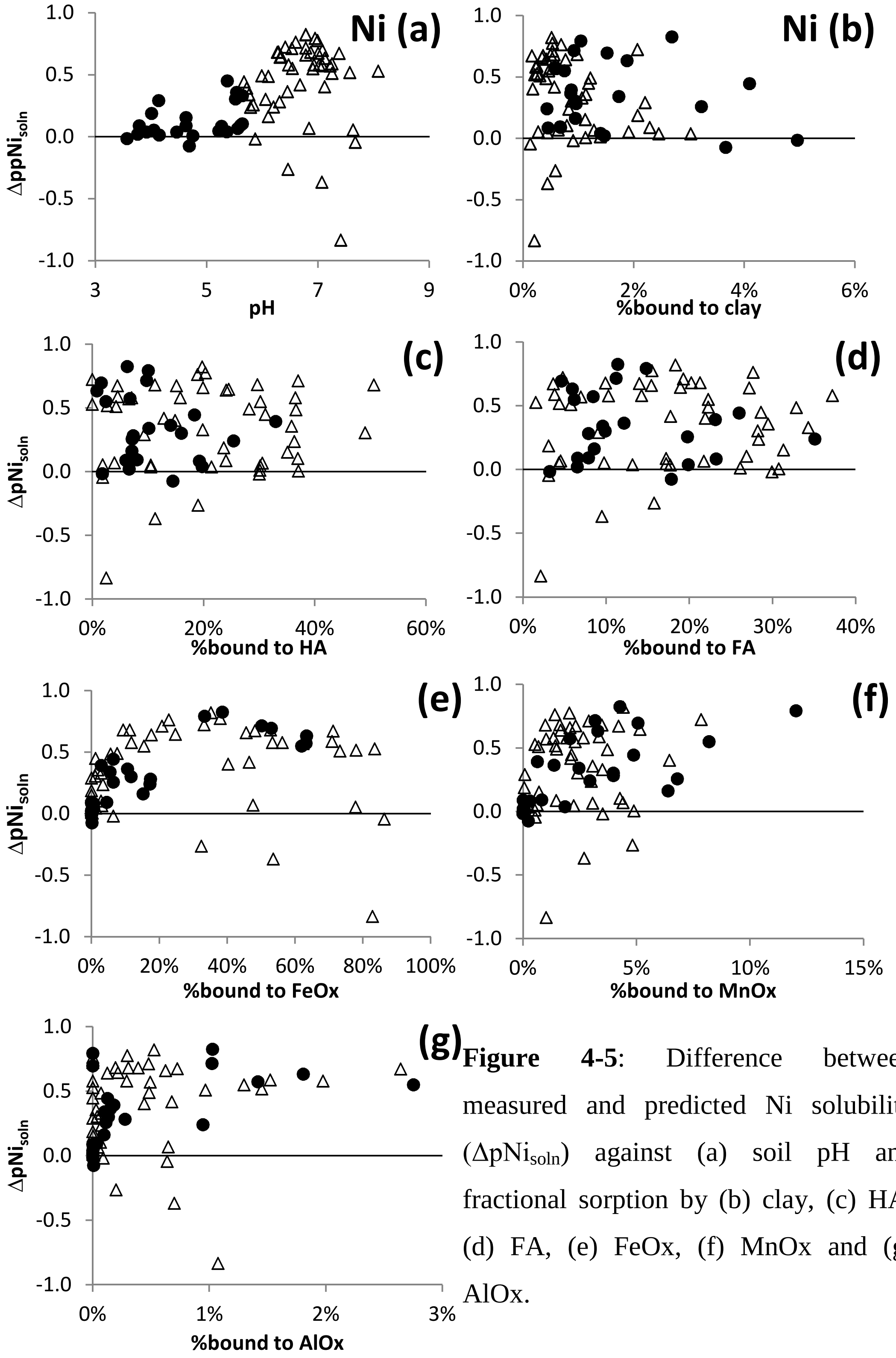


Figure 4-5: Difference between measured and predicted Ni solubility (ΔpNi_{soln}) against (a) soil pH and fractional sorption by (b) clay, (c) HA, (d) FA, (e) FeOx, (f) MnOx and (g) AlOx.

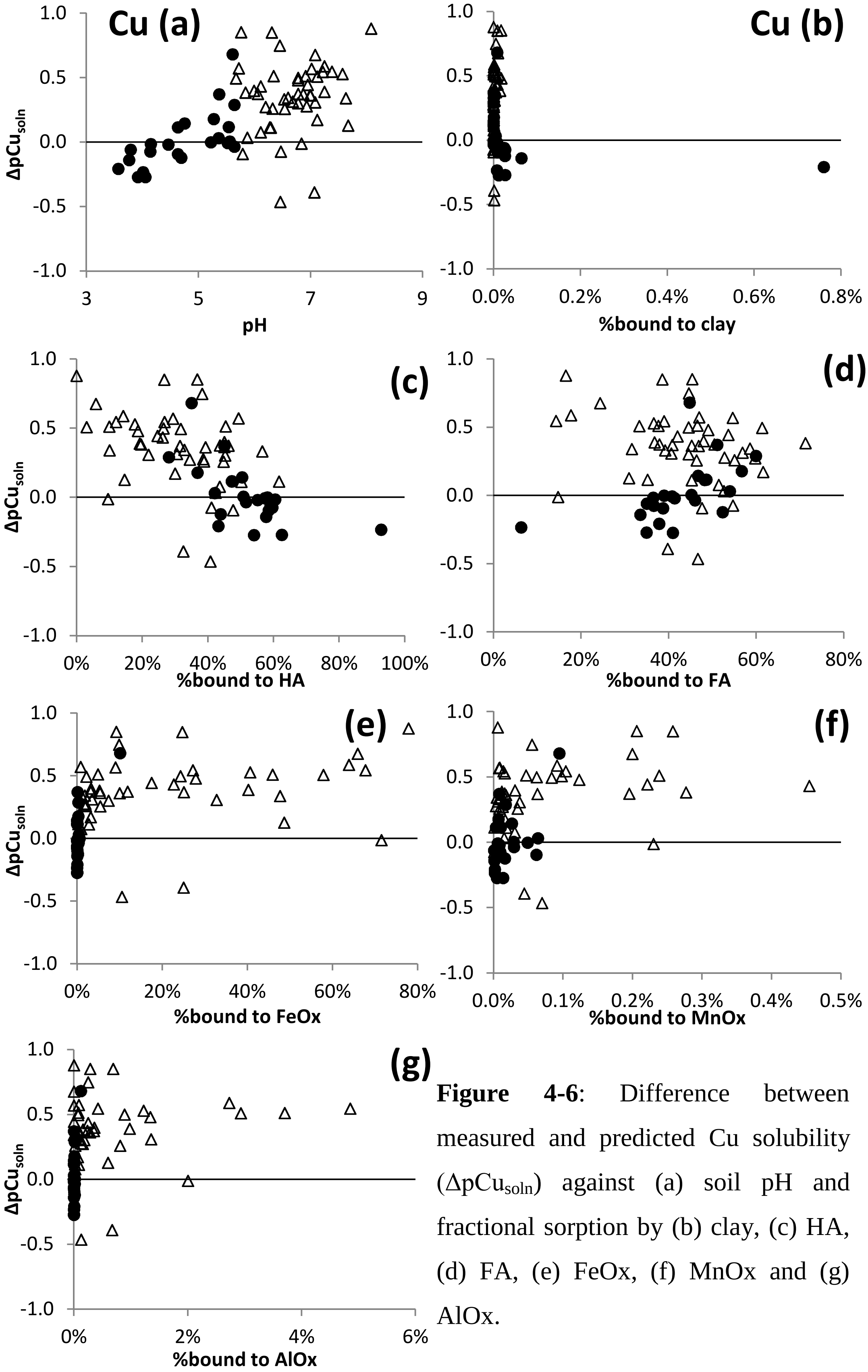


Figure 4-6: Difference between measured and predicted Cu solubility (ΔpCu_{soln}) against (a) soil pH and fractional sorption by (b) clay, (c) HA, (d) FA, (e) FeOx, (f) MnOx and (g) AlOx.

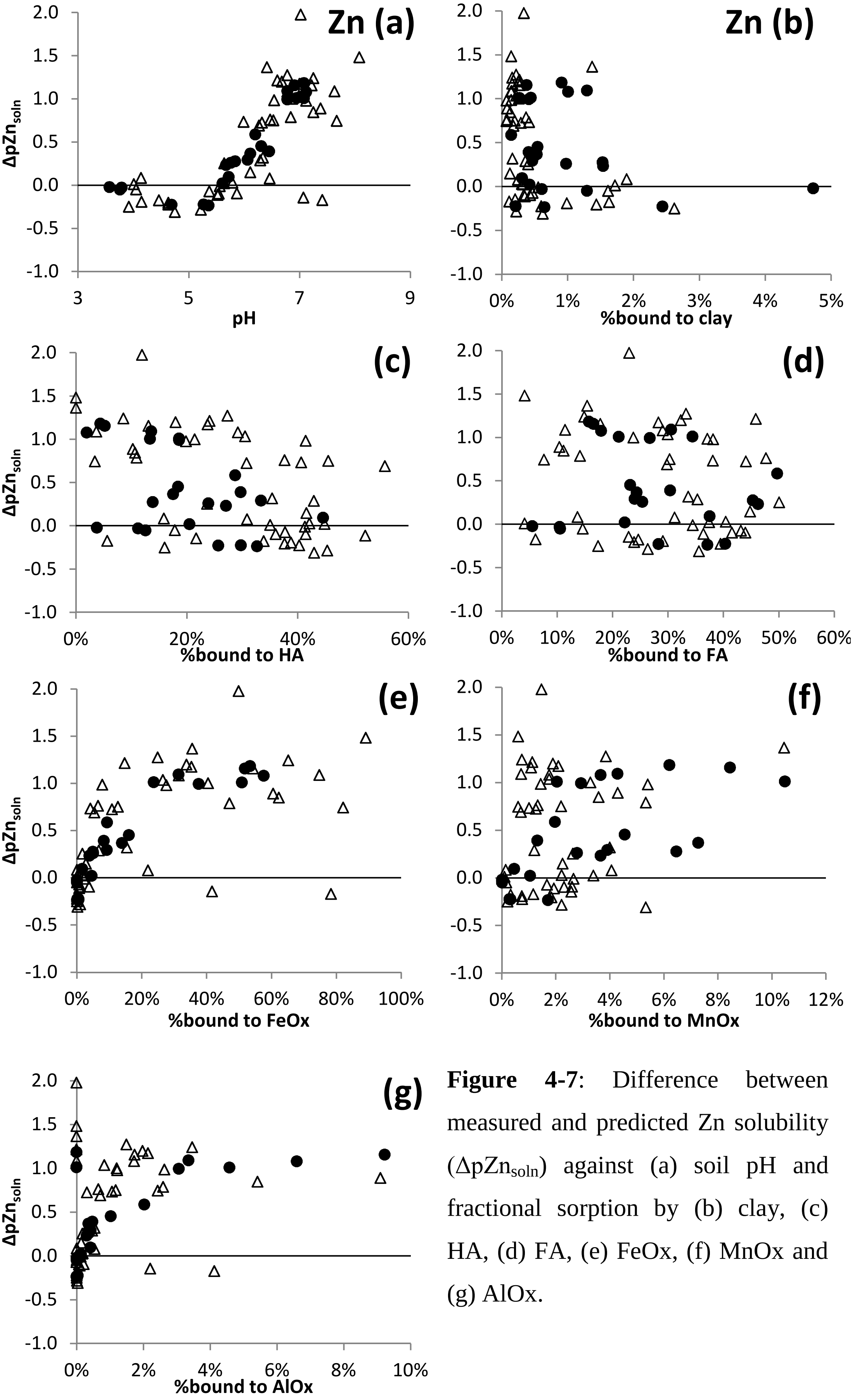


Figure 4-7: Difference between measured and predicted Zn solubility (ΔpZn_{soln}) against (a) soil pH and fractional sorption by (b) clay, (c) HA, (d) FA, (e) FeOx, (f) MnOx and (g) AlOx.

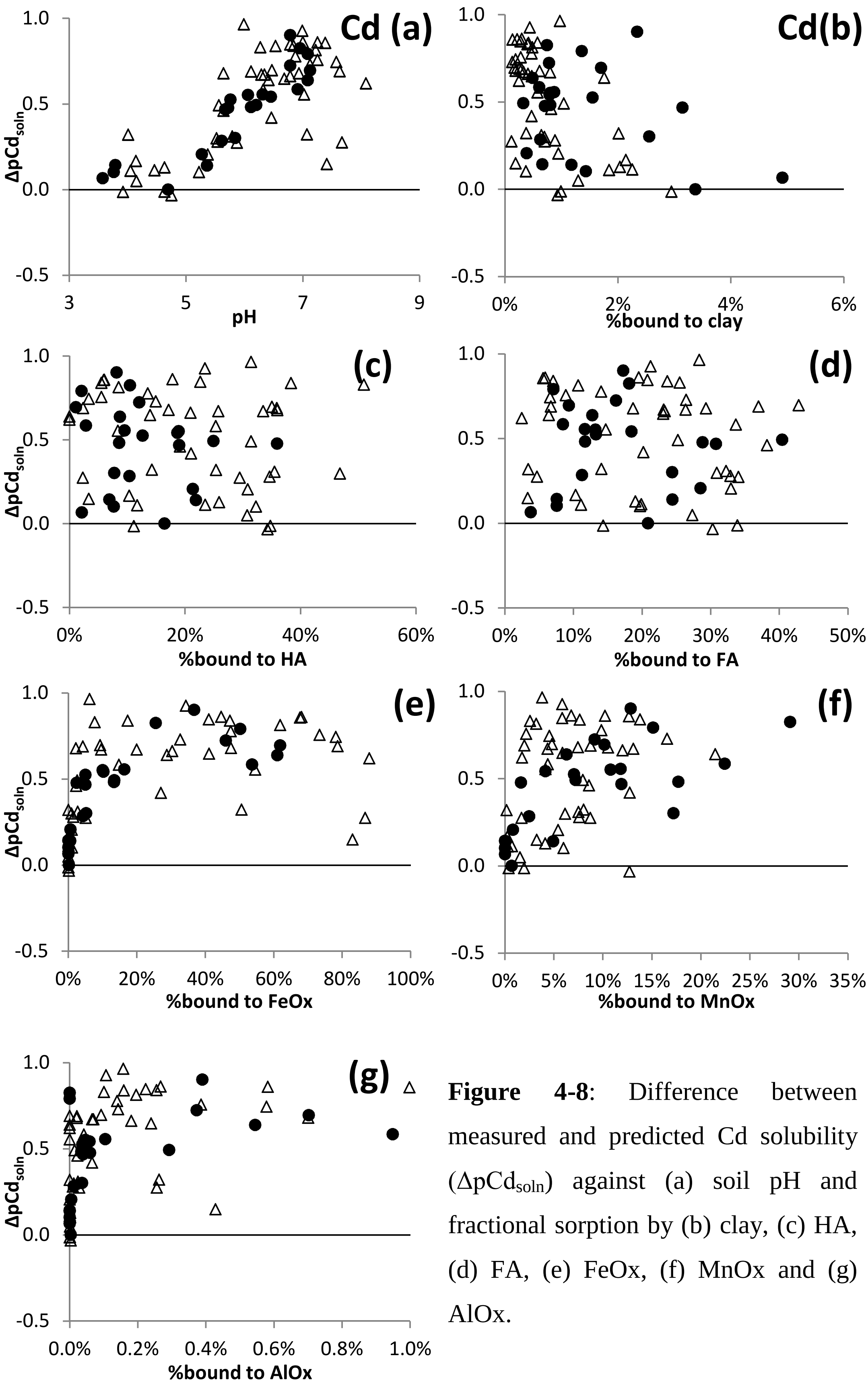


Figure 4-8: Difference between measured and predicted Cd solubility (ΔpCd_{soln}) against (a) soil pH and fractional sorption by (b) clay, (c) HA, (d) FA, (e) FeOx, (f) MnOx and (g) AlOx.

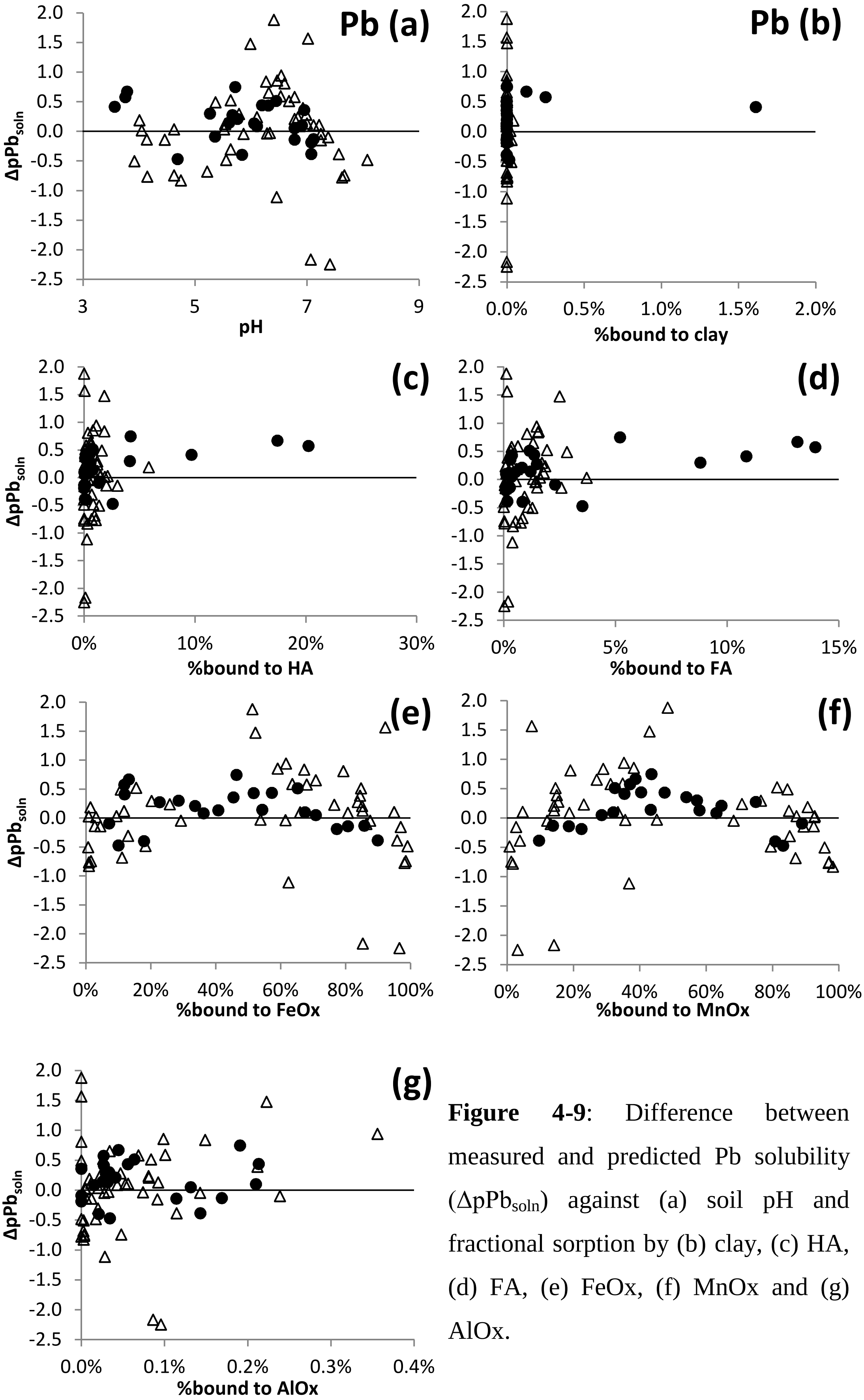


Figure 4-9: Difference between measured and predicted Pb solubility (ΔpPb_{soln}) against (a) soil pH and fractional sorption by (b) clay, (c) HA, (d) FA, (e) FeOx, (f) MnOx and (g) AlOx.

A significant underestimation of measured solubility (> RSD) was seen for a small number of soils (Fig. 4-2; WV7 for Ni, Cu and Pb; WV10 for Pb). This may be a result of overestimation of some of the binding phases. For example, for soil WV7, WHAM predicted 83%, 70%, and 97% of binding on Fe oxides for Ni, Cu and Pb, and the bias was -0.83, -1.62 and -2.25 respectively; this implies over prediction of metal binding on Fe oxides. As it has just been discussed, generally the model algorithms *underestimated* the metal binding on mineral oxides. Therefore, overestimation of metal binding suggests that, for these soils, the DCB method (Section 2.1.4) extracted more Fe than was actually present in the Fe (hydr)oxides phase, leading to an overestimation of the Fe oxide content. The source of this Fe may have been Fe^{II} and Fe^{III} bound to humus which would cause overestimation of metal binding for two reasons. First, mistakenly ascribing humus-bound Fe to a Fe (hydr)oxide binding phase, as discussed above and, second, possibly failing to include humus-bound Fe as a competitor ion which would increase metal solubility.

4.4.3.3 Optimization of Fe-oxide characteristics and binding constants:

The apparent correlation between the bias in pM_{soln} and mineral oxides content in soils implies the parameters governing metal adsorption on oxides may require adjustment. Buekers et al. (2008b) found that the reactive surface area of oxyhydroxides is a sensitive parameter for modelling metal solubility, especially in pH-neutral and calcareous soils and in Fe-oxyhydroxides dominated soils. The surface areas of soil oxyhydroxides varies among different types of pure Fe-oxyhydroxides and they might be larger in soil, because of impurities that are built into the oxyhydroxide structure, defective structures in the oxyhydroxide crystal, the partitioning of the oxyhydroxide in the soil and increased surface area caused by the formation of thin surface coatings on other minerals (Buekers et al., 2008b). For instance, Eusterhues et al. (2005)

estimated that the surface area of iron oxyhydroxides in soil was in the range 200-800 m^2 g^{-1}, based on surface area measurements of soils before and after removal of iron oxyhydroxides. The default value of surface area of Fe oxides in WHAM is 600 m^2 g^{-1} and so this was adjusted to 800 m^2 g^{-1} to test the model sensitivity to this variable. Improvement in prediction is shown in Table 4-4 for Ni, Zn and Cd as there is a decrease in RSD and a slightly increase in R but for Cd only. However, there was still an overestimation of metal solubility. Surface area values greater than 800 m^2 g^{-1} were not assessed because of a lack of any literature support. It might also be more appropriate to adjust the binding constants relating to Ni, Zn and Cd sorption affinity to Fe and Mn oxides to achieve better model prediction in solubility (Linde et al., 2007). Further improvements could possibly be made by including the interaction of DOM with oxide surfaces and its effect on oxy-anion binding (Gustafsson, 2006; Weng et al., 2009). But it has also been argued that, the capacity for further improvements might be limited by intrinsic uncertainties related to the heterogeneous character of natural organic matter (Groenenberg et al., 2010) and other reactive soil constituents.

Table 4-4: Performance of WHAM-VII modelling Ni, Cu, Zn, Cd and Pb solubility using default and adjusted FeOx surface areas.

(FeOx) surface area		Ni	Cu	Zn	Cd	Pb
Default surface area (600 m^2 g^{-1})	RSD	0.51	0.45	0.83	0.63	0.64
	R	0.94	0.74	0.94	0.94	0.85
	Δp(M)	0.40	0.23	0.58	0.56	-0.02
Adjusted surface area (800 m^2 g^{-1})	RSD	0.48	0.44	0.78	0.58	0.64
	R	0.94	0.74	0.94	0.95	0.85
	Δp(M)	0.36	0.21	0.54	0.52	-0.09

4.4.3.4 Over prediction of metal solubility in high pH soils by WHAM

This section briefly explores further reasons for the general over-prediction of metal solubility by WHAM - especially in alkaline soils.

Initially designed for the aqueous environment, WHAM-VII recognises only a limited suite of binding phases. Besides those already considered in this study (clays, Fe, Mn and Al oxides, HA and FA), the model also includes silica and quartz. However, there are other binding phases in soils which may actively adsorb metal cations especially in high pH soils. For example, calcite ($CaCO_3$) and hydroxyapatite ($Ca_5(PO_4)_3(OH)$), which are only likely to present in soils with high pH (> 7.0 and > 6.0 respectively), can adsorb metal cations by surface replacement of Ca^{2+} on mineral surface sites (Davis et al., 1987; Ahmed et al., 2008; Bailey et al., 2005). Other binding phases may have much smaller effect on metal binding than the major constituents (HA, FA and mineral (hydr)oxides), but tend to be more important in high pH soils due to the development of negative surface charge. The exclusion of these phases in the WHAM model may be the primary reason for the general over-prediction of solution metal concentration at high pH.

It is widely recognised that adsorption of metals on mineral (hydr)oxides increases with pH and occurs over a short range of pH which varies between different (hydr)oxides; Fischer et al. (2007) show the typical trend for a range of metals adsorbed on goethite. This is partly controlled by an intrinsic affinity of the metal for the oxide surface and partly by the surface charge on the oxide. If the chemical affinity of the metal ion for the oxide outweighs electrostatic repulsion, then it is even possible for cations to be adsorbed on to oxides below their point of zero charge (PZC). A summary of measured PZC values for (hydr)oxides and clay minerals can be found in Kosmulski (2009) with relatively low values found in Mn (hydr)oxides (4 to

4.5) and high values for goethite (8.1 to 9.5). Soils are likely to have a range of (hydr)oxides, which may vary in PZC, surface morphology/area, crystallinity and degree of surface contamination with sorbed anions and humus acids. All of these factors together will generate a range of adsorption strengths for metal cations and a range of apparent PZC values. As pH rises, therefore, the *diversity* of oxide surfaces involved in metal adsorption will increase. Therefore, if a speciation model generates error in predicting metal binding on (hydr)oxides surfaces, the effect is likely to be more significant in high pH soils as there is an accumulation of error contributed by each oxides phase. This may also explain the limited improvement in prediction by optimizing the parameter for Fe oxides only (Section 4.4.3.3).

As a sub model of WHAM-VII, Model VII (or VI) was fairly robust in predicting metal binding and speciation on humic and fulvic acids, both in particulate and colloidal phases, which are more important in modelling metals in low pH or/and organic rich soils. In contrast, the surface complexation model for describing metal speciation on mineral (hydr)oxides may be relatively weak in terms of its simplicity in (i) combining all soil (hydr)oxide adsorbents into just three types (Al, Mn and Fe oxides) and (ii) using a single parameter to describe heterogeneity for all metals (Lofts and Tipping, 1998) and oxides (default setting). Such a simplification is reasonable to avoid the need for a huge database limiting the applicability of the model. However, it may also be the reason for greater deviation from measured values in predicting solubility in high pH soils in which mineral (hydr)oxides are the dominant metal binding phases.

WHAM prediction of solubility in Urban soils generally produced a poorer correlation than that for MA soils. This may be the result of slightly higher pH values in Urban soils (Section 3.3.1) as a result of alkaline wastes such as cement, concrete etc.

(Bridges, 1991; Rosenbaum et al., 2003). The higher metal sorption capacity seen in some brownfield soils, compared to non-urban soils, is thought to involve processes of both sorption and precipitation (Markiewiez-Patkowska et al., 2005; Mclean and Bledsoe, 1992). Some particular examples include (i) a large bias (ΔpM) observed for Zn and Pb with values of 1.7 and 1.8 respectively in soil WV21; (ii) extremely low metal solution concentrations which lead to greater experimental error in some urban soils - e.g. Zn in NG13 with pH 7.0 and total Zn concentration of only 26 mg kg^{-1}; (iii) a very high binding capacity in the sewage farm soils studied due to organic matter from the sewage sludge resulting in overestimation of Cd solution concentrations in NG14 and NG15.

4.4.4 Comparison between WHAM-VI and WHAM-VII

As one of the main sub-models in the WHAM-VII assemblage model, Model-VII is identical to VI with respect to its formulation of total monodentate binding sites, proton binding constants and electrostatic effects, but is a simpler model than Model VI (WHAM-VI) in its postulated multidentate metal binding sites. Although there are fewer model parameters, Model VII was parameterized on twice as many datasets as were used for Model VI. The new model is therefore believed to provide better descriptions of metal binding at higher pH, and to have more internally consistent parameter values (Tipping et al., 2011).

A comparison was made to assess possible improvements of WHAM-VII over its predecessor in modelling trace metal solubility in whole soils. The predicted solubility from the two models is compared in Fig. 4-10 and summarized in Table 4-5. The predicted results are well correlated for all five metals with R values (>0.96).

However, in contrast to the other four metals, large deviations between the two models are seen for Pb (RSD = 0.74); WHAM-VI output shows a much greater deviation from measured values of pM_{soln} compared to WHAM-VII (ΔpM = 0.64 and -0.02 respectively). Overall, therefore, there appears to be a significant improvement in modelling Pb solubility using WHAM-VII with a minor improvement for Ni, but virtually no change for Cu, Zn and Cd. Better prediction of Zn, Cd and Pb solubility in minespoil soils was found by Marzouk et al. (2013) in using WHAM-VII instead of WHAM-VI, however, the difference was very small comparing the scale of the deviation from measured values.

Table 4-5: Summary of comparing WHAM-VI and WHAM-VII predicting solution concentration of 5 trace metals.

		Ni	**Cu**	**Zn**	**Cd**	**Pb**
WHAM-VI vs VII	RSD	0.08	0.10	0.16	0.08	0.74
	R	1.00	0.98	0.99	1.00	0.96
	Δp(M)	0.00	0.01	0.05	0.05	-0.65
WHAM-VI predicted vs measured	RSD	0.51	0.44	0.72	0.57	1.03
	R	0.94	0.75	0.96	0.95	0.74
	Δp(M)	0.40	0.23	0.53	0.51	0.64
WHAM-VII predicted vs measured	RSD	0.51	0.45	0.83	0.63	0.64
	R	0.94	0.74	0.94	0.94	0.85
	Δp(M)	0.40	0.23	0.58	0.56	-0.02

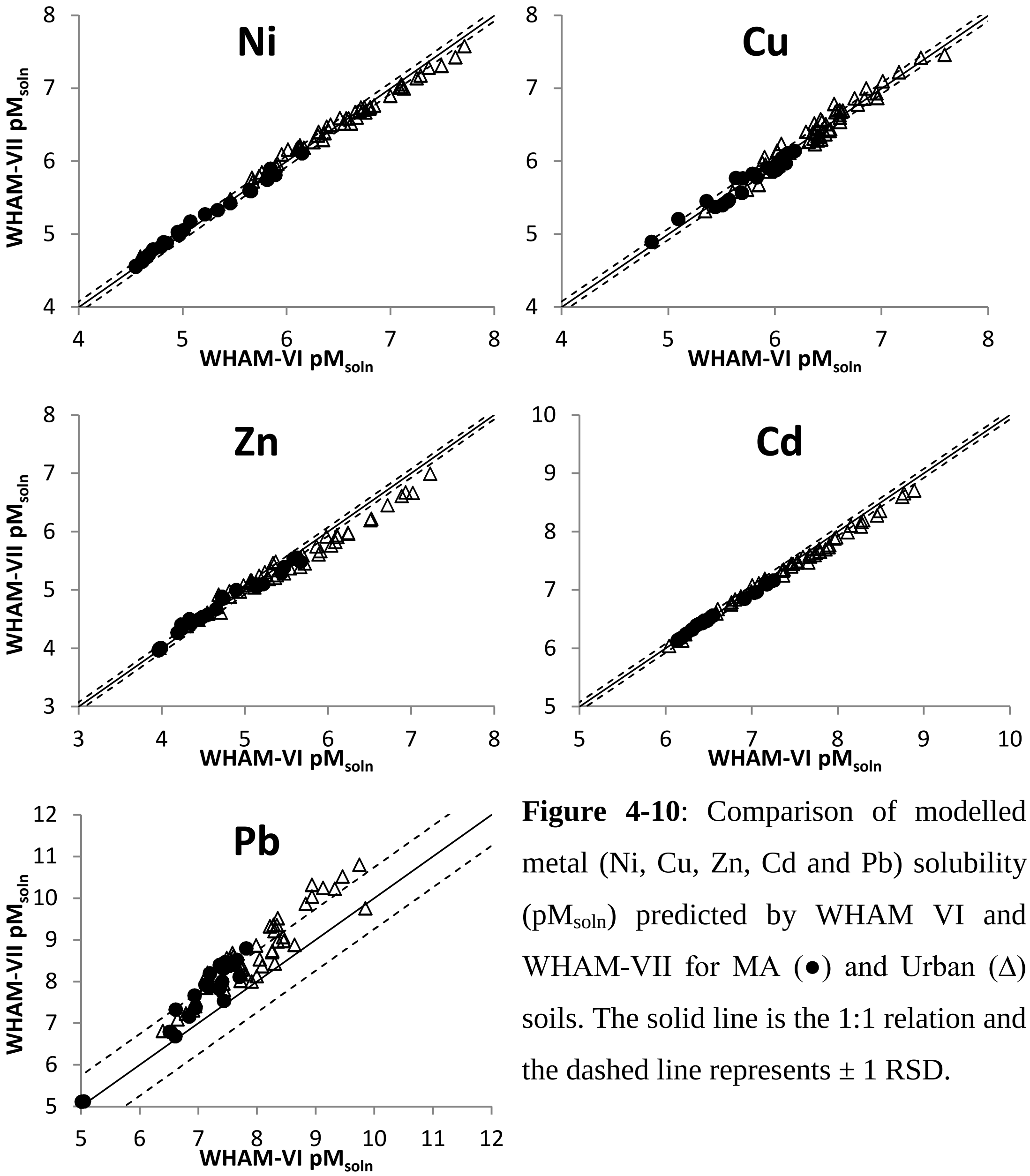


Figure 4-10: Comparison of modelled metal (Ni, Cu, Zn, Cd and Pb) solubility (pM_{soln}) predicted by WHAM VI and WHAM-VII for MA (●) and Urban (Δ) soils. The solid line is the 1:1 relation and the dashed line represents ± 1 RSD.

4.5 Conclusions

Both simple regression models and mechanistic geochemical speciation models can be used to predict trace metal solubility in whole soil suspensions. The empirical model may provide an apparently superior fit to the solubility data. However, such a comparison is probably unreasonable because the regression model coefficients are optimised to fit the data whereas the mechanistic model has been independently parameterised using studies on single, 'purified' geocolloidal adsorbents.

Furthermore, whereas the mechanistic model incorporates allowance for changing conditions, such as solid:solution ratio, metal ion competition, changes in anion complexation, temperature, ionic strength etc., the robustness of the regression model remains untested in this, and other, regards.

The difference between the MA and Urban soils illustrated that metal solubility is not only determined by soil characteristics but is also strongly affected by variation in the sources of contamination and possibly the duration of contact with a soil environment. The advantage of using E-values to represent the reactive metal fraction in predictions of solubility is very clear for WHAM-VII, but not for the ‘locally parameterised’ regression model. This is almost certainly due to strong links between pH and E-value becoming subsumed into the coefficient nominally describing the direct influence of pH on metal solubility.

Overestimation of the solution concentration from WHAM-VII was observed for all five metals, and was most strongly correlated with soil pH. Fractionation information from WHAM suggested that the source of the model’s underestimation of metal binding most likely lay with Fe and Mn oxides for Ni, Zn and Cd and HA for Cu. Prediction of metal solubility was improved by increasing the notional surface area of Fe oxides, however it is clear that a more systematic investigation of (i) model deficiencies and (ii) the potential for other mechanisms of metal ion retention in soils is required.

A comparison of WHAM versions VI and VII suggested very little difference for Cu, Cd and Zn and a marginal improvement for Ni. However it was clear that a substantial correction of the over-prediction of Pb solubility had been achieved by reformatting and re-parameterising the model in version VII.

5 CHAPTER 5: LABILITY OF LEAD IN SOIL: EFFECTS OF SOIL PROPERTIES AND CONTAMINANT SOURCE

5.1 Overview

Industrial pollution, combustion of leaded petrol and mining activities have increased environmental Pb concentrations since the Industrial Revolution. The Agency for Toxic Substances & Disease Registry (ATSDR, 2012) lists Pb as the second most dangerous substance that threatens human health, not only because of its high toxicity but also due to its wide dispersion around the globe (Nriagu, 1996; Watmough and Hutchinson, 2004; Bacon et al., 2006). Human exposure to Pb, via inhalation of particles and ingestion of contaminated soil, can result in physiological damage, particularly to the nervous system. Due to these health concerns, the commercial use of Pb has been considerably reduced over the last three decades and unleaded petrol introduced to reduce aerial Pb deposition (Erel et al., 2002; Flament et al., 1996; Johnson et al., 1995). However, soils retain a large reservoir of anthropogenic Pb, derived partly from atmospheric deposition (Emmanuel and Erel, 2002; Sterckeman et al., 2000). Young children are at greatest risk from ingestion of soil contaminated with Pb because of their compulsive hand-to-mouth response (Wixson and Davies, 1994) and their greater sensitivity to Pb toxicity (Kayhanian, 2012). Potentially, soil Pb may also enter the biogeochemical cycle through plant uptake, surface runoff and leaching into groundwater systems, depending upon the reactivity and solubility of the soil-borne Pb burden. Therefore, to fully quantify the risks associated with Pb it is useful to assay the labile pool of Pb in soil, which is more closely linked to Pb bioavailability and solubility than the total concentration in soil (Meers et al., 2007; Sauvé et al., 2000; Tongtavee et al., 2005).

The reactivity and solubility of Pb in soil is generally lowest in alkaline soils with high mineral oxide and organic carbon contents (see Chapter 3 and 4). It may also be limited by formation of discrete mineral phases such as anglesite ($PbSO_4$) or Pb jarosite [$Pb_{0.5}Fe_3(SO_4)_2(OH)_6$] in acidic environments (Ruby et al., 1996) and chloropyromorphite ($Pb_5(PO_4)_3Cl$) which has been widely reported (Lang and Kaupenjohann, 2003). Association of Pb with different soil fractions has been investigated using sequential extraction procedures (SEP) by several workers (Bacon et al., 2006; Emmanuel and Erel, 2002; Imperato et al., 2003; Liu et al., 2003; Teutsch et al., 2001; Thornton et al., 2008). However, the application of such results to risk assessment may be limited because labile Pb is unlikely to correspond to any single SEP fraction (Atkinson et al., 2011).

Lead has four stable isotopes, 204, 206, 207 and 208. The isotopes of ^{206}Pb, ^{207}Pb and ^{208}Pb are radiogenic, representing the end of decay chains of ^{238}U, ^{235}U and ^{232}Th respectively. Local variations in uranium and thorium content as well as difference in their half-life can therefore result in wide location-specific variation of Pb isotopic ratios (isotopic abundance), known as the Pb isotopic signature. Thus, the Pb isotopic signature in particles found in aerosols (e.g. Shiharata et al., 1980; Erel et al., 1997; Bacon et al., 1996), sediments (e.g. Graney et al., 1995; Monna et al., 1999) and soils (e.g. Komarek et al., 2007; Puchelt et al., 1993) can be used for approximate geo-location. More recently, several researchers have successfully determined the source of Pb in individual SEP extraction steps for soils receiving both natural (parent material) and anthropogenic Pb (typically from coal or leaded petrol) (e.g. Thornton et al., 2008; Komarek et al., 2007). It has been shown that Pb from parent material was primarily associated with the residual fraction whilst anthropogenic Pb was distributed amongst carbonate, humus and Fe-oxide phases (Emmaunuel and Erel, 2002; Teutsch et al., 2001).

This chapter represents an extension of work by Atkinson et al. (2011) who studied the Pb fractionation of 4 soils with distinctive metal sources and soil properties. The objective was to broaden the investigation of the effect of soil properties and sources of contamination on Pb lability in soils, as determined by isotopic dilution. Soils from three sites (108 samples in total) were analysed for chemical properties likely to affect Pb solubility, total Pb isotopic signature, fractionation by sequential extraction and Pb_E. The sites covered a wide range of soil properties and natural and anthropogenic sources of Pb including petrol (‘Rural Roadside’ soils), sewage sludge (‘Sewage Farm’ soils) and typical urban soils from a systematic geochemical survey (‘London’ soils). Regression was used to examine the influence of soil properties within each set of soils. The sources of Pb in both labile and non-labile pools in roadside soils were estimated by measuring the isotopic ratios $^{206}Pb/^{207}Pb$ and $^{208}Pb/^{207}Pb$. An attempt was made to quantify the relative contribution from two likely sources (indigenous coal and ore Pb and leaded petrol) to the labile (isotopically exchangeable) soil Pb fraction to determine whether Pb from petrol remains more labile than indigenous soil Pb despite the withdrawal of leaded petrol for in the UK several decades ago.

5.2 Material & Methods

5.2.1 Soil sampling

Three sets of soils, with different sources of Pb contamination, were used in this study. Twenty one topsoils (0-20 cm) were collected from 4 sites along a major road (the A6) leading to the M1 motorway in a rural area of Nottinghamshire, UK (52°48’N, 1°16’W), where soils were likely to have received petrol-derived Pb from heavy traffic (designated ‘Rural Roadside’). Associated sub-soils (below 20 cm depth) were sampled at two roadside sites: nine samples from three replicate cores at one site at three depth intervals (20-26, 26-30 and 30-40 cm) and twelve samples from triplicate cores at a second site, at 10 cm intervals between 20 and 60 cm. Sixteen topsoils were sampled from five fields at a sewage processing farm in Nottinghamshire, UK (52°57’N, 1°02’W) (designated ‘Sewage Farm’ soils). A further 50 topsoils (5-20 cm) were sub-sampled from the Geochemical Baseline Survey of the Environment (G-BASE) “London Earth” sample archive of the British Geological Survey (BGS) (Johnson et al., 2005). The ‘London’ soils selected were a subset from the systematic survey of the Greater London Authority area, chosen to cover 13 different land uses and a range of soil properties including pH, organic matter content and total Pb concentration.

5.2.2 Soil characterization

Soil samples were air-dried and sieved to < 2 mm. Soil pH was measured in 0.01 M $CaCl_2$ (1 : 2.5 soil : solution ratio) after shaking for 30 min. Soil organic matter content (SOM) was estimated from loss on ignition (%LOI) (Section 2.1.2.1). Available phosphate was determined using the Olsen method and a colorimetric assay (Rowell, 1994). Sub-samples of Rural Roadside and Sewage Farm soils were agate ball-milled and total Pb concentration was measured at the University of Nottingham (UoN) (Section 2.2.2). The Pb concentration in the London soil samples was determined on powder pellets by wavelength dispersive x-ray fluorescence (XRF) spectrometry using PANalytical MagiX-PRO PW2440 and Axios-Pro spectrometers at BGS, each fitted with 4kW Rh-anode x-ray tubes. The manufacturer's SuperQ software was used to account for matrix effects and correct for spectral line overlap interference. The XRF lower detection limit was 1.3 mg kg^{-1}, the calibration extended to 1,000 mg kg^{-1} and data were reported up to 10,000 mg kg^{-1}. Three of the London soils, in which total Pb concentrations exceeded 10,000 mg kg^{-1} measured by XRF, were assayed by ICP-MS following acid digestion at the UoN.

5.2.3 Measurement of Pb isotopic abundances

The isotopic abundances of Pb isotopes (IA) in the HF/$HClO_4$/HNO_3 acid digestates of Rural Roadside and Sewage Farm soils were measured by ICP-MS (Thermo-Fisher Scientific, model X-Series^{II}) at the UoN. The London soils were assayed at the BGS (Agilent, model 7500cx). The analytical method and instrument settings used at the UoN are described in Chapter 2 (Section 2.3) and at BGS in Izquierdo et al, (2012). Both instruments employed a large number of sweeps with short quadrupole dwell times, to minimise plasma flicker, and used repeated assays of the Pb isotope CRM NIST 981 to drift-correct mass bias effects (Baker et al., 2004). The known Pb isotopic ratios (^{206}Pb/^{207}Pb and ^{208}Pb/^{207}Pb) in leaded petrol, UK Pb ore and UK coal, were used to assess the likely sources of Pb in the soils. The ^{206}Pb/^{207}Pb and ^{208}Pb/^{207}Pb ratios for petrol-derived Pb have been estimated at 1.067 ± 0.007 and at 2.340 ± 0.011 respectively (Monna et al., 1997). The Pb isotopic signature of galenas (PbS) from Derbyshire and Leicestershire were considered as representative for ore Pb with isotopic ratio values of $^{206}Pb/^{207}Pb$ = 1.182 ± 0.004 and $^{208}Pb/^{207}Pb$ = 2.458 ± 0.002 (Rohl, 1996). For Pb in UK coal published average isotopic ratios for England and Wales were used (Farmer et al., 1999): $^{206}Pb/^{207}Pb$ = 1.184 ± 0.005 and $^{208}Pb/^{207}Pb$ = 2.461 ± 0.012. UK ore and coal are also used here as a proxy for soil parent material. The ratio of $^{206}Pb/^{207}Pb$ is generally adopted as the most sensitive to change in environmental studies (Bacon et al., 1996) due to the similar isotopic abundance of the isotopes minimising instrumental bias and maximising precision.

The Pb isotopic abundances of the labile fraction for all soils were measured in the solution phase of 1g soil (< 2 mm) suspended in 30 ml of 0.01 M $Ca(NO_3)_2$ for three days. Quality assurance was confirmed by measuring the isotopic ratios $^{206}Pb/^{207}Pb$ and $^{208}Pb/^{207}Pb$ in 8 equilibrated $Ca(NO_3)_2$ solutions at both institutions (UoN and BGS); the mean ratio of $^{206}Pb/^{207}Pb$ values (UoN/BGS) was 1.001 with SD = 0.002.

5.2.4 Exponential model for Pb isotopic ratio in soil profile

An exponential model for the change in the isotopic ratio $^{206}Pb/^{207}Pb$ of soil Pb with depth was fitted to profile data from the Rural Roadside soils. The predicted isotopic ratio throughout the profile was calculated from Eq. 5-1.

$$IR_D = IR_{PM} + \left(IR_0 - IR_{PM}\right)\exp^{(-kD)} \qquad (5\text{-}1)$$

where IR_0 and IR_{PM} are the isotopic ratios of Pb at the surface (0 cm) and in the soil parent material respectively, k is an exponential constant, IR_D is the isotopic ratio ($^{206}Pb/^{207}Pb$) of total Pb in the soil at a specified depth (D, cm). The parameters IR_0,

IR_{PM} and k were optimized for the best fit to measured values of IR_D using the *Solver* function in Microsoft Excel.

5.2.5 Lead lability measured by isotopic dilution

The isotopic dilution method used for measuring E-values for Pb (Pb_E) is described in Chapter 2 (Section 2.3). The isotopic compositions of the labile and non-labile Pb fractions were determined to investigate the source of Pb in each case. The isotopic abundance in the non-labile pool was calculated using Eq. 5-2 showing ^{206}Pb as an example:

$$^{206}\mathrm{IA}_{\mathrm{NL}} = \frac{^{206}\mathrm{IA}_{\mathrm{total}} - \left(^{206}\mathrm{IA}_{\mathrm{L}} \times \mathrm{P}_{\mathrm{L}}\right)}{\left(1 - \mathrm{P}_{\mathrm{L}}\right)} \qquad (5\text{-}2)$$

where $^{206}IA_L$ and $^{206}IA_{NL}$ are the isotopic abundances of ^{206}Pb in the labile (0.01 $Ca(NO_3)_2$ extracts) and non-labile (NL) pools respectively; P_L is the molar proportion of labile Pb in the soil calculated from the value of $\%Pb_E$.

The isotopic ratios (^{206}Pb/^{207}Pb and ^{208}Pb/^{207}Pb) of petrol Pb and coal Pb were used to estimate the proportion of each source of Pb in the labile and non-labile pools. The assumption that Pb came only from these two sources is a simplification but it indicates whether Pb from petrol is more labile than Pb from local parent material which has an isotopic signature similar to that of UK coal or Pb ore minerals. The proportion of labile Pb in the petrol-derived Pb pool was estimated by linear interpolation of isotopic ratios (Eq. 5-3):

$$\%\mathrm{Pb}_{\mathrm{E(petrol)}} = \frac{\left(\dfrac{\mathrm{IR}_{\mathrm{coal}} - \mathrm{IR}_{\mathrm{L}}}{\mathrm{IR}_{\mathrm{coal}} - \mathrm{IR}_{\mathrm{petrol}}}\right) \times \mathrm{Pb}_{\mathrm{E}}}{\left(\dfrac{\mathrm{IR}_{\mathrm{coal}} - \mathrm{IR}_{\mathrm{total}}}{\mathrm{IR}_{\mathrm{coal}} - \mathrm{IR}_{\mathrm{petrol}}}\right) \times \mathrm{Pb}_{\mathrm{total}}} \times 100\% \qquad (5\text{-}3)$$

where $\%\mathrm{Pb}_{\mathrm{E(petrol)}}$ is the proportion (%) of petrol-derived Pb that is isotopically exchangeable, IR_{L} and $\mathrm{IR}_{\mathrm{total}}$ are the isotopic ratios (^{206}Pb/^{207}Pb or ^{208}Pb/^{207}Pb) of labile Pb (0.01 M $Ca(NO_3)_2$ extracts) and total soil Pb ($HF/HClO_4/HNO_3$ acid digestion) respectively; $\mathrm{Pb}_{\mathrm{total}}$ is total Pb concentration (mg kg^{-1}) measured by acid digestion. Equation 5-3 is an extension of a widely used simple binary mixture model, based on interpolation between the isotopic ratios of prescribed end-members, discussed by Komárek et al., (2008). It has been estimated that this approach produces a maximum error of about 2.0% at the midpoint across the full range of possible source compositions (0 – 100% petrol-derived Pb).

5.2.6 Predicting the labile fraction of Pb from soil properties

The relationship between labile fraction of Pb (Pb_{E}) and soil characteristics including pH, %LOI, available phosphate ($\mathrm{P}_{\mathrm{avail}}$, mg kg^{-1}) and total Pb concentration ($\mathrm{Pb}_{\mathrm{total}}$, mg kg^{-1}) in soils was investigated using multiple regression (Eq. 5-4); the constants (k_0 – k_4) were optimized using the stepwise regression function in *Minitab version 16.2.2*.

$$\mathrm{LogPb}_{\mathrm{E}} = \mathrm{k}_1\log_{10}\mathrm{Pb}_{\mathrm{total}} + \mathrm{k}_2\mathrm{pH} + \mathrm{k}_3\%\mathrm{LOI} + \mathrm{k}_4\mathrm{P}_{\mathrm{avail}} \qquad (5\text{-}4)$$

Correlation coefficients between measured and modelled values of Pb_{E} and residual standard deviations for the model fit were used to assess the contribution of each soil variable to the prediction of labile Pb.

5.2.7 Sequential extraction of Pb

A sequential extraction procedure (SEP, adapted from Li and Thornton, 2001) was applied to 32 soil samples selected to cover a range of values of the variables in Eq. 5-4 (Pb_{total}, pH, %LOI and P_{avail}). Soils included 10 Rural Roadside (both topsoils and subsoils), 12 Sewage Farm and 10 London soils with a range of land uses. Full details of the SEP are described in Chapter 2 (Section 2.2.3).

5.3 Results and discussion

5.3.1 Soil characteristics

Soil characteristics are shown in Table 5-1. Rural Roadside sites had a wide range of pH values (4.6-8.5) with slightly lower %LOI in the subsoil. Available phosphate concentration and total Pb concentration were relatively low compared to the other two sets of soils. Average Pb concentration in top soils was approximately double that in subsoils suggesting anthropogenic inputs. Sewage Farm soils had a narrower pH range (5.8-7.0), higher %LOI (mean=16%) and very high available phosphate content (53-380 mg kg^{-1}). Lead concentrations varied from 55-712 mg kg^{-1} with lower concentrations in fields where pH was slightly higher and %LOI and available phosphate were lower suggesting lower historical sewage sludge inputs. The G-BASE dataset for soils in Nottinghamshire (BGS; 636 samples) gives a background Pb concentration range from 13.8 to 976 (mg kg^{-1}) with an average of 145 mg kg^{-1} and median of 100 mg kg^{-1}. The London soils covered a wide pH range (3.3-7.3) and had similar values of %LOI (mean = 9.7%) to the Rural Roadside soils. Available phosphate was higher than for the Rural Roadside soils but much lower than for soils from the Sewage Farm. Lead concentrations were very high in the London soils (median= 940 mg kg^{-1}) and showed greater variation (99-22600 mg kg^{-1}) between different land uses, than soils sampled elsewhere. Many urban sites in the UK are heavily contaminated with metals; the G-BASE dataset for urban soils (BGS; 13583 samples gives a median of 128 mg kg^{-1} and a range of 2.1 to 10,000 mg kg^{-1}. The latter figure is certainly exceeded and simply represents the maximum quantification limit for XRF.

Table 5-1: Land use and soil characteristics for the three site locations studied.

		pH			Loss on ignition (%)			Available phosphate (mg kg^{-1})			Total Pb content (mg kg^{-1})		
Land use	**n**	**Mean**	**Min**	**Max**	**Mean**	**Min**	**Max**	**Mean**	**Min**	**Max**	**Mean**	**Min**	**Max**
Rural Roadside	**42**	**5.67**	**3.92**	**7.57**	**8.31**	**2.59**	**20.1**	**7.88**	**1.92**	**29.7**	**51.5**	**14.0**	**233**
Surface	21	5.71	4.17	7.01	11.8	7.05	20.1	9.31	1.92	29.7	65.7	25.1	233
Subsoil	21	5.62	3.92	7.57	4.84	2.59	8.20	6.44	1.99	26.3	37.2	14.0	161
Sewage Farm	**16**	**6.32**	**5.86**	**6.74**	**16.2**	**5.42**	**29.3**	**194**	**53.0**	**380**	**390**	**55.4**	**712**
Field 1	4	6.29	6.14	6.57	10.9	9.60	11.8	167	142	190	273	229	324
Field 2	2	6.66	6.58	6.74	6.47	6.38	6.56	138	128	147	78.8	55.4	102
Field 3	6	6.14	5.86	6.47	24.7	19.7	29.3	283	182	380	610	433	712
Field 4	2	6.44	6.34	6.53	21.4	19.5	23.4	171	126	216	592	511	673
Field 5	2	6.49	6.44	6.53	5.78	5.42	6.14	61.1	53.0	69.1	74.8	67.8	82.0
London	**50**	**6.58**	**3.29**	**7.31**	**9.70**	**4.25**	**17.5**	**22.0**	**1.09**	**207**	**1990**	**99.2**	**22600**
Commercial and residential	4	7.05	6.93	7.14	9.55	6.66	14.4	18.1	10.5	25.8	1850	166	3970
Deciduous woodland established	2	5.11	3.29	6.92	16.9	16.6	17.3	21.6	14.7	28.5	1220	162	2280
Domestic Garden	22	6.73	4.95	7.25	9.02	5.35	16.9	13.5	1.09	42.4	1850	172	7210
Grass moor	2	6.12	5.00	7.24	5.10	4.25	5.95	1.61			548	99.2	996
Graveyard	1	6.76			5.45			n.a			115		
Hospital Grounds	1	7.31			6.11			13.5			420		
Major roads/verge	2	6.39	5.57	7.20	9.07	8.98	9.17	15.8	14.6	17.0	468	428	507
Minor metalled roads/verge	3	6.23	5.56	6.69	12.3	7.89	17.5	122	37.2	207	755	213	1730
Park	4	6.00	5.28	6.88	10.9	9.50	12.4	19.1	6.31	28.2	3260	217	9040
Recreational area	3	6.92	6.70	7.07	12.0	9.73	15.4	33.7	1.41	80.8	3330	641	6600
School	1	5.98			8.27			2.25			193		
Urban open space	2	6.51	6.40	6.61	12.0	9.90	14.0	n.a.			370	205	534
Urban open space tended but unproductive	3	7.00	6.78	7.30	8.59	4.96	11.5	27.3	19.8	34.9	3850	123	22600

n.a. not available (limited amount of sample)

The isotopic characteristics of the 108 soils are shown in Fig. 5-1a) alongside typical values for leaded petrol, UK ore and UK coal. The range of values measured indicates that Pb in the soils investigated had multiple origins (Bacon et al., 1996). The majority of samples fell close to a mixing line, formed by a plot of the isotopic ratio $^{208}Pb/^{207}Pb$ against $^{206}Pb/^{207}Pb$, which strongly suggests that soil Pb originated from the parent material and leaded petrol sources used as indicative end members.

The Rural Roadside subsoils had isotopic signatures for Pb that were close to those of UK coal, with more than 75% of the soils falling within the range delimited by Pb ore and coal (Fig. 5-1b). This is as expected considering the joint influences of the local Derbyshire Pb ore field and Nottinghamshire-Leicestershire coalfields on alluvial and fluvio-glacial deposits in the Trent Valley and surrounding areas. In contrast, Rural Roadside topsoils had isotopic signatures dominated by petrol Pb at the surface with decreasing petrol Pb contributions with depth (Fig. 5-2). The exponential model (Eq. 5-1) fitted the measured values at both roadside sites showing that Pb had an isotopic signature tending towards that of petrol towards the soil surface whereas, at depth, the isotopic ratio of the parent material (IR_{PM}: site 1 = 1.188 and site 2 = 1.186) were in the range of indigenous coal and ore Pb (1.174 - 1.189). This may suggest a slow movement of petrol-derived Pb into the subsoil as reported in many other studies (e.g. Bacon et al., 1996; Erel et al., 1997; Teutsch et al., 2001; Bacon and Hewitt, 2005). The difference in the optimized constant (k = 0.11 and 0.08) between the two profiles may be a result of soil pH with more movement of petrol-derived Pb in the soil profile with the lower pH. However, given the low solubility of Pb in soil it is also likely that physical movement (e.g. earthworm activity) has contributed to the downward movement of petrol-derived Pb.

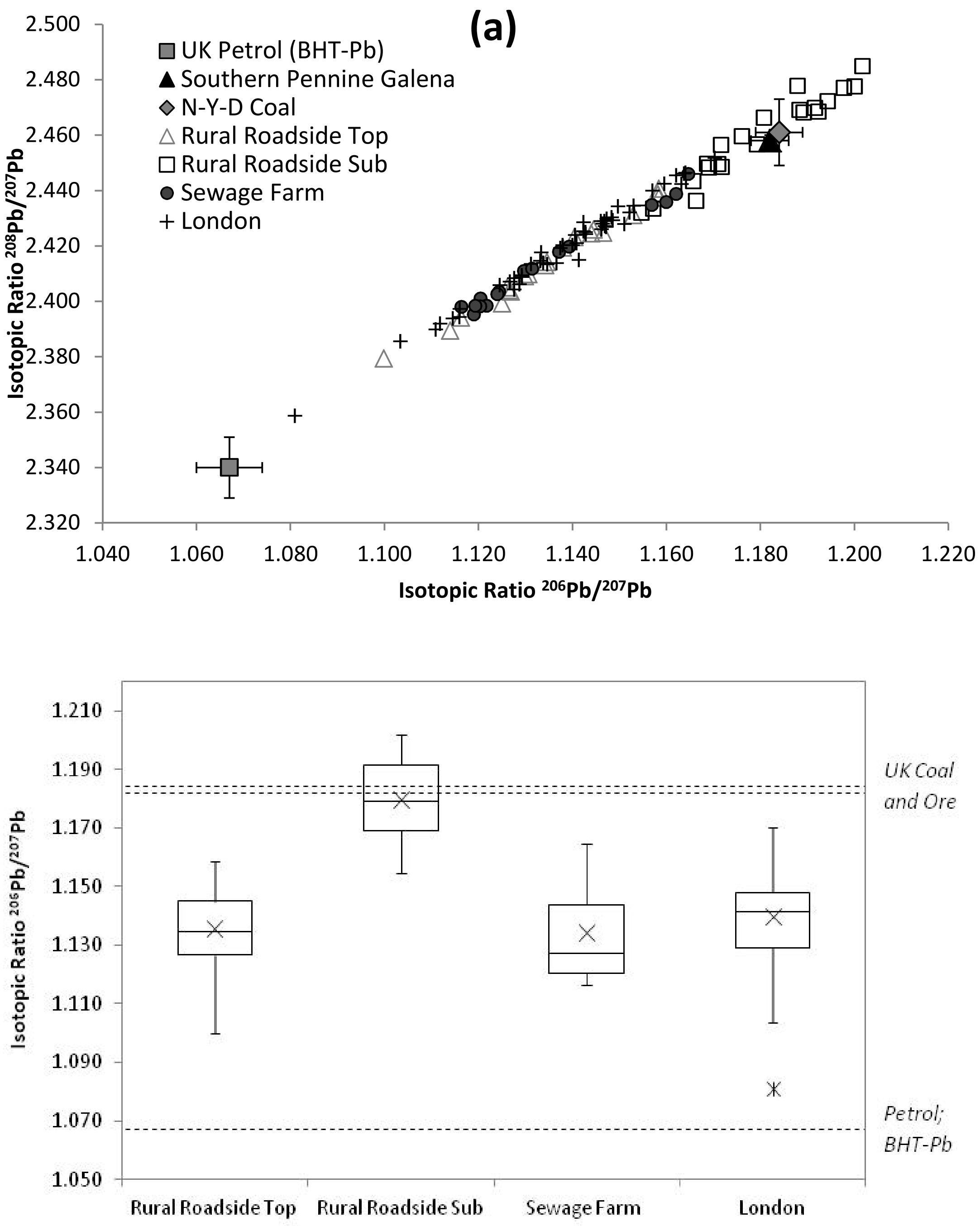


Figure 5-1: **a)** Lead isotopic characteristics in soils from three sites: Rural Roadsides (n = 42) (top- and sub-soils), a Sewage Farm (n = 16) and the G-BASE London Earth geochemical survey (n = 50). Isotopic ratios ^{206}Pb/^{207}Pb and ^{208}Pb/^{207}Pb are shown as a mixing line with median values for UK coal (Nottinghamshire-Yorkshire-Derbyshire, Farmer et al., 1999), South Pennine Pb ore and leaded petrol shown for comparison. **b)** Lead isotopic ratio ^{206}Pb/^{207}Pb shown as a box and whisker plot for the three sites; the higher and lower quartile represents the highest and lowest 25% of the data respectively; whiskers indicates variability outside the upper and lower quartiles; the horizontal broken lines show values for the three reference sources, the mean value (×) and outliers are marked as an asterix.

The majority of London soils (Fig. 5-1a), fell in a range similar to that of the Rural Roadside topsoils (1.127 – 1.146) and consistent with values reported for London aerosols of c.1.135 (Noble et al, 2008), suggesting a similar mix of Pb from both indigenous coal/ore and petrol sources. The origins of Pb in urban soils are difficult to identify unequivocally by isotopic ratio because there are so many possible sources. McGill et al. (2003) suggested that Pb in urban soils may come from paint, roofing materials, water pipes, coal burning, etc., and each manufactured source may be a mix of parent sources. In the Sewage Farm soils, fields with lower Pb concentrations, indicating lower sludge inputs, showed a greater influence from indigenous Pb (UK ore or coal) whereas soils with greater Pb contents showed a stronger influence of petrol-derived Pb, suggested by Atkinson et al. (2011) to arise from run-off to sewers from roads. There was therefore a strong correlation (r = -0.92) between the isotopic ratio and the total Pb content in the Sewage Farm soils which implies a single source of background (indigenous) soil Pb and a consistent isotopic signature in the sewage sludge input (closer to petrol). The same situation may broadly apply to the Rural Roadside topsoils (r = -0.72) and this is supported by the depth profile shown in Fig. 5-2. For London soils however, the correlation coefficient between isotopic ratio and total Pb content was not significant, suggesting a more complex mix of sources. Farmer et al. (2011) studied the relationship between total Pb concentration and the isotopic ratio ^{206}Pb/^{207}Pb in 27 urban soils in the city of Glasgow, Scotland and also suggested that the isotopic ratio of total Pb represented a complex mixture of Pb deposited from a variety of sources since the Industrial Revolution.

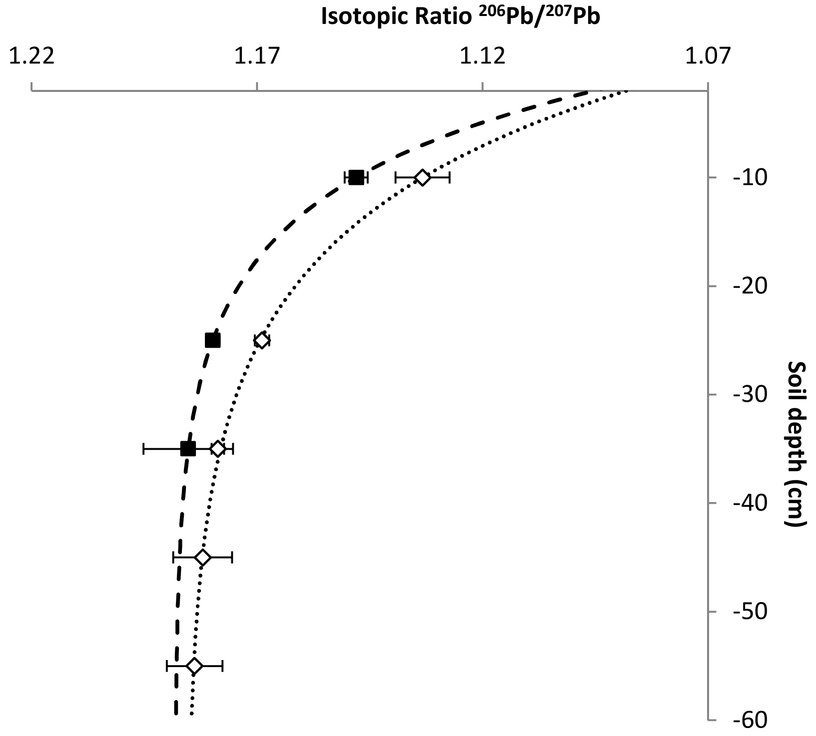
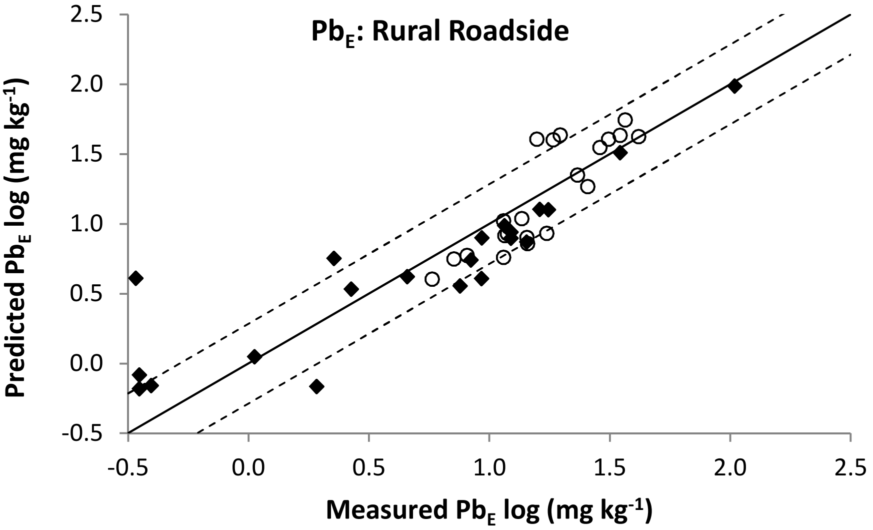


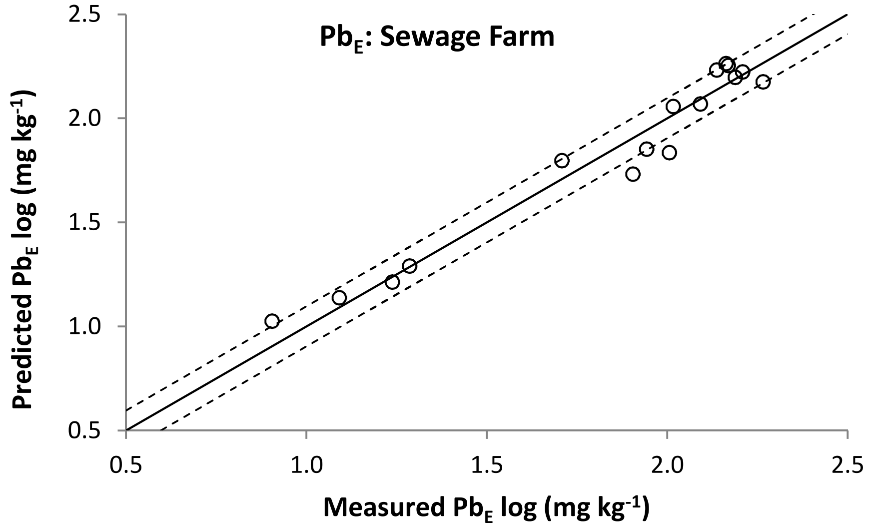
Figure 5-2: Measured and predicted values of Pb isotopic ratio $^{206}Pb/^{207}Pb$ in two Rural Roadside soil profiles. The average soil pH values were pH 7.9 for site 1 (■) and pH 5.1 for site 2 (◊). The line indicates the fit of Eq. 5-1. Error bars show standard error of three replicates.

5.3.2 The effect of soil properties and contaminant sources on Pb lability

In a study of Pb bioaccessibility, the source of Pb was found to be less important than the concentration and physico-chemical influences of soil properties (Farmer et al., 2011). In the current investigation, the median Pb labilities (%Pb_E) measured for the Rural Roadside, Sewage Farm and London soils were quite similar at 21%, 24% and 30% respectively. However, Figure 5-3 also shows a wider range of Pb lability in the Rural Roadside soils than in both the Sewage Farm and London soils. Assuming a similar Pb source from parent material and contamination dominated by petrol-derived Pb for the Rural Roadside and Sewage Farm sites, the difference in lability between these two sets of soils is likely to be determined by soil properties. Thus, an attempt was made to predict Pb_E from total Pb concentration and soil properties including pH, %LOI and available-P using regression (Eq. 5-4). Comparisons of predicted and measured Pb_E for the soils are shown in Fig. 5-4.

Figure 5-3: Box and whisker plot showing lability of Pb (%Pb_E) for soils from Rural Roadside (n=42), Sewage Farm (n=16) and London (n=50) sites; the higher and lower quartile represents the highest and lowest 25% of the data respectively; whiskers indicates variability outside the upper and lower quartiles; the mean value (×) and outliers (asterix) are also shown.





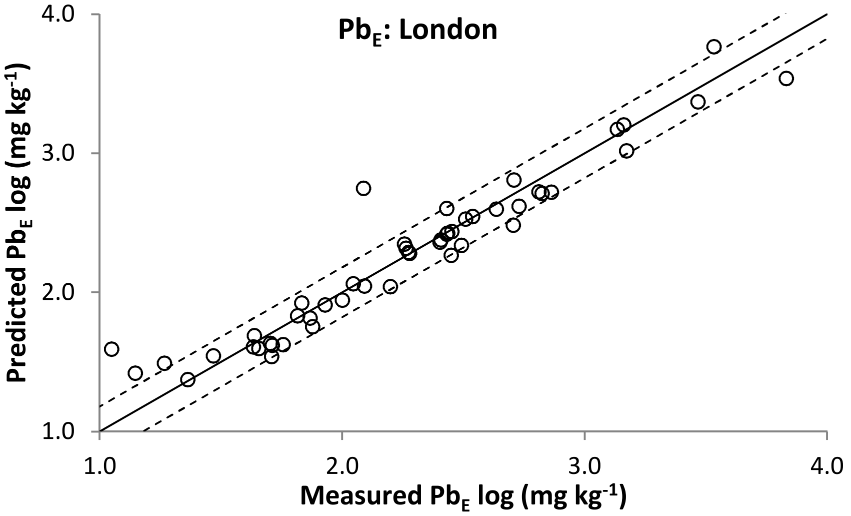


Figure 5-4: Prediction of Pb_E (mg kg^{-1}; log$_{10}$ scale) in soils from Rural Roadside, Sewage Farm and London sites from the soil variables Pb_{total}, pH and %LOI (Eq. 5-4). For the Rural Roadside sites both topsoils (○), and subsoils (♦) are shown.

Stepwise regression showed that soil pH was the dominant parameter in predicting Pb_E in Rural Roadside soils, accounting for 40.6% of the total variance. However, for the Sewage Farm and London soils, with their restricted range of pH values, it was clear that soil Pb content was the primary determinant of Pb_E, accounting for 96% and 90% of the variance respectively. The other soil characteristics provided marginal improvements to the model fitting. Table 5-2 gives the values of the optimised coefficients, RSD and correlation coefficients associated with combinations of the soil parameters in Eq. 5-4. Available P (k_4 in Eq. 5-4) was found to be non-significant in improving the prediction Pb_E for all three sets of soils.

Table 5-2: Linear regression coefficient, RSD and R^2 values of equations for model prediction of Pb_E ($\log_{10}$ scale). The regression (Eq. 5-4) was parameterized from values of Pb_E (mg kg^{-1}) measured on soils from the Rural Roadside, Sewage Farm and London sites separately.

Roadside	**k_1 ($\log_{10}Pb_{total}$)**	**k_2 (pH)**	**k_3 (%LOI)**	**RSD**	**R^2**
	0.61	-	-	0.51	0.38
	1.47	-0.26	-	0.29	0.76
	1.29	-0.24	0.024	0.29	0.78
Sewage Farm					
	0.75	-	-	0.17	0.96
	1.08	-0.13	-	0.10	0.96
London					
	0.79	-	-	0.25	0.90
	1.04	-0.12	-	0.20	0.90
	1.13	-0.12	-0.03	0.18	0.92

Calcareous subsoils from the Rural Roadside sites, had the lowest $\%Pb_E$, ranging from 0.6-12.7%, where Pb from parent material is likely to be fixed within soil particles or may be in the form of discrete Pb minerals such as cerrusite ($PbCO_3$) or pyromorphite (Li and Thornton, 2001). There was a greater proportion of labile Pb in topsoils than in subsoils, as a result of petrol-derived Pb input, which is likely to be more labile than Pb from parent material. Lead-containing particles from petrol would be expected to dissolve in soils and adsorb to geocolloidal phases. Teutsch et al. (2001) found that petrol Pb was predominately bound to carbonate and Fe-(hydr)oxides, but presumably this will depend on soil composition. Values of Pb_E were over-predicted for deeper subsoils with high pH values (>7.5) and low amounts of labile Pb (< 0.4 mg kg^{-1}), probably originating almost entirely from soil parent material.

Although sources of Pb contamination, in the Sewage Farm soils may have been similar to those from the Rural Roadside sites, 75% of $\%Pb_E$ fell in the lower range of 20-27% (Fig. 5-3). This was probably because of the relatively constant pH in these soils and their extraordinarily high phosphate concentration (Tye et al., 2003). Contaminant Pb in the Sewage Farm soils is mainly from a single source, sewage sludge; the history of sludge application also determines the concentrations of soil humus and available phosphate. There is therefore a strong co-variance between the soil Pb content and the factors likely to affect its lability (Brazauskiene et al., 2008): soil Pb content was highly correlated with both %LOI (r= 0.97) and P_{avail} (r= 0.75). This, coupled with an on-site liming policy to maintain a limited range of soil pH values, around pH 6.5 (Severn Trent Ltd, pers. comm.), probably explains the very restricted range of $\%Pb_E$ values observed so that Pb_{total} explained 96% of the variation in Pb_E.

In the fifty London soils, although land use, soil properties and possible sources of Pb all covered a wider range than at the other two sites, the range of Pb lability was relatively small with 75% of the %E-values falling in the range 16−26% (Fig. 5-3).

This was a larger range than found in the Sewage Farm soils but much smaller than seen for the Rural Roadside soils and so soil properties had only a minor effect on E-values with Pb_{total} accounting for 90% of the variation in Pb_E (Table 5-2). Appleton et al. (2012) studied the bioaccessibility of Pb on the same subset of London soil samples and concluded that total Pb concentration was the single significant predictor of bioaccessible Pb. Soil pH, which accounted for only 0.7% of the variation in Pb_E and an improvement in RSD of just 0.05, was quite high (mean pH=6.6; SD = 0.75), which may have contributed to the relatively narrow range of $\%Pb_E$ values. However, it is difficult to draw general conclusions regarding the factors controlling Pb lability in the London soils because of the complex mix of historical and current Pb sources and land uses. The highest level of lability found in a London soil (74.9%), was from a park planted with deciduous woodland with an exceptionally low pH value of 5.28.

5.3.3 Fractionation of lead as measured by sequential extraction (SEP)

Figure 5-5 shows the mean proportion of Pb in each SEP fraction and the proportion of Pb that was labile (isotopically exchangeable) or non-labile. The SEP results indicate that the ‘inert’ residual pool (F5) was the largest fraction for all three sets of soils (37.2% for Rural Roadside soils, 77.6% for Sewage Farm soils, and 41.8% for London soils), which corresponds qualitatively with the results from other studies of Pb in contaminated soils (Thums et al., 2008; Thornton et al., 2008; Liu et al., 2003). The remaining Pb was bound mainly to Fe and Mn oxides (F3) and to humus (or sulphide) (F4). There was no consistent agreement between Pb_E and any single SEP fraction, as also found for Pb by Atkinson et al. (2011) and for Cd by Ahnstrom and Parker (2001). However, in the Sewage Farm soils, the SEP residual fraction (78%) could be identified closely with the non-labile pool of Pb (76%). This may be because the high concentration of phosphate in these soils caused formation of a discrete Pb-phosphate phase which was not accessible to any of the SEP extractants. For both the Rural Roadside and London soils, however, Pb bound into other fractions also contributed to the non-labile pool. Non-labile Pb in the Rural Roadside soils (68%) corresponded quite well with a combination (70%) of F3 (oxides) + F5 (residual) of the SEP. A similar situation applied for most of the London soils, where the non-labile fraction (76%) also corresponded with F3 and F5 combined (74%) by omitting one exceptionally acidic soil (pH 3.29). It is perhaps less likely that humus-bound Pb contributes substantially to the non-labile Pb fraction (Atkinson et al., 2011).

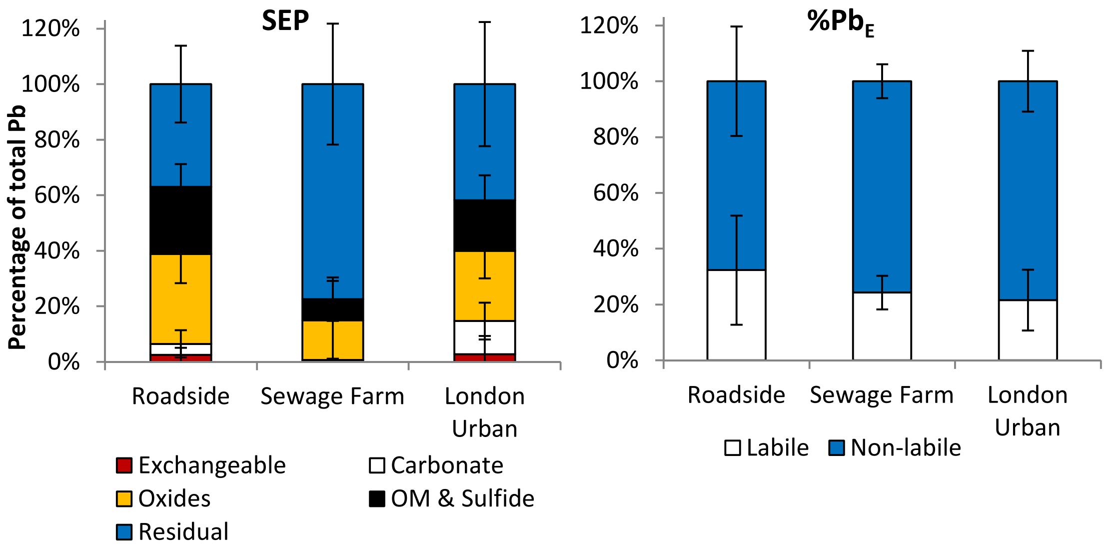


Figure 5-5: Comparison of average soil Pb fractions (%), determined by a sequential extraction procedure (SEP), with isotopically exchangeable Pb (%Pb$_E$) for a sub set of soils from Rural Roadside, Sewage Farm and London sites. Error bars represent standard deviation of data.

5.3.4 Differences between the Pb isotopic signature of labile and non-labile pools

Previous studies have suggested that Pb from petrol and coal combustion particulates is more soluble in dilute acid extractions than Pb from parent material which tends to be associated with the residual pool of SEPs (Erel et al., 1997; Li et al., 2011). In the current study, the Rural Roadside soils provided the best evidence of a link between isotopic signature and Pb$_E$, suggesting incomplete mixing of petrol-derived Pb with the indigenous soil Pb pool. In Fig. 5-6, for Rural Roadside soils, the ratio of ^{206}Pb/^{207}Pb in the non-labile fraction to ^{206}Pb/^{207}Pb in the labile fraction for roadside soils is shown as a function of ^{206}Pb/^{207}Pb in the whole soil. The difference in the proportion of petrol-derived Pb in topsoils and subsoils is clear from their respective positions on the x-axis: for sub soils all ^{206}Pb/^{207}Pb ratios were ≥ 1.15 and for top soils, the values were all ≤ 1.16. Complete mixing of petrol-derived and indigenous soil Pb would produce a uniform value of 1.0 on the Y-axis for all soils. Where the ratio is > 1, this suggests that petrol-derived Pb is over-represented in the labile fraction compared to the non-labile Pb fraction. Figure 5-6 indicates that the labile Pb fraction in both topsoils and subsoils contained a greater proportion of petrol-derived Pb than was found in the non-labile fraction. This suggests that petrol-derived Pb had moved down the soil profile and contributed to the labile fraction, either by leaching in solution or through physical mixing of the soil; a similar trend has been seen in alluvial soils in the English Midlands (UK) by Izquierdo et al. (2012).

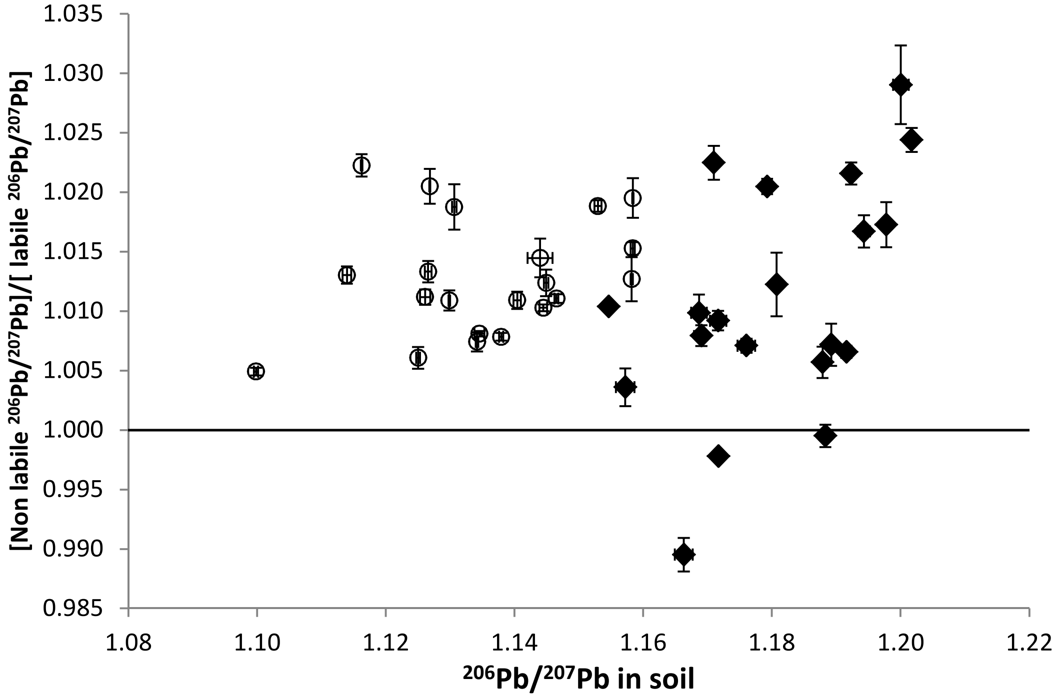


Figure 5-6: The ratio of ^{206}Pb/^{207}Pb in the non-labile Pb fraction to ^{206}Pb/^{207}Pb in the labile Pb fraction for Rural Roadside soils as a function of the isotopic ratio ^{206}Pb/^{207}Pb in the whole soil (from acid digestate) for topsoils (○), and subsoils (♦). Error bars show the standard error of two replicates.

Nevertheless, the labile and non-labile Pb fractions in topsoils did not simply align with the isotopic signatures of petrol-derived Pb and indigenous coal or ore sources respectively. Table 5-3 summarizes the average values of $^{206}Pb/^{207}Pb$ in labile and non-labile soil Pb fractions at the four Rural Roadside sites. Clear evidence of fixation of petrol-derived Pb in the topsoils is shown by displacement of $^{206}Pb/^{207}Pb$ ratios for *non-labile* Pb towards that of petrol-derived Pb. Similarly, Pb from parent material clearly contributed to the labile fraction in both topsoils and subsoils, presumably released by weathering processes (Emmanuel and Erel, 2002). The average $^{206}Pb/^{207}Pb$ ratio in the non-labile fraction in subsoils, however, does not suggest substantial fixation of petrol-derived Pb at depth, whilst other studies have suggested that anthropogenic Pb in sub-soils may be found fixed within (SEP) residual phases (Teutsch et al. 2001).

Table 5-3: Average values of the isotopic ratio $^{206}Pb/^{207}Pb$ in labile and non-labile Pb fractions in soils from the Rural Roadside sites.

	Surface soils		**Sub-soils**	
Site	$^{206}Pb/^{207}Pb$ Labile	$^{206}Pb/^{207}Pb$ Non labile	$^{206}Pb/^{207}Pb$ Labile	$^{206}Pb/^{207}Pb$ Non labile
1	1.133	1.140	1.167	1.183
2	1.122	1.133	1.173	1.182
3	1.148	1.168	n.a.	n.a.
4	1.112	1.125	n.a.	n.a.

n.a. not available

Figure 5-7 shows the proportion of labile Pb (%Pb_E) in petrol-derived and indigenous coal fractions (from Eq. 5-3) for the three sites as a box and whisker diagram. In Rural Roadside soils, there was a significant difference in Pb lability between petrol-derived Pb (40%) and indigenous coal Pb (29%) (paired t-test). There was a relatively strong correlation between soil pH and the proportion of petrol-derived Pb that was labile ($p<0.001$, $r = -0.55$). Nevertheless, the data again clearly show that both sources of Pb contribute to both the labile and non-labile pools of soil Pb. For Sewage Farm and London soils, the values of %Pb_E for Pb originating from petrol and indigenous coal were not significantly different. For some of the Sewage Farm soils, as suggested by Atkinson et al. (2011), the co-existence of large phosphate concentrations may cause rapid fixation of petrol-derived Pb during sewage sludge processing which then contradicts the pattern seen in the Rural Roadside soils where petrol-derived Pb was clearly more labile. For the London soils, despite having the widest range of possible Pb sources (Fig. 5-1), the proportions of petrol- and indigenous coal Pb that were isotopically exchangeable were very close (average of 23.1% and 22.1% respectively). This suggests a remarkably consistent level of assimilation into the soil and a geochemical "ageing" to a similar reactivity for Pb from all sources and is consistent with the narrow range of Pb_E seen across this dataset (Fig. 5-3).

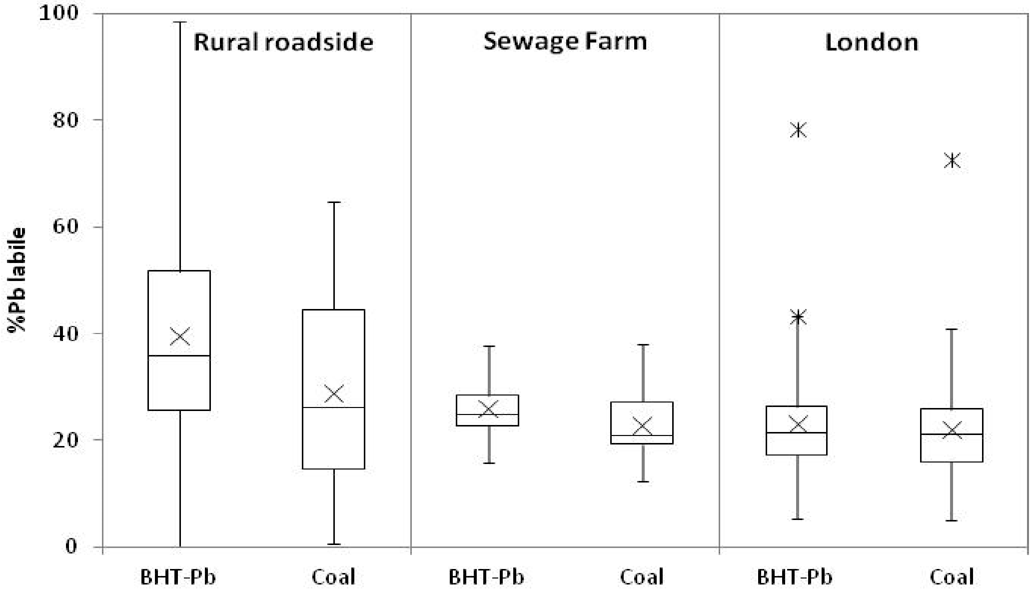


Figure 5-7: Box and whisker plot showing the estimated proportion (%) of labile Pb in soil Pb pools originating from either petrol or indigenous coal at the Rural Roadside, Sewage Farm and London sites; the higher and lower quartile represents the highest and lowest 25% of the data respectively; whiskers indicates variability outside the upper and lower quartiles; the mean value (×) and outliers are marked as an asterix.

5.3.5 Limitations to the binary model of Pb source apportionment

There are substantial limitations to the assumption that Pb in soil originates from just two major sources. First, the assumption that a clear linear mixing line between two end members indicates the presence of just two sources can be challenged. The analytical constraints of quadrupole ICP-MS, which preclude accurate determination of ^{204}Pb isotopic abundance, have encouraged a reliance on plots such as $^{208/207}$Pb against $^{206/207}$Pb (Fig. 5-1) in source apportionment studies. However, as Ellam (2010) has shown, multiple geological sources of Pb are likely to cover a limited range of values on such plots. Thus, in combination with input from the 'Broken Hill Type' lead (BHT-Pb) used in petrol, with a low $^{206/207}$Pb ratio, an apparently linear plot is likely to emerge, erroneously suggesting that there are mixtures of just two end members. The second, more obvious, problem arises from the simple assumption that BHT-Pb is synonymous with petrol-derived Pb; in fact imported BHT-Pb was widely used in industrial applications in the UK in the 20th century (Vane et al., 2011; Chenery et al., 2012). Thus Bacon et al. (1996) found that the historical trend in the $^{206/207}$Pb ratio in archived Park Grass herbage samples declined ahead of the introduction of tetra-ethyl Pb in petrol, indicating industrial inputs from BHT-Pb into atmospheric aerosols in the UK from as early as 1900. For the three datasets presented in the current study, it may be reasonable to assume that Pb in the Rural Roadside soils is predominantly a combination of petrol-derived and indigenous soil Pb. The Sewage Farm soils, historically amended with sewage from Nottingham, must have a greater input of non-petrol BHT-Pb even though Pb from urban road runoff is likely to form a substantial part of the soil Pb burden. The London soils, although also substantially affected by petrol Pb, are likely to be the most affected by non-petrol BHT-Pb. An extreme example from the London dataset is Sample 654432

(Table 5-4) with a soil Pb content of almost 23,000 mg kg^{-1} and the most pronounced BHT signature of all the soils studied ($^{206/207}$Pb ratio = 1.081 (BGS, total Pb) and 1.082 (UoN, labile Pb)). Intuitively it is highly unlikely that most of the soil Pb originated from petrol; In current study, it was estimated that this would have required the equivalent of Pb from more than 30 L petrol per kg soil. The sample site is located close to a dockland area and scrutiny of a local map from the early 20th century, shows the historical presence of a ‘White Lead Works’ ($2PbCO_3$,$Pb(OH_2)$ for paint etc) within 400 m of the site – possibly using imported Australian or Canadian lead.

Table 5-4: Soil characteristics, Pb_E and Pb isotopic ratio for Rural Roadside (a), Sewage Farm (b) and London (c) soils

(a)	Depth	Soil pH (in 0.01 M $CaCl_2$)	Total Pb	Pb_E	LOI	Avail-P	Pb IR in soil		Labile Pb IR	
Sample	(cm)		(mg kg^{-1})		%	(mg kg^{-1})	206/207	208/207	206/207	208/207
Site 1_1	0-20	6.10	47	12	8.8	4.2	1.138	2.419	1.131	2.408
Site 1_2	0-20	6.80	48	7	8.5	1.9	1.140	2.423	1.131	2.408
Site 1_3	0-20	6.73	48	8	8.8	4.4	1.144	2.424	1.133	2.411
Site 1_4	0-20	6.30	48	11	14.6	3.2	1.144	2.426	1.135	2.413
Site 1_5	20-26	6.95	39	3	5.8	3.0	1.157	2.433	1.154	2.430
Site 1_6	26-30	7.50	15	2	4.4	6.1	1.202	2.485	1.180	2.452
Site 1_7	30-40	7.46	60	0	4.2	3.9	1.166	2.443	1.178	2.445
Site 1_8	0-20	7.00	43	6	7.1	5.9	1.153	2.431	1.137	2.418
Site 1_9	20-26	7.12	63	2	5.3	2.0	1.171	2.449	1.150	2.430
Site 1_10	26-30	7.45	22	1	4.1	3.1	1.194	2.472	1.179	2.455
Site 1_11	30-40	7.57	18	0	4.1	5.2	1.192	2.468	1.170	2.444
Site 1_12	0-20	6.65	49	11	7.1	3.1	1.147	2.425	1.135	2.412
Site 1_13	20-26	6.70	41	5	5.7	2.4	1.155	2.432	1.143	2.420
Site 1_14	26-30	7.24	14	0	3.8	5.3	1.200	2.478	1.171	2.445
Site 1_15	30-40	7.43	15	0	3.6	6.4	1.198	2.477	1.179	2.462
Site 2_1	0-20	4.64	66	31	15.5	5.5	1.127	2.403	1.118	2.400
Site 2_2	0-20	4.27	66	20	13.0	12.2	1.127	2.404	1.119	2.396
Site 2_3	0-20	4.17	65	16	11.2	4.9	1.134	2.413	1.130	2.407
Site 2_4	0-20	4.13	62	18	11.4	20.8	1.130	2.409	1.123	2.399
Site 2_5	20-30	3.95	30	16	5.9	9.3	1.169	2.450	1.161	2.446
Site 2_6	30-40	3.92	25	12	4.9	2.0	1.176	2.460	1.169	2.446
Site 2_7	40-50	4.05	25	12	4.8	11.1	1.169	2.448	1.162	2.435
Site 2_8	50-60	4.25	24	12	5.2	3.8	1.172	2.456	1.162	2.434
Site 2_9	0-20	4.53	71	42	13.2	23.6	1.125	2.399	1.120	2.398
Site 2_10	20-30	4.35	161	104	7.6	11.4	1.172	2.448	1.174	2.453
Site 2_11	30-40	4.22	25	14	3.4	7.0	1.181	2.466	1.174	2.451
Site 2_12	40-50	4.19	20	8	3.0	2.5	1.188	2.478	1.183	2.463
Site 2_13	50-60	4.29	17	9	2.6	2.3	1.188	2.469	1.189	2.466
Site 2_14	0-20	5.32	109	37	16.1	9.4	1.145	2.518	1.134	2.416
Site 2_15	20-30	4.43	69	35	8.2	26.4	1.166	2.436	1.176	2.453
Site 2_16	30-40	4.76	44	18	5.1	14.3	1.179	2.457	1.160	2.444
Site 2_17	40-50	5.07	35	9	5.1	2.7	1.189	2.468	1.183	2.463
Site 2_18	50-60	5.15	20	8	4.9	5.4	1.192	2.470	1.185	2.465
Site 3_1	0-20	5.08	32	12	8.4	2.7	1.158	2.439	1.147	2.426
Site 3_2	0-20	4.74	33	14	8.5	3.9	1.158	2.441	1.147	2.423
Site 3_3	0-20	4.78	25	14	8.1	2.7	1.158	2.437	1.148	2.421
Site 4_N1	0-20	5.72	63	23	16.6	29.7	1.135	2.414	1.127	2.403
Site 4_N2	0-20	5.36	65	29	20.1	25.3	1.126	2.405	1.117	2.395
Site 4_N3	0-20	6.67	55	14	10.6	11.3	1.131	2.410	1.118	2.395
Site 4_S1	0-20	6.98	233	35	10.3	7.3	1.100	2.379	1.104	2.384
Site 4_S2	0-20	6.98	64	17	11.4	5.6	1.116	2.394	1.107	2.385
Site 4_S3	0-20	7.01	88	26	18.0	7.8	1.114	2.389	1.096	2.377

(b) **Sample**	**Soil pH** **(in 0.01 M $CaCl_2$)**	**Total Pb** **(mg kg^{-1})**	**Pb_E** **(mg kg^{-1})**	**LOI** **%**	**Avail-P** **(mg kg^{-1})**	**Pb IR in soil** **206/207**	 **208/207**	**Labile Pb IR** **206/207**	 **208/207**
6A	6.14	256	51	11.4	153.7	1.137	2.418	1.136	2.415
6B	6.57	324	88	10.8	190.4	1.130	2.411	1.133	2.416
6C	6.24	229	80	9.60	183.7	1.139	2.420	1.135	2.416
6D	6.19	282	101	11.8	141.8	1.131	2.412	1.136	2.418
7A	6.58	55	8	6.56	146.8	1.165	2.446	1.159	2.437
7B	6.74	102	19	6.38	128.5	1.157	2.435	1.151	2.431
8A	6.26	596	184	24.5	291.7	1.116	2.398	1.115	2.401
8B	5.86	631	147	23.8	379.9	1.120	2.401	1.122	2.402
8C	6.03	433	104	19.7	252.8	1.122	2.398	1.124	2.409
8D	6.08	683	145	25.5	314.9	1.119	2.395	1.117	2.397
8E	6.17	608	154	29.3	276.4	1.120	2.398	1.125	2.402
8T	6.47	712	137	25.2	182.4	1.119	2.398	1.121	2.400
9A	6.34	673	162	23.4	216.3	1.124	2.403	1.128	2.410
9B	6.53	511	124	19.5	126.1	1.124	2.403	1.123	2.411
10A	6.53	82	17	6.14	53.0	1.160	2.436	1.154	2.432
10B	6.44	68	12	5.42	69.1	1.162	2.439	1.144	2.434

(c)	**Soil pH**	**Total Pb**	**Pb$_E$**	**LOI**	**Avail-P**	**Pb IR in soil**		**Labile Pb IR**	
Sample	**(in 0.01 M CaCl$_2$)**	**(mg kg^{-1})**	**(mg kg^{-1})**	**%**	**(mg kg^{-1})**	**206/207**	**208/207**	**206/207**	**208/207**
641188	7.30	1416	272	9.3	n.a.	1.138	2.420	1.145	2.426
641198	7.14	2237	123	6.7	25.8	1.142	2.425	1.142	2.425
641201	6.93	1006	190	10.0	n.a.	1.147	2.430	1.142	2.423
641221	6.98	641	111	9.7	1.4	1.164	2.446	1.161	2.446
641228	7.07	166	9	7.1	n.a.	1.149	2.429	1.145	2.425
650221	7.07	2762	432	15.4	80.8	1.170	2.452	1.163	2.450
650266	6.88	1864	270	9.5	28.2	1.150	2.434	1.148	2.427
650324	6.98	2154	647	7.5	23.1	1.147	2.427	1.149	2.424
650373	5.56	320	65	11.5	37.2	1.152	2.432	1.143	2.422
650436	7.07	1512	345	7.3	27.5	1.128	2.408	1.136	2.425
650499	5.28	9044	6771	10.4	6.3	1.141	2.415	1.141	2.417
650763	6.74	7212	2933	6.7	19.9	1.153	2.434	1.151	2.433
650773	7.24	996	253	6.0	n.a.	1.133	2.418	1.133	2.416
650803	6.74	1828	536	9.2	12.0	1.143	2.425	1.144	2.422
650903	5.98	193	51	8.3	2.3	1.140	2.420	1.138	2.417
650930	6.90	245	43	6.8	15.6	1.129	2.406	1.125	2.408
650932	5.57	428	124	9.2	17.0	1.111	2.390	1.109	2.390
651006	6.69	213	43	7.9	n.a.	1.127	2.407	1.125	2.405
651013	6.92	2284	507	16.6	28.5	1.160	2.442	1.162	2.445
651021	6.43	1731	255	17.5	207.4	1.116	2.397	1.119	2.400
651100	6.40	534	85	14.0	n.a.	1.137	2.414	1.140	2.423
651133	5.80	217	51	11.3	22.8	1.147	2.427	1.144	2.425
651344	7.21	927	158	15.5	4.7	1.112	2.392	1.110	2.389
651438	5.00	99	29	4.3	1.6	1.164	2.446	1.160	2.433
651654	3.29	162	62.5	17.3	14.7	1.141	2.421	1.137	2.419
651871	6.74	919	311	7.3	4.1	1.134	2.414	1.130	2.407
651954	6.75	1546	180	15.5	42.4	1.146	2.428	1.151	2.432
651975	6.87	172	18	7.7	n.a.	1.125	2.406	1.120	2.401
652570	6.74	1247	269	9.7	15.6	1.133	2.415	1.136	2.417
652631	7.23	2587	668	10.0	5.9	1.157	2.440	1.157	2.440
652900	6.66	237	45	10.1	20.2	1.129	2.409	1.128	2.412
653407	6.29	301	75	10.3	n.a.	1.135	2.413	1.130	2.410
653570	6.70	6600	1450	10.9	19.0	1.162	2.445	1.161	2.441
653588	4.95	187	74	5.7	1.9	1.151	2.428	1.148	2.426
654281	6.62	1153	283	8.2	1.1	1.143	2.424	1.141	2.423
654432	6.91	22572	3409	11.5	34.9	1.081	2.359	1.082	2.360
654464	7.20	507	100	9.0	14.6	1.146	2.426	1.146	2.426
654542	7.12	845	281	6.9	8.7	1.147	2.429	1.143	2.426
654699	7.25	4056	729	16.9	6.3	1.146	2.429	1.144	2.427
654950	6.75	779	183	5.3	11.5	1.129	2.409	1.126	2.410
654976	7.05	3974	511	14.4	10.5	1.131	2.413	1.131	2.412
655376	6.76	115	23	5.5	n.a.	1.163	2.442	1.157	2.435
655522	6.78	123	14	5.0	19.8	1.137	2.419	1.136	2.417
655783	7.07	1377	323	6.4	3.8	1.115	2.394	1.113	2.393
655952	6.00	3798	1363	5.7	1.2	1.142	2.429	1.148	2.430
656000	6.44	4530	1488	12.0	37.9	1.103	2.386	1.102	2.382
656271	7.31	420	68	6.1	13.5	1.128	2.404	1.125	2.406
656359	6.61	205	51	9.9	n.a.	1.141	2.424	1.135	2.424
656463	6.03	952	188	12.4	n.a.	1.148	2.430	1.145	2.427
656571	6.90	230	57	7.8	7.3	1.116	2.394	1.116	2.399

5.4 Conclusion

In summary, both the soil properties and sources of contamination influence the isotopic exchangeability of Pb. In Rural Roadside soils, there was a relatively clear distinction between Pb from parent material and the main source of aerial contamination (petrol-derived Pb); soil pH was the most important factor in controlling Pb lability. However, in Sewage Farm soils, with a limited range of soil pH (c. pH 6.5), the co-existence of high phosphate concentration from sewage sludge may limit the magnitude and range of Pb lability seen by formation of Pb-phosphate minerals. Despite a wide range of land uses, and possible Pb sources, the Pb lability of most of the London soils fell into a restricted range (16-26%). This may be because of relatively high soil pH (mean=6.6; SD=0.75), but it is difficult to draw an unequivocal conclusion because of the complex mix of Pb sources present.

There was no consistent agreement between E-value and any single SEP fraction in all three sets of soils. However, the non-labile Pb in both Rural Roadside and London soils was likely to be a combination of Pb occlusion within Fe oxides (F3) and primary minerals (F5 of the SEP), whilst for Sewage Farm soils, the SEP residual fraction (78%) could be identified broadly with the (isotopically) non-labile pool of Pb (76%) – possibly a consequence of Pb-phosphate interactions.

There was evidence in Rural Roadside soils that petrol-derived Pb remained more labile than Pb from native ore or coal, and the proportion of petrol-derived Pb that was labile was strongly correlated with soil pH. However, both petrol-derived and native (coal) Pb contributed to both the labile and non-labile fraction by slow physical reactions such as fixation and weathering. In some of the Sewage Farm soils, the existence of high phosphate concentrations results in petrol-derived Pb having a lower lability than expected when compared with native soil Pb.

6 CHAPTER 6: LABILITY OF COPPER, ZINC AND LEAD ON HUMIC ACID

6.1 Overview

Geochemical models, such as WHAM (VI & VII) (Lofts and Tipping, 2011; Tipping, 1998) and the NIC(C)A-Donnan Model (Kinniburgh et al., 1999), are based on the hypothesis that trace metals sorbed on humic substances are entirely labile. However, there is increasing evidence that a non-labile fraction may exist in suspended colloidal phases. For example, Ma et al. (2006a) studied 18 soils and found non-isotopically exchangeable Cu in a suspended soil colloidal phase and suggested it was likely to be associated with dissolved organic matter. Similar suggestions have been made by Lombi et al. (2003) and Nolan et al. (2009) for Ni, Cu and Zn. Jackson et al. (2005) in a study of dissolved organic matter (DOM) in river water, measured Ni and U distribution in the colloidal phase and concluded that Ni was ‘more labile’ than U. By contrast Laborda et al. (2011), used asymmetrical flow field flow fractionation (AsFIFFF) to separate dispersed colloidal phases of isotopically spiked compost material and, from measurements of the isotopic ratio within different size fractions including the solution phase, they concluded that all Cu and Pb was labile.

It is accepted that metals can be held by very strong binding sites on humic substances through the formation of ‘multidentate’ complexes with carboxylic and phenolic groups or by specific binding with N and S groups which are known to preferentially bind metals such as Cr^{III}, Fe^{III}, Cu and Hg strongly (Tipping, 2002). However, strong chemical bond formation does not necessarily result in non-labile complex formation and there have been very few studies that have attempted to determine the presence of such a metal fraction on humic substances. It has been shown that both isotopic dilution (ID) approaches and single extraction with EDTA can be used to estimate the labile fraction of several trace metals in soils (Degryse et al., 2009); the labile fractions determined by these two methods are often quite closely correlated (Gabler et al., 2007). However, to measure metal lability on a dissolved system (e.g. on DOM), a fractionation technique is required to separate the macromolecular DOM from the inorganic complexes which are assumed to be labile. This approach then allows either the measurement of metals complexed to EDTA or the isotopic abundance of a spike isotope in the non-DOM solution phase. Recently, size exclusion chromatography (SEC) coupled with inductively coupled plasma mass spectrometry (ICP-MS) has been applied to the study of various trace element distributions on DOM including humic acids (Jackson et al., 2005; Kozai et al., 2013; Liu and Cai, 2010; Liu et al., 2011; Schmitt et al., 2001) and these studies have shown that SEC is an efficient way to separate dissolved humic substances based on molecular size/weight.

The aim of the investigation was to determine the lability of Cu, Zn and Pb on humic acid (HA) so that the hypothesis that ‘all trace metal in dissolved HA is labile’ could be tested. Using SEC-ICP-MS, two HAs were fractionated and the change in Cu, Zn and Pb distribution between HA and inorganic complexes characterized in the presence of EDTA and added stable isotopes. An EDTA extraction was undertaken after incubation of HA with a metal spike enriched with ^{65}Cu, ^{70}Zn and ^{204}Pb to determine the extractable metal fractions. In addition, the isotopically exchangeable Cu, Zn and Pb fractions (E-values) were also measured on dissolved humic acids ($M_{E\text{-}HA}$) incubated with Cu, Zn and Pb (normal isotopic mix) for a range of time periods. This was achieved using ^{65}Cu, ^{70}Zn and ^{204}Pb as the spike isotope and measuring isotopic ratios for Cu, Zn and Pb in the humic acid phase and as inorganic complexes using SEC-ICP-MS for phase separation.

6.2 Materials and methods

6.2.1 Humic acid preparation

Humic acid was previously extracted from a grassland soil (G-HA) (Nottinghamshire, UK) and an acid peatland soil (P-HA) (North Yorkshire, UK) freeze dried and preserved in glass containers in the dark (Marshall et al., 1995). The ash content (g kg^{-1}) was measured by dry combustion at 500°C for 8 hours. A modified $Ba(OH)_2$ method (Schnitzer and Khan, 1972) was used to determine the total acid group concentration (acidity in mol_c kg^{-1}) including a step of pre-dissolution of humic acid in 0.1 M NaOH for complete hydration (Marshall et al., 1995). Stock humic acid solutions (16 g L^{-1}) were prepared by dissolving in 0.5 M NaOH before adjusting to pH 7.0 with nitric acid. Total *residual* Cu, Zn and Pb concentrations (before incubation/spiking) were measured on dilutions of this stock in a matrix of 0.005 M EDTA using ICP-MS. Detail of the instrument settings and processing of data are described in Chapter 2, Section 2.2.6. These general chemical properties are listed in Table 6-1.

Table 6-1: Origins and selected chemical properties of humic acid samples

	Origin	**Ash content***	**Acidity***	**Cu**	**Zn**	**Pb**
		g kg^{-1}	**mol_c kg^{-1}**	**mg kg^{-1}**	**mg kg^{-1}**	**mg kg^{-1}**
G-HA	Permanent grassland soil	23.5	5.50	151	168	27.7
P-HA	*Sphagnum* moss peat	6.70	7.00	81.6	97.4	27.1

*Ash content and acidity were measured by Marshall et al. (1995).

6.2.2 Determination of EDTA-extractable Cu, Zn and Pb

Humic acid dissolved in NaOH, at concentrations of 4.0 g L^{-1} (G-HA) or 4.4 g L^{-1} (P-HA) , were incubated with a mixed stable isotope enriched solution: 168 µg L^{-1} ^{65}Cu (≥ 99.0%), 30.8 µg L^{-1} ^{70}Zn (≥ 92.7%) and 17.7 µg L^{-1} of ^{204}Pb (≥ 99.4%). Metal concentrations were chosen to be high enough to give a measurable ICP-MS signal after complex-formation without resulting in significant competition for functional groups. The concentration of functional groups can be calculated from acidity (Table 6-1), as 0.022 mol_c L^{-1} in this incubated solution for G-HA and 0.031 mol_c L^{-1} for P-HA, while the mixed stable isotope solution contained 6.2 µmolc L^{-1} of metals. After equilibration for 40 or 160 days at 18°C the spiked solutions were diluted with 0.1 M Tris buffer at pH 8.2 (to give final concentrations of 1.0 g L^{-1} for G-HA and 1.1 g L^{-1} for P-HA) and equilibrated for 2 hours with a range of Na-EDTA concentrations (0, 0.0005, 0.0025, 0.01, 0.025 and 0.05 M). Samples were analysed by size exclusion chromatography ICP-MS (SEC-ICP-MS, see Section 6.2.4) to determine metal-EDTA, metal-Tris and metal-HA complexes.

6.2.3 Isotopically exchangeable Cu, Zn and Pb

Isotopic exchange kinetics and E-values for Cu, Zn and Pb ($M_{E\text{-}HA}$) were determined on G-HA (4.0 g L^{-1}) and P-HA (4.4 g L^{-1}) incubated for 40 days with a mixed-metal spike prepared from Cu and Zn ICP-MS standard solutions (with natural isotopic abundance; at concentrations of 240 µg L^{-1} and 320 µg L^{-1} respectively) and a Pb NIST SRM (981) standard (52.5 µg L^{-1}) to give a substantial increase in the total metal concentration in HAs. The samples were then further spiked with a stable isotope solution to give a significant increase in ICP-MS signal. Final concentrations of ^{65}Cu, ^{70}Zn and ^{204}Pb were 118.0 µg L^{-1}, 61.5 µg L^{-1} and 35.5 µg L^{-1} respectively.

Immediately following addition of the isotopic spike, samples were diluted with Tris buffer (to 1.0 g L^{-1} for G-HA and 1.1 g L^{-1} for P-HA) and analysed using SEC-ICP-MS following isotopic equilibration times of between 35 min and 3 days.

6.2.4 SEC ICP-MS Analysis

The settings for SEC are described in Chapter 2, Section 2.2.6.2. Samples of 100 µl HA solution were injected into an eluent stream (0.1 M of Tris buffer, pH 8.2) with a flow rate of 1 ml min^{-1} into the ICP-MS for isotope analysis. Eight isotopes ^{63}Cu, ^{65}Cu, ^{66}Zn, ^{70}Zn, ^{204}Pb, ^{206}Pb, ^{207}Pb and ^{208}Pb were measured with a single dwell time of 40 mS. The remaining ICP-MS settings are described in Section 2.3.1. A correction factor (K-factor) was applied to correct for mass discrimination to each measured isotopic ratio as described in Section 2.3.2.2.

Trace metal calibration in SEC-ICP-MS can present problems because it is unlikely that inorganic standards would survive transition of the SEC column given the pH of the mobile phase. A solution to this is to use HA as a carrier for the metal standards. Calibration standards were therefore prepared by adding known concentrations of Cu, Zn (ICP-MS standard solutions) and Pb (NIST SRM (981)) to a 2 g L^{-1} solution of G-HA. The entire chromatogram for the most abundant isotopes (^{63}Cu, ^{66}Zn, and ^{208}Pb) was integrated using PlasmaLab software (version 2.5.1.276, Thermo Electron, 2003). An integrated blank chromatogram (2 g L^{-1} G-HA without added metal) was subtracted from equivalent metal-amended samples to generate calibration curves for Cu, Zn and Pb as counts per second (CPS) versus concentration (with $R^2 \geq 0.99$ for Cu and Pb, $R^2 \geq 0.92$ for Zn). Total Cu, Zn and Pb concentrations in metal-incubated humic acids were calculated using these ‘standard addition’ calibration curves.

The isotopic ratio of Tris-complexed Cu, Zn and Pb used to calculate $M_{E\text{-}HA}$ was adapted from Eq. 2-2, and is shown in Eq. 6-1 for Cu in which ^{65}Cu is the spike isotope and ^{63}Cu is the native isotope measured:

$$M_{E\text{-}HA}=\left(\frac{M_{HA}}{W}\right)\left(\frac{C_{spk}V_{spk}}{M_{spk}}\right)\left(\frac{^{65}IA_{spk}-{}^{63}IA_{spk}R_{ss}}{^{63}IA_{HA}R_{ss}-{}^{65}IA_{HA}}\right) \qquad (6\text{-}1)$$

Where M_{HA} and M_{spk} are the average atomic masses of Cu, Zn and Pb in the (unspiked) HA solution and the spike solution respectively, W is the weight of humic acid (kg), C_{spk} is the gravimetric concentration of Cu in the spike solution, V_{spk} is the volume of spike added (L), IA is the molar isotopic abundance of ^{65}Cu or ^{63}Cu in the spike solution or HA (unspiked), and R_{ss} is the isotopic ratio of ^{65}Cu to ^{63}Cu in the spiked HA solution measured in the separated non-humic phase of the chromatogram; the latter phase is equivalent to the separated supernatant solution in a soil suspension used for measurement of a soil E-value.

6.2.5 Modelling Cu, Zn and Pb isotope distribution on humic acid:

The extent of movement of spike isotope from the non-HA phase (mainly Tris complexes) into HA-complexed forms as a function of contact time was modelled using an adapted exponential equation originally developed to describe the degree of mixing of radioactive isotopes with total soil metal pools (Sinaj et al., 1999; Young et al., 2007), Eq. 6-2:

$$P_{(t)}=P_{(1)}\left[t+P_{(1)}{}^{1/n}\right]^{-n}+P_{(M)} \qquad (6\text{-}2)$$

Where P is the proportion of spiked ^{65}Cu, ^{70}Zn or ^{204}Pb in the solution phase (Tris complexes) at a specified time (t) or after 1 min (1), and n is an empirical constant. The proportion of humic acid metal (all isotopes) in solution $P_{(M)}$ is the value of $P_{(t)}$ at infinite time and assumes that the measured metal concentration in solution reflects an equilibrium state. The constants of $P_{(1)}$, $P_{(M)}$ and n were optimized by minimizing the residual standard deviation (RSD) between the measured and modelled $P_{(t)}$ using the *Solver* function in *Microsoft Excel*.

6.3 Results and discussion

6.3.1 General characterization of Cu, Zn and Pb on dissolved humic acid

Chromatograms showing both HAs incubated with Cu, Zn and Pb for 40 days, eluted in a Tris buffer, are shown in Fig. 6-1. The distribution of Cu, Zn and Pb between HA and Tris complexes is reasonably clear, with larger organic species having a shorter residence time than smaller inorganic ones, and differences in the behaviour of the three metals are also apparent. All three metals were detected in both the excluded volume (eluted at ~ 7 min) and in later eluting fractions. The first sharp peak represented the excluded fraction which was the HA-metal fraction larger than 300,000 Da. After this, a broad peak (7.5 - 13 min) was seen in all chromatograms presumably reflecting the distribution of molecular masses of the HA but also probably arising from degrees of HA interaction with the column matrix, either with hydrophobic aromatic functional groups (Asakawa et al., 2011) or charged surfaces, leading to changes in retention time (Chin and Gschwend, 1991; Jackson et al., 2005; Pelekani et al., 1999). Comparatively, there was a higher proportion of Zn-Tris complexation (peak at c. 14-15 min) than Cu and Pb implying relatively weaker binding of Zn to HA.

All Zn and Pb complexes were eluted within 15 min whereas Cu continued to elute for up to 20 min, probably a result of Cu-Tris complex formation during elution, as the two NH_2 group in Tris form a relatively stable complex with Cu. The effect of Tris on Cu elution is influenced by pH. McPhail and Goodman (1984) showed that decreasing the pH from 10.4 to 8.0 diminished a substantial proportion (~ 40%) of Cu-Tris signal. Therefore, it is reasonable to assume that at pH 8.2 Tris buffer is able to complex a substantial amount of Cu, with a relatively small effect on HA-Cu complexes.

A difference in metal distribution between humic acids was also clear (Fig. 6-1). Lower metal concentrations were observed in the excluded volume for P-HA, and the average size of the second peak range was smaller than that for G-HA for Cu and Zn. Although the total number of acid functional groups is higher in P-HA than in G-HA (Table 6-1), a larger proportion of Zn/Pb-Tris complexes in P-HA solutions implies weaker binding of Zn and Pb by P-HA than by G-HA. This may result from differences in factors relating to functional groups such as binding site density which can affect the ability of metals to form strong multi-dentate bonds (Lee et al., 1993).

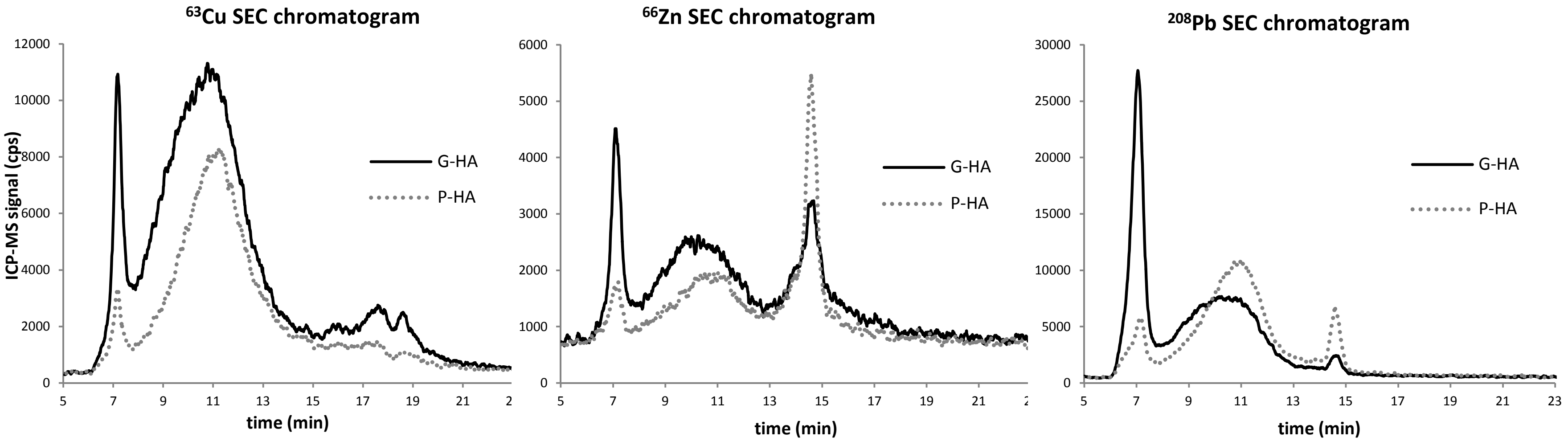


Figure 6-1: SEC-ICP-MS chromatograms of ^{63}Cu, ^{66}Zn and ^{208}Pb showing complexation with grassland soil humic acid (1.0 g L^{-1}) and peat humic acid (1.1 g L^{-1}). Both humic acids were incubated with Cu, Zn (natural isotopic abundance) and Pb (NIST SRM (981) standard) for 40 days. For the grassland soil humic acid, the final Cu, Zn and Pb concentrations were 151 µg L^{-1}, 168 µg L^{-1} and 27.7 µg L^{-1}; for the peat humic acid the final Cu, Zn and Pb concentrations were 89.7 µg L^{-1}, 107.2 µg L^{-1}, and 29.82 µg L^{-1} respectively.

Metal recovery was calculated and showed close to 100% Cu recovery for both G-HA (103%) and P-HA (94%). There was a c. 10% loss of Zn (87% and 90% recovery for G-HA and P-HA respectively) and 30-40% loss of Pb (64% and 69% recovery for G-HA and P-HA). This may be a consequence of Zn or Pb dissociation from HA during elution or a result of experimental error as the ICP-MS signal was low for both Zn and Pb. However, as the results presented here mainly rely on isotopic ratio and relative proportions of metal in HA and Tris complexes, losses during elution should have a minimal effect on the results. Relatively low Pb recovery was also reported by Schmitt et al. (2001) in a study using SEC fractionation of organic matter in lake water which indicated the presence of labile Pb-organic matter complexes, and hence it appears that Pb can strongly interact with the column material and consequently have a low recovery.

6.3.2 Integration of Cu chromatograms

As shown in Fig. 6-1, the distribution of ^{63}Cu and ^{65}Cu on humic acid forms a continuous chromatogram with a poorer distinction between HA-bonded and non-HA forms, compared to the chromatograms for Zn and Pb spiked HA solutions. To calculate the value of $M_{E\text{-}HA}$ for Cu on HA it was necessary to integrate the ^{65}Cu and ^{63}Cu present as a Tris complex; from this a ^{65}Cu/^{63}Cu isotopic ratio could then be calculated. To avoid using an arbitrary division between HA-bound Cu and Tris-bonded Cu a test was carried out to establish the effect of choosing to integrate across different time points within the chromatogram.

The G-HA solution was spiked with a ^{65}Cu enriched solution and equilibrated for 72 hours before analysis (Fig. 6-2). Ten points were selected on the chromatogram between 5 and 23.3 min. The final point at 23.3 min was where the whole sample was assumed to have been eluted through the SEC column. The chromatogram for ^{63}Cu was integrated in the same way at the same time points. Integration was undertaken in both directions along the time axis i.e. from high molecular weight (MW) to low MW (5 min to 23.3 min) and from low MW to high MW (23.3 min to 5 min). Due to the presence of a non-labile Cu fraction in HA (explained in later Sections) the isotopic ratio of ^{65}Cu/^{63}Cu in the Tris complexes (labile fraction) was higher than that in the HA (Fig. 6-2). The two time series converged at a ^{65}Cu/^{63}Cu ratio of 2.181 when the whole chromatogram was integrated at time t = 5 or t = 23 min. A clear change in the ^{65}Cu/^{63}Cu ratio was observed where contributions to the ratio resulted from both HA-bound and Tris-bound species. When integrating from the high MW end (the start) of the chromatogram isotopic ratios before 13.3 min were not affected by the presence of (^{65}Cu-enriched) Cu-Tris complexes. Similarly Tris-complex Cu species eluted after 16.3 min were unaffected by the HA complexes containing a lower ^{65}Cu/^{63}Cu isotopic ratio (Fig. 6-2). Therefore, only isotopic ratios calculated after 16.3 min were used in the calculation of the $M_{E\text{-}HA}$ for Cu in HA.

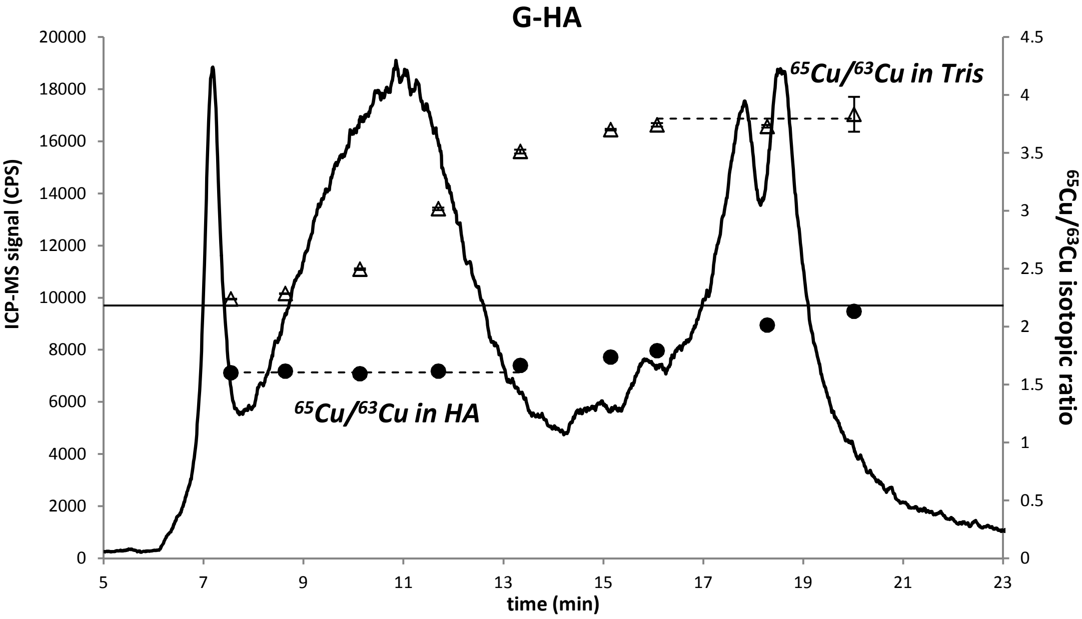


Figure 6-2: A ^{65}Cu chromatogram of G-HA solution 72 hours after spiking with ^{65}Cu (broken line). The isotopic ratio ^{65}Cu/^{63}Cu measured using different integration times are shown where chromatograms were progressively integrated from left (solely HA) to right (●), or from the right hand side (solely Tris-Cu complexes) to left (Δ). The dash lines represent the final isotopic ratios for the HA-Cu and Tris-Cu complexes. Error bars are the standard error of duplicates.

6.3.3 EDTA extraction of Cu, Zn and Pb on humic acid

Concentrations of total oxyacid groups on the HAs were 5.5×10^{-3} M and 7.7×10^{-3} M for the G-HA and P-HA solutions respectively. Concentrations of EDTA carboxyl groups in the extracting solutions used (Section 6.2.2), ranged from 2×10^{-3} M (in 0.0005 M EDTA) to 0.2 M (in 0.05 M EDTA). The highest concentrations of EDTA therefore should have been able to complex all labile Cu, Zn and Pb in preference to the HA whereas at the two lowest EDTA concentrations HAs may have been strong competitors for metal bound to EDTA. Figure 6-3 shows ^{63}Cu and ^{65}Cu chromatographs for P-HA with 0.0005 M and 0.05 M EDTA. It should be noted that the ^{63}Cu arises solely from residual Cu in the 'purified' HA which has resisted acid elution and dialysis during the preparation of the HA; the ^{65}Cu was equilibrated with the HA for 40 days prior to extraction for 2 h with EDTA (Section 6.2.2). A decrease in ICP signal is observed for both isotopes bound to HA with an increase in the metal bound EDTA peak, as the EDTA concentration was increased. However, even though it might be expected that 0.05 M EDTA would be able to complex all Cu in the system, a significant proportion of Cu (> 40%), for both residual (eg ^{63}Cu) and spiked (^{65}Cu) isotopes, remained bonded to both HAs.

Single extraction with 0.05 M EDTA has been widely used for estimating labile trace metals in soils using an extraction time of just 2 hours (Marzouk et al., 2013). Therefore, it is reasonable to classify the remaining Cu fraction on HAs as non-labile. The peak representing Cu-Tris complexes diminished, as Cu-EDTA was a stronger complex and easily out-competed Tris. It should also be noted that the EDTA peak shifted to a longer elution time at higher EDTA concentration for reasons that are not clear but may indicate changes in speciation during elution within the SEC column.

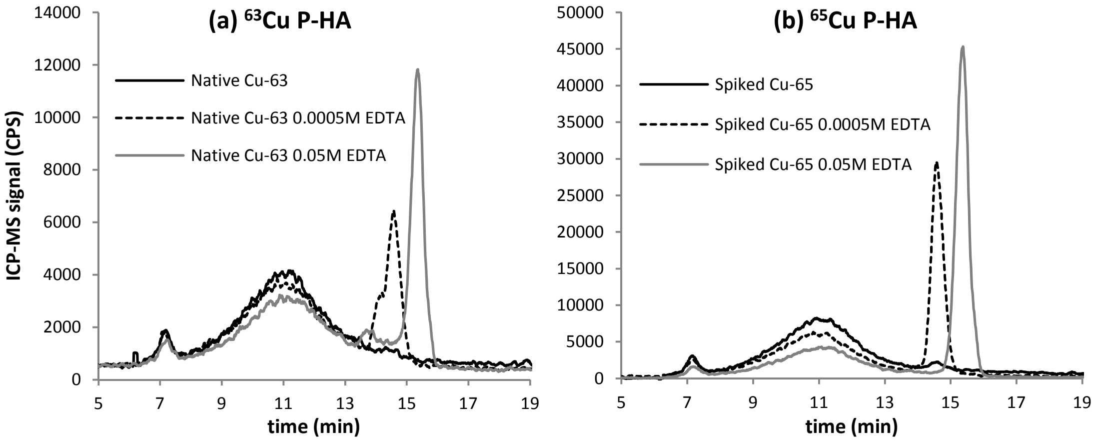


Figure 6-3: SEC-ICP-MS chromatograms of residual ^{63}Cu (a) and spiked ^{65}Cu (b) in peat humic acid (P-HA; 1.1 g L^{-1}) solutions at two EDTA concentrations. The humic acid was incubated with ^{65}Cu for 40 days before EDTA was added at concentrations of 0.0, 0.0005 and 0.05 M (Section 6.2.2).

For all three metals, %EDTA extractable metal increased with EDTA concentration (Fig. 6-4), most obviously at EDTA concentrations > 0.01 M (0.04 M total oxyacid groups). This may suggests that EDTA functional groups at lower EDTA concentrations were close to saturation with available cations present in the sample either from Tris or HA (i.e. not just Cu, Zn and Pb). A significant change in %EDTA extractable metal with HA-metal equilibration time (40 or 160 days) was observed while residual metal (residual contamination within the 'purified' HA) was least extractable. This indicates that fixation of isotopes occurred during incubation (40 and 160 days) and that the residual metal was held in an even more stable form. This ageing effect for metals bound to humic substances has also been reported in other studies (Burba, 1994; Rate et al., 1993) and two hypotheses have been proposed to explain the phenomenon. Choppin and Clark (1991) suggested that there was a movement of metals from weak binding sites to strong sites when the macromolecular structure of HA opens as a result of charge repulsions between ionized acidic functional groups enabling the aqueous solute to access otherwise hydrophobic domains within the DOM (McCarthy et al., 1989). An alternative theory, known as the 'thermodynamic' hypothesis (Cabaniss, 1990; Rate et al., 1993) is that it simply takes time for metals such as Cu, Zn and Pb to migrate to the strongest binding sites while being effectively impeded by bonding to weaker sites.

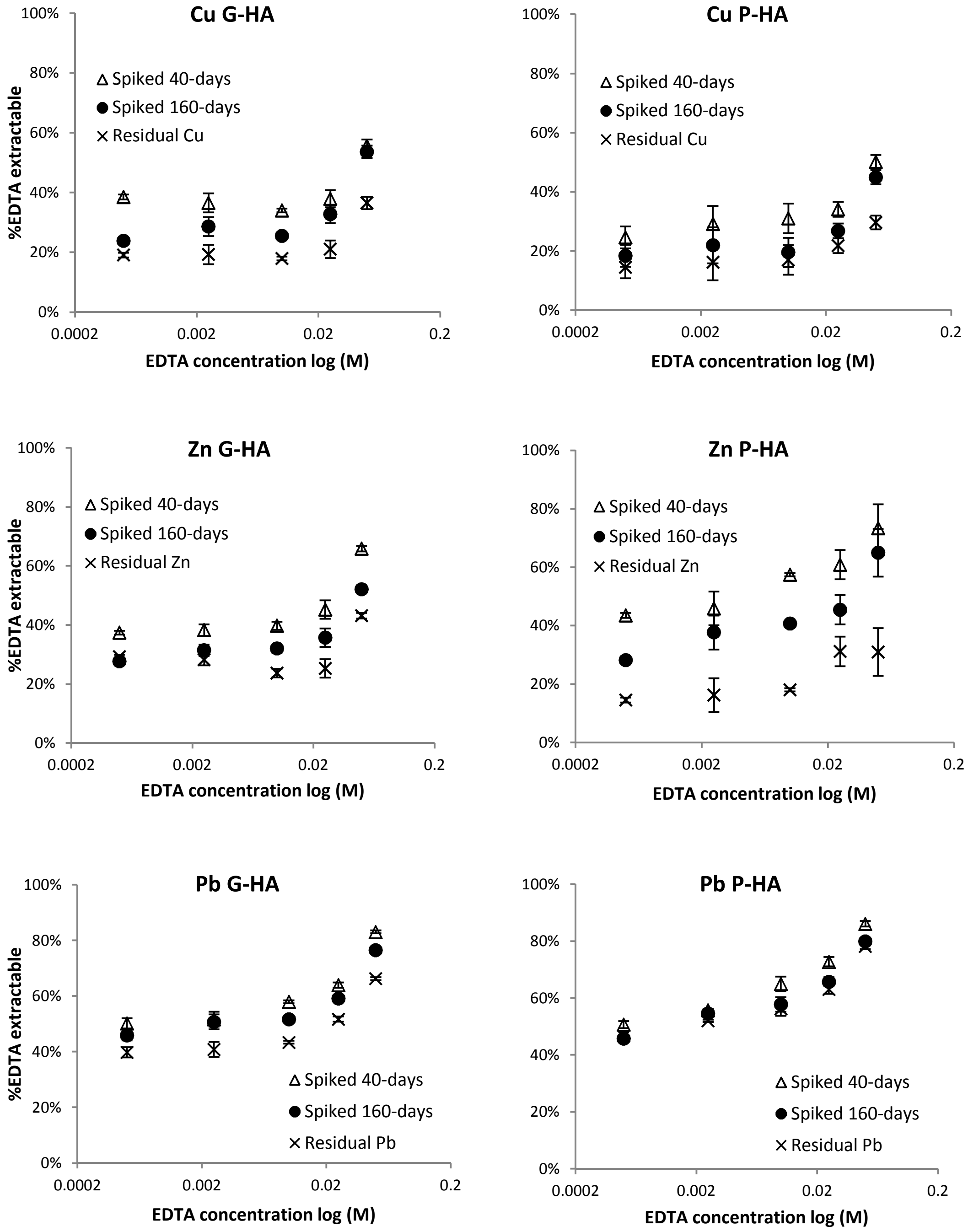


Figure 6-4: Proportion of Cu, Zn and Pb, both residual (×) and spiked incubated for 40 days (Δ) or 160 days (●), extracted by EDTA from G-HA and P-HA humic acids as a function of EDTA concentration (M; $\log_{10}$ scale). Error bars are the estimated standard error between duplicates from the native isotopes, i.e. ^{63}Cu, ^{66}Zn and ^{208}Pb.

Of the three metals, Cu had the lowest %EDTA extractable with only 60% extracted in 0.05 M EDTA, confirming that Cu is more strongly bound to humic substances than Pb or Zn (Burba, 1994). Although it has been suggested that Cu binding to soil HAs is stronger than to peat HAs (Sahu and Banerjee, 1990), the results in Fig. 6-4 suggest the reverse. However this may be a consequence of the relatively low concentrations of residual (before incubation/spiking) metal concentration in P-HA than that in G-HA (Table 6-1), resulting in a higher proportion being bound to (less abundant) strong binding sites. Comparison of the behaviour of extractable Zn with G-HA and P-HA could not be established due to the low abundance of ^{70}Zn, however, the difference between extractable native (^{66}Zn) and spiked Zn was more pronounced in P-HA than in G-HA suggesting that Zn in P-HA was more labile. The %EDTA-extractable Pb was similar in both HAs. Although it is generally accepted from modelling practices that Pb is strongly bound with particulate organic matter in soil (Gustafsson et al., 2011; Guo et al., 2006), its lability in dissolved HA is relatively low, which may be result of its larger radius of cation being difficult to form inner sphere HA complexes.

6.3.4 Isotopically exchangeable Cu, Zn and Pb on humic acid

The distribution of ^{63}Cu (post-incubation) and spiked ^{65}Cu distributions are shown in Fig. 6-5a for metal-incubated G-HA and P-HA after spiking and equilibration for 3 and 2 days respectively. On the HA fraction for both G-HA and P-HA, the distribution of the spiked ^{65}Cu follows that of the ^{63}Cu, which was incubated as ICP-MS standard Cu for 40 days before spiked with ^{65}Cu enriched solution (Section 6.3.1), implying a relatively open HA structure on the basis of this time scale of measurement. Closer investigation by comparing the isotopic ratio on three HA fractions that were free from the influence of Tris complexes (6 - 7.5 min; 7.5 - 8.6 min and 8.6 - 10.1

min) (see Section 6.3.2) confirmed there to be no significant difference between the three fractions (T-test, $P > 0.1$). Other authors have also found little difference in the binding of Zn and Cd by different fractions of HAs and suggested that the forward reactions were almost complete within a few minutes (Ma et al., 1999). Comparison of the distribution of incubated isotopes in non-spiked samples to the spiked samples also shows no change in the SEC chromatogram. This suggests that, after spiking, no conformational changes occur in the humic structure (such as a more compact structure due to the formation of multidentate complexes or reduction of repulsive interactions).

The modelled proportion of spiked ^{65}Cu in Tris complexes agrees well with the measured proportion for both HAs (Fig. 6-5b; Eq. 6-2). Both showed a rapid dissociation of the spike from Tris complexes and subsequent sorption by HAs. The optimized constants and values of RSD are shown in Table 6-2. As a result of large analytical error for measuring ^{70}Zn on G-HA, Eq. 6-2 failed to predict its proportion on HA by consistently predicting 50% of ^{70}Zn on G-HA. Model prediction for the proportion of ^{70}Zn in P-HA and ^{204}Pb in both HAs however, does show variation as a function of contact time. The optimized constant ‘n’ demonstrates that for both Cu and Pb, the isotopic exchange reaction was more rapid in G-HA than that in P-HA. The values of $P_{(M)}$ suggested that, after 2-3 days of contact, the system was close to equilibrium. Comparing all three metals indicated that there was a greater proportion of Cu-Tris complexes at equilibrium, which may be a consequence of the strength of Cu-Tris complexes compared to those formed with Pb and Zn. The stability constants for Tris complexes of Cu^{2+}, Zn^{2+} and Pb^{2+} at 25°C measured in an ionic strength of 0.1 M are 4.05 ± 0.02, 1.94 ± 0.03 and ≤ 2.7, respectively (Fischer et al., 1979).

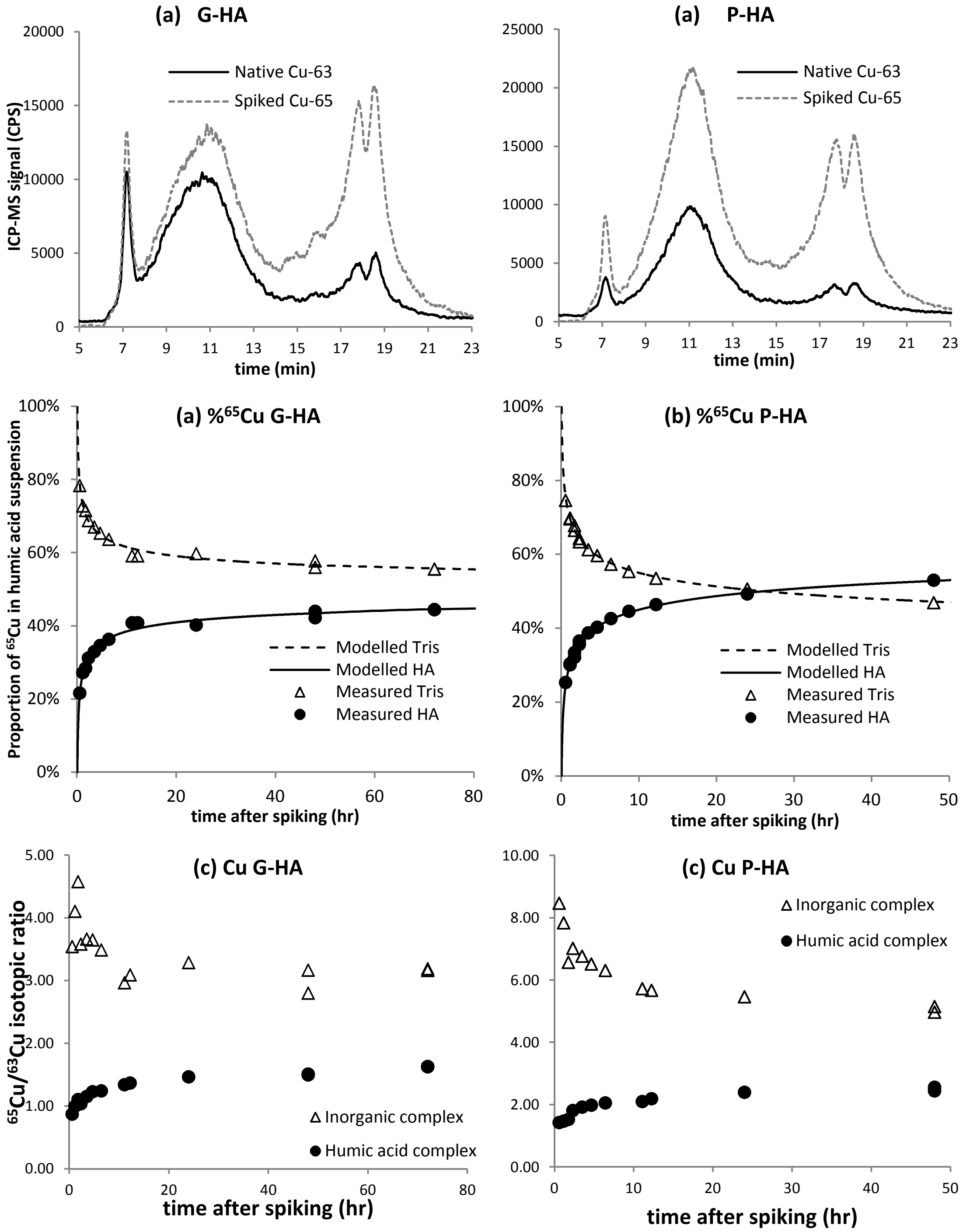


Figure 6-5: **(a)** SEC-ICP-MS chromatograms of spiked ^{65}Cu and incubated ^{63}Cu on grassland soil humic acid (72 hours after spiking) and for peat humic acid (48 hours after spiking). The humic acids were previously incubated with mixed metal standards for 40 days. **(b)** Proportions of ^{65}Cu-humic acid complexes in whole ^{65}Cu chromatogram as a function of time after spiking. **(c)** Isotopic ratio of ^{65}Cu/^{63}Cu on humic acid complexes (●) and Tris complexes (Δ) as a function of time after spiking.

Table 6-2: Values of the parameterized model constants and RSD

		G-HA			P-HA	
	Cu	**Zn**	**Pb**	**Cu**	**Zn**	**Pb**
$P_{(1)}$	0.86	*1.00*	1.00	0.99	1.00	1.00
$P_{(M)}$	0.48	*0.50*	0.18	0.21	0.39	0.00
n	0.29	*16.96*	0.54	0.17	0.17	0.20
RSD	0.009	*0.090*	0.037	0.005	0.043	0.088

The change in Cu isotopic ratio of Tris complexes and HAs as a function of time after spiking are shown in Fig. 6-5c. The difference between the ratio in Tris complexes and that in the HA phase of the chromatogram implies the existence of non-labile Cu in humic acid. Isotopic ratios for Tris-complexed metal were used to calculate $M_{E\text{-}HA}$ according to Eq. 6-1. Lability (%$M_{E\text{-}HA}$) was then calculated by dividing $M_{E\text{-}HA}$ (mg kg^{-1}) by total metal concentration measured after SEC elution (Fig. 6-6). Strong evidence of non-labile Cu in both G-HA and P-HA was observed and values of % $M_{E\text{-}HA}$ after 2-3 days (48-72 hours) were very close to the %EDTA extractable measured in 0.05 M EDTA extracts of the HAs spiked with ^{65}Cu and incubated for 40 days (Fig. 6-4). The lability of Cu in G-HA was slightly greater than that in P-HA. The measured %$M_{E\text{-}HA}$ increased with time probably as a result of the spiked isotopes accessing less accessible isotopic exchangeable binding sites within the HA structure. A stable value of %$M_{E\text{-}HA}$ was only obtained for Cu >24 hr after spiking (Fig. 6-6). Therefore, to avoid fixation of isotopes on HAs whilst allowing sufficient time for metal to access all isotopically exchangeable binding sites it is prudent to use an equilibrium time of 24 to 72 hrs for measuring such $M_{E\text{-}HA}$.

Despite substantial experimental error in measuring $M_{E\text{-}HA}$ for Zn, results suggest the existence of non-labile Zn in P-HA (<60%). Non-labile Zn was likely to exist on G-HA, as the lability of Zn was ~72.5% after 72 hours (3 days) equilibration. However, as previously mentioned, the large analytical errors make it impossible to determine the exact lability of Zn on G-HA.

Lead on G-HA showed a lability of ~50-60% which was less than the proportion of 0.05 M EDTA extractable Pb (79.8%). As Pb is weakly bound to HAs, it is reasonable to conjecture that non-labile Pb on G-HA was mobilized by high concentration of EDTA. For P-HA however, Pb was almost 100% labile after being equilibrated for 48 hours measured using the ID method, while the EDTA extractable was 86.1% for the ^{204}Pb after 40 days incubation. A potential explanation is the relatively shorter equilibrium time (2 hours) in the EDTA extraction not being sufficient to access all the labile Pb on the HAs. It was not possible to confidently establish % $M_{E\text{-}HA}$ values for Pb on the HAs, because of the relatively low recovery gained for Pb (64% and 69% for G-HA and P-HA respectively). It is perhaps reasonable to assume that the spiked ^{204}Pb may be lost more easily than post-incubated HA-Pb during elution through the SEC column, therefore the low apparent ^{204}Pb abundance measured in the Tris phase will result in an inflated E-value for Pb held on humic acid.

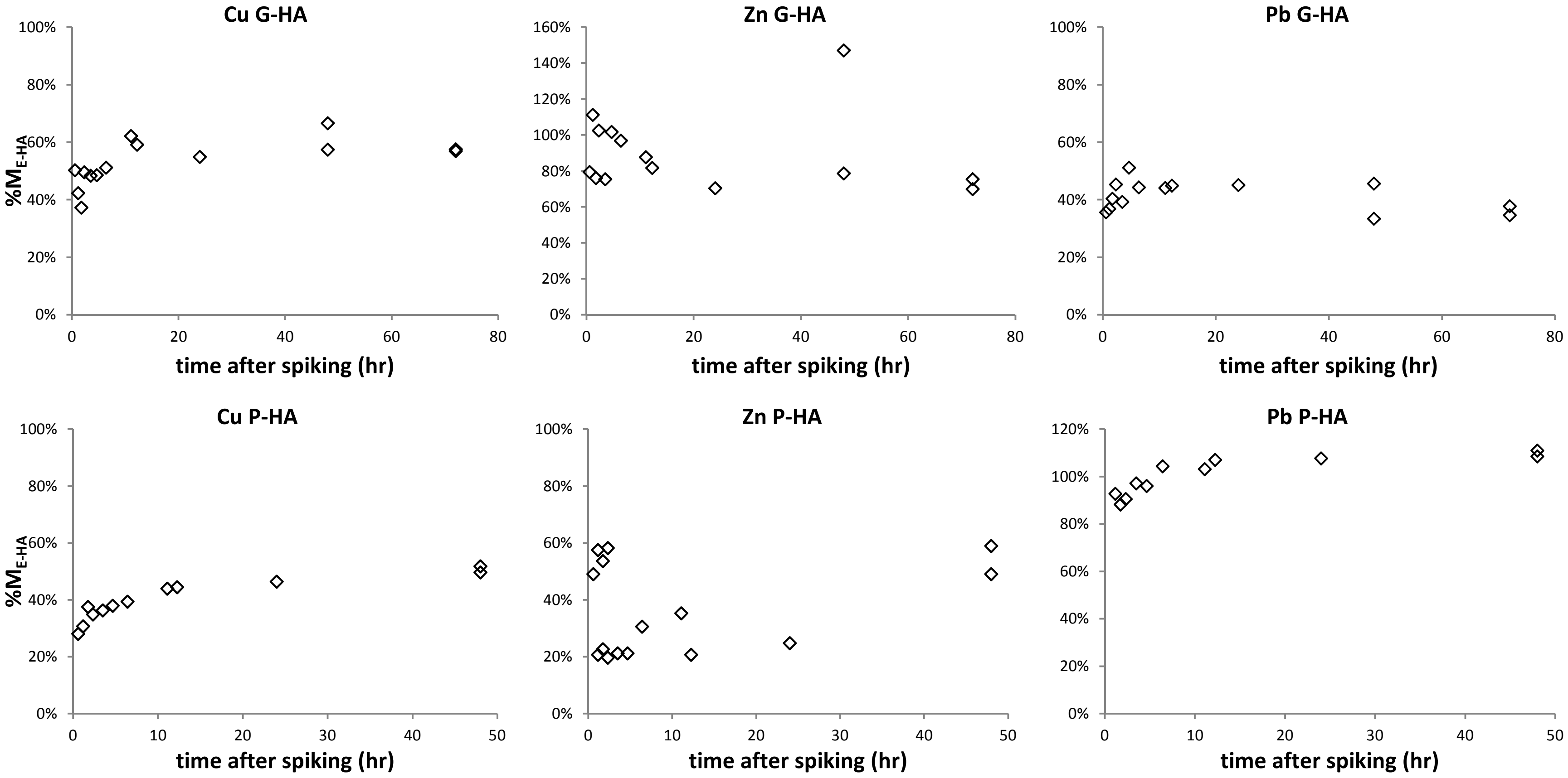


Figure 6-6: %E-values (%$M_{E\text{-}HA}$) for Cu, Zn and Pb on grassland soil (G-HA) and peat (P-HA) humic acids as a function of spike isotope equilibration time.

6.4 Conclusions

Non-labile Cu, Zn and Pb fraction has been observed on humic acids extracted from grassland and peat soils, by using SEC in conjunction with EDTA extraction and isotopic dilution. Evidence of metal fixation on HA during 40 and 160 days incubation was observed for all three metals using EDTA extraction. Results indicated that the proportion of non-labile Cu on HAs can be 40-50%. The findings suggest that the assumption made in geochemical speciation models that all metals bound to dissolved humic acid are labile, needs further verification and testing. It would also be interesting to extend the approach used here to including fulvic acid which is believed to be more active in increasing trace metal mobility. However, this technique may be limited by the potential interaction of weakly bounded metal (in this case, Zn and Pb) with the gel matrix of the column.

7 CHAPTER 7: CONCLUSIONS AND FUTURE WORK

7.1 Overview

The work presented in this thesis used multi-element stable isotope dilution (ID) to measure a discrete reactive metal phase in soil, thus providing a unique tool to study the effect of contact time, soil properties and contaminant source on Ni, Cu, Zn, Cd and Pb lability. Lability was investigated on soils incubated for an extended period with metal salts and a logistic model established to describe the relationships observed (Chapter 3). Metal lability in Urban soils with historical contamination was compared with model predictions and the deviations observed interpreted from the properties of the original metal contaminant (Chapters 3 & 5). Metal solubility and fractionation were predicted from an extended Freundlich equation, and the mechanistic model WHAM-VII (Tipping et al., 2011), using isotopically exchangeable metal as a determining variable (Chapter 4). Evidence for extended Pb lability in soils contaminated by petrol combustion was assessed by combining measurements of lability from isotopic dilution and source apportionment from Pb isotopic ratios (Chpater 5). A unique means of following the kinetics of metal binding to dissolved humic acid was also developed to test the long-standing concept that metal bonded to soil organic matter is wholly labile (Chapter 6).

7.2 Main findings

7.2.1 Operational aspects of the isotopic dilution technique: pros and cons

Recent analytical improvements in the measurement of stable and radiogenic isotopes at low concentrations, using techniques such as ICP-MS, has meant that the facilities required for isotopic dilution are available in many laboratories. Potentially the method can be utilised for any element that has more than one stable isotope or long-lived radioactive isotopes (e.g. ^{129}I, Shetaya et al., 2012).

As shown in Chapter 3 ID is less likely to overestimate metal lability compared to chemical extraction approaches where, for example, acid may mobilize non-labile metals bound with mineral (hydr)oxides and carbonate surfaces. However overestimation of E-value can occur if non-labile SCP-metal complexes are present, as demonstrated here using an additional resin purification step. Previous studies suggested that the non-labile SCP-metal complexes were most likely to be associated with DOM (Ma et al., 2006a; Lombi et al., 2003; Marzouk 2012). In this study, this presence of non-labile SCP-metal complexes was more obvious in high pH soils with low metal lability, but in most cases produced an overestimation of %E-value of less than 2% for both metal salt-amended and urban soils. Isolation of the true isotopic abundance in the labile metal fraction using a chelating resin may increase experimental error, both as a consequence of resin contamination and because the metal content extracted by resin may be too small to be accurately measured. There is also a risk of resin extracting the non-labile fraction. Therefore, when measuring E-values for soils with relatively low SOC content, the resin purification step may not be necessary.

The choice of isotopic spike concentration is also important. Inadequate spike concentration may result in small changes in isotopic ratio which are not quantifiable by ICP-MS; whereas high spike concentrations will increase the experimental cost and may increase the likelihood of acid dissolution of non-labile soil metal. Other workers (Gabler et al., 2007) have used chemical extraction of labile soil metal to estimate the level of isotopic spike required. This work (Chapter 3) demonstrated that a spike level that changes the isotopic abundance by 20% from the total native metal concentration is normally sufficient to give a reliable increase in the isotopic ratio by ICP-MS. The isotope concentration of the spike solution may be lower when measuring mineral and/or alkaline soils with low metal lability.

Isotopes with a low natural abundance are preferentially chosen as the spike isotope so that a very small addition results in a significant increase in isotopic abundance. In choosing a spike isotope it is also important to consider analytical issues that may result from isobaric interferences in ICP-MS analysis. Using small spike additions is also important to keep costs low. However, typically the cost of the stable isotopes used for each sample, run with 4 replicates, amounts to less than £ 0.20 which is trivial compared to the operational costs of the ICP-MS.

Solubility modelling, using the labile fraction as an input, is problematic when metal lability is overestimated by simple chemical extraction (Chapter 4). Although, single chemical extraction is faster than using ID (c.1 hr for a 0.43 M HNO_3 extraction c.f. 6 to 7 days for ID) in most cases the ID method gave a more reliable result, as judged by prediction of solubility. This was particularly clear in soils with neutral or high pH as the nitric acid mobilized non-labile metal and dissolved potential binding phases such as Ca carbonate. The 0.43 M HNO_3 extraction method may only be suitable for estimating lability in organic rich acidic soils (Chapter 3). The additional time required using the ID approach should also be explored further to establish whether e.g. the (pre)equilibrium time can be shortened without affecting the results.

7.2.2 Effect of soil properties on metal lability

The current study on metal salt amended (MA) and urban contaminated soils (Nottingham, Wolverhampton and London) suggested that both soil properties and sources of contamination can affect the lability of trace metals and that the effect was metal specific. In soils where metals were added in solution (in this case as dissolved metal nitrate salts, Chapter 3), the lability was a result of metal fixation in soils during a period of 3 years incubation and possibly 10 years of dry store. Soil pH was the most important factor controlling the lability of Ni, Cu, Zn, Cd and Pb, although the effect was less significant for Cu.

The mechanisms by which soil properties determine metal fixation have been investigated in several studies (e.g. Ma et al., 2006b, Tye et al., 2003). In this work (Chapter 3) an 'S-shaped' logistic equation was used to describe lability as a function of soil pH, as the single most important soil property determining metal E-value. The advantage of this model over a multiple-regression function is that it offers more realistic boundaries to model outcomes (i.e. %E-values within the 0 – 100% range). The results suggested that mineral (hydr)oxides are important fixation phases for Ni, Zn and Cd, as metals diffuse into micropores and/or incorporate into the mineral structure (Jacquat et al., 2009; Manceau et al., 2000; Degryse et al., 2011); while Pb may also be strongly affected by organic matter in soils as it was also found in other modelling practices for Pb sorption in soils (Gustafsson et al., 2011; Guo et al., 2006). Soils contaminated by petrol-derived Pb also showed evidence that lability was dependent on soil properties and in rural roadside soils pH was again the most important factor.

7.2.3 Effect of contaminant sources on metal lability

Although, for soil amended with metals in the laboratory and rural roadside soils, empirical models have been shown to be relatively successful in predicting lability from soil properties, such models are not applicable for historically contaminated urban soils as the effect of contaminant source largely subsumes the effect of soil properties on metal lability. Metals in such soils are typically less labile and less soluble than in soils amended with dissolved metal salts, even after an extended period of equilibration. Investigation of rural roadside soils for Pb showed that, although both petrol-derived and indigenous coal Pb contributed to both the labile and non-labile fractions, there was clear evidence that petrol-derived Pb remained more labile than Pb from indigenous ore or coal, and the proportion of petrol-derived Pb that was labile was strongly correlated with soil pH.

In soils contaminated by a wide range of Pb sources sequential extraction showed that no single SEP fraction reflected the labile fraction of Pb; in agreement with previous studies by Atkinson et al. (2011) and Ahnstrom and Parker (2001). However, the non-labile Pb in both roadside and urban soils from London was likely to be a combination of Pb occlusion within Fe oxides and primary minerals, whilst for sewage sludge amended soils, the SEP residual fraction could be identified very closely with the (isotopically) non-labile pool of Pb possibly a consequence of Pb-phosphate interactions, either during sludge processing or following application to soil.

The urban soils collected in Nottingham, Wolverhampton (Urban soils in Chapter 3) and London (Chapter 5) were contaminated with metals from complex mixed sources. It is difficult to draw unequivocal conclusions about precisely how different sources affect metal lability as the original form of the metal when it was added is generally unknown.

Therefore, in future work, speciation information could assist in studies investigating metal binding to solid phases in historically contaminated urban soils. This will help to understand which metal species are isotopically exchangeable and how lability is affected. A limited number of studies on Zn and Cd have attempted to couple ID with advanced spectroscopic techniques such as X-ray Absorption Spectroscopy (XAS) (Sarret et al., 2004; Ahmed et al., 2009; Degryse et al., 2011). Degryse et al. (2011) found that the 'sorbed fraction' of Zn was always larger than the 'isotopically exchangeable fraction' but was unable to establish the precise identity of the labile adsorbed and non-labile species.

7.2.4 Predicting solubility from E-values

Both Freundlich based empirical models and mechanistic geochemical speciation models can be used to predict trace metal solubility in whole soil suspensions. The empirical model developed in Chapter 4 may provide an apparently superior fit to the solubility data. However, such a comparison is probably unreasonable because the regression model coefficients are optimised to fit the data whereas mechanistic models are independently parameterised using studies on single, 'purified' geocolloidal adsorbents. Furthermore, whereas mechanistic models allow for changing conditions, such as solid:solution ratio, metal ion competition, changes in anion complexation, temperature, ionic strength etc., the robustness of a regression model parameterised on an 'operationally uniform' dataset remains untested in this, and other, regards.

Over-prediction of solution metal concentration by WHAM-VII was observed for Ni, Cu, Zn, Cd and Pb, and was most strongly correlated with soil pH. Fractionation information from WHAM suggested that the source of the model's underestimation of metal binding most likely lay with Fe and Mn oxides for Ni, Zn and Cd and HA for Cu. Prediction of metal solubility was improved by increasing the notional surface area of Fe oxides. Another explanation of the over prediction is the absence of potential binding phases in the model, such as calcium carbonate, calcium phosphate and clay edges which are likely to be much more important in high pH soils. With increasing knowledge of cation binding to different soil fractions, it is possible to include more binding phases into mechanistic models. Thus, for example, Ahmed et al. (2008) studied the kinetics of Cd sorption, desorption and fixation by suspended particulate calcite using radioactive ^{109}Cd. Including description of such additional processes is likely to improve model predictions especially in high pH soils where the most significant underestimation of binding was observed. Therefore, a more systematic investigation of (i) model deficiencies and (ii) the potential for other mechanisms of metal ion retention in soils is required.

Apart from development of the WHAM model, further work is also required in determination of input variables. As the model was parameterized on pure substances, prediction of solubility relies on precise determination of the variables which include the reactive metal fraction and important binding phases. The advantage of using E-values to represent the 'total reactive metal fraction', instead of 0.43 M HNO_3 extractable metal, has already been demonstrated in Chapter 4 as it reduces the deviation between the measure and predicted solubility. It is reasonable to assume that estimation of binding phases may also affect model prediction. For example, in the current study, the concentrations of HA and FA in soil were directly measured as DOC following alkaline extraction. By contrast, in several published studies particulate HA has been estimated from total organic carbon (Buekers et al., 2008b; Weng et al., 2002). Figure 7-1 shows the difference between measured and estimated

HA for both MA and Urban soils, assuming that SOM was all HA with 50% of that HA active and the remainder inert (Weng et al., 2002). Generally, estimated HA was higher than measured values and the deviation was greater in Urban soils. Evidence of coal observed at many of the Urban soil sampling sites is likely to contribute to the large overestimation of HA content (e.g. WV28 and WV29). Overestimation of the HA binding phase would reduce the difference between measured and predicted solubility, especially in high pH Urban soils where overestimation of the solubility was observed. This may also lead to incorrect speciation information being given by WHAM. Another problem may be the determination of clay content, as WHAM does not classify different clay minerals but instead uses a single parameter of cation exchange capacity for all clay content. Sorption of trace metals on clay surfaces is strongly affected by the mineralogy of the clay (Tack, 2010), which cannot be easily estimated according to particle size. No account is taken of specific adsorption on clay edges within the WHAM model.

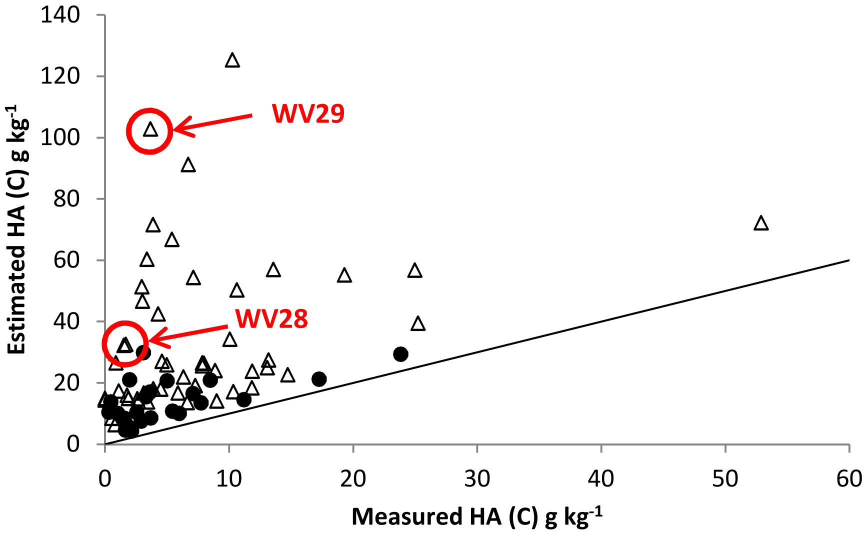


Figure 7-1: Comparison between measured and estimated C content in HA (g kg^{-1}) in soils (● MA soils, Δ Urban soils). The solid line is the 1:1 line.

WHAM also uses the explicit assumption of all metal bound to HA and FA is labile. Although the model accommodates a small number of very strongly binding organic sites on HA and FA there is still the assumption of instantaneous, reversible equilibrium that characterises labile metal. However, non-labile Cu, Zn and Pb fractions were observed on humic acids extracted from grassland and peat soils in the current work, by using SEC in conjunction with EDTA extraction and ID. Evidence of metal fixation on HA during 40 days incubation was observed for all three metals with a substantial proportion (≥14%) of metal left on HA after 0.05 M EDTA extraction and an even larger proportion after 160 days fixation (≥22%).

The isotopic abundance in the labile fraction after reaching equilibrium was used to calculate the metal lability on HA and the results indicated that the proportion of non-labile Cu on HA can be 40-50%. The SEC approach combined with the ID method could be applied to quantify metal lability in other metal fractions in the solution phase such as FA and possibly nano-particulate mineral (hydr)oxides. The results would certainly help to understand metal lability in the solution phase as well as improve model prediction of metal speciation in soils.

8 REFERENCES

Ahmed, I.A.M., Crout, N.M.J., Young, S.D. (2008) Kinetics of Cd sorption, desorption and fixation by calcite: A long-term radiotracer study. *Geochimica Et Cosmochimica Acta* **72**, 1498-1512.

Ahmed, I.A.M., Young, S.D., Crout, N.M.J. (2006) Time-dependent sorption of Cd2+ on CaX zeolite: Experimental observations and model predictions. *Geochimica Et Cosmochimica Acta* **70**, 4850-4861.

Ahmed, I.A.M., Young, S.D., Mosselmans, J.F.W., Crout, N.M.J., Bailey, E.H. (2009) Coordination of Cd^{2+} ions in the internal pore system of zeolite-X: A combined EXAFS and isotopic exchange study. *Geochimica Et Cosmochimica Acta* **73**, 1577-1587.

Ahnstrom, Z.A.S., Parker, D.R. (2001) Cadmium reactivity in metal-contaminated soils using a coupled stable isotope dilution-sequential extraction procedure. *Environ. Sci. Technol.* **35**, 121-126.

Almas, A.R., Lofts, S., Mulder, J., Tipping, E. (2007) Solubility of major cations and Cu, Zn and Cd in soil extracts of some contaminated agricultural soils near a zinc smelter in Norway: modelling with a multisurface extension of WHAM. *Eur. J. Soil Sci.* **58**, 1074-1086.

Anschutz, P., Zhong, S.J., Sundby, B., Mucci, A., Gobeil, C. (1998) Burial efficiency of phosphorus and the geochemistry of iron in continental margin sediments. *Limnology and Oceanography* **43**, 53-64.

Antelo, J., Fiol, S., Marino, S., Arce, F., Gondar, D., Lopez, R. (2009) Copper adsorption on humic acid coated gibbsite: comparison with single sorbent systems. *Environ. Chem.* **6**, 535-543.

Appleton, J.D., Cave, M.R., Wragg, J. (2012) Modelling lead bioaccessibility in urban topsoils based on data from Glasgow, London, Northampton and Swansea, UK. *Environ. Pollut.* **171**, 265-272.

Arunachalam, J., Emons, H., Krasnodebska, B., Mohl, C. (1996) Sequential extraction studies on homogenized forest soil samples. *Sci. Total Environ.* **181**, 147-159.

Asakawa, D., Iimura, Y., Kiyota, T., Yanagi, Y., Fujitake, N. (2011) Molecular size fractionation of soil humic acids using preparative high performance size-exclusion chromatography. *J. Chromatogr. A* **1218**, 6448-6453.

Asakawa, D., Kiyota, T., Yanagi, Y., Fujitake, N. (2008) Optimization of conditions for high-performance size-exclusion chromatography of different soil humic acids. *Analytical Sciences* **24**, 607-613.

Atkinson, N.R., Bailey, E.H., Tye, A.M., Breward, N., Young, S.D. (2011) Fractionation of lead in soil by isotopic dilution and sequential extraction. *Environ. Chem.* **8**, 493-500.

ATSDR. (2012). Toxicological profile for RDX. Atlanta, GA: U.S. Department of Health and Human Services. Agency for Toxic Substances and Disease Registry.

Ayoub, A.S., McGaw, B.A., Shand, C.A., Midwood, A.J. (2003) Phytoavailability of Cd and Zn in soil estimated by stable isotope exchange and chemical extraction. *Plant and Soil* **252**, 291-300.

Bacon, J.R., Farmer, J.G., Dunn, S.M., Graham, M.C., Vinogradoff, S.I. (2006) Sequential extraction combined with isotope analysis as a tool for the investigation of lead mobilisation in soils: Application to organic-rich soils in an upland catchment in Scotland. *Environ. Pollut.* **141**, 469-481.

Bacon, J.R., Hewitt, I.J. (2005) Heavy metals deposited from the atmosphere on upland Scottish soils: Chemical and lead isotope studies of the association of metals with soil components. *Geochimica Et Cosmochimica Acta* **69**, 19-33.

Bacon, J.R., Jones, K.C., McGrath, S.P., Johnston, A.E. (1996) Isotopic character of lead deposited from the atmosphere at a grassland site in the United Kingdom since 1860. *Environ. Sci. Technol.* **30**, 2511-2518.

Bailey, E.H., Mosselmans, J.F.W., Young, S.D. (2005) Time-dependent surface reactivity of Cd sorbed on calcite, hydroxylapatite and humic acid. *Mineralogical Magazine* **69**, 563-575.

Baker, J., Peate, D., Waight, T., Meyzen, C. (2004) Pb isotopic analysis of standards and samples using a Pb-207-Pb-204 double spike and thallium to correct for mass bias with a double-focusing MC-ICP-MS. *Chemical Geology* **211**, 275-303.

Basta, N.T., Tabatabai, M.A. (1992) Effect of cropping systmes on adsorption of metals by soils. II. Effect of pH. *Soil Science* **153**, 195-204.

Benedetti, M.F., VanRiemsdijk, W.H., Koopal, L.K., Kinniburgh, D.G., Gooddy, D.C., Milne, C.J. (1996) Metal ion binding by natural organic matter: From the model to the field. *Geochimica Et Cosmochimica Acta* **60**, 2503-2513.

Bergkvist, P., Berggren, D., Jarvis, N. (2005) Cadmium solubility and sorption in a long-term sludge-amended arable soil. *J. Environ. Qual.* **34**, 1530-1538.

Bermond, A. (2001) Limits of sequential extraction procedures re-examined with emphasis on the role of H+ ion reactivity. *Anal. Chim. Acta* **445**, 79-88.

Biester, H., Scholz, C. (1997) Determination of mercury binding forms in contaminated soils: Mercury pyrolysis versus sequential extractions. *Environ. Sci. Technol.* **31**, 233-239.

Boddey, R.M., deOliveira, O.C., Alves, B.J.R., Urquiaga, S. (1995) Field application of the N-15 isotope dilution technique for the reliable quantification of plant-associated biological nitrogen fixation. *Fertilizer Research* **42**, 77-87.

Bonten, L.T.C., Groenenberg, J.E., Weng, L., van Riemsdijk, W.H. (2008) Use of speciation and complexation models to estimate heavy metal sorption in soils. *Geoderma* **146**, 303-310.

Brazauskiene, D.M., Paulauskas, V., Sabiene, N. (2008) Speciation of Zn, Cu, and Pb in the soil depending on soil texture and fertilization with sewage sludge compost. *Journal of Soils and Sediments* **8**, 184-192.

Bridges, E.M. (1991). Waste materials in urban soils, in: Bullock, P., Gregory, P.J., British Society of Soil Science., Nature Conservancy Council. (Eds.), *Soils in the urban environment*. Blackwell Scientific, Oxford, pp. ix, 174 p.

Bruemmer, G.W., Gerth, J., Tiller, K.G. (1988) Reaction kinetics of the adsorption and desorption of nickel, zinc and cadmium by goethite: I. Adsorption and diffusion of metals. *Journal of Soil Science* **39**, 37-52.

Buekers, J., Amery, F., Maes, A., Smolders, E. (2008a) Long-term reactions of Ni, Zn and Cd with iron oxyhydroxides depend on crystallinity and structure and on metal concentrations. *Eur. J. Soil Sci.* **59**, 706-715.

Buekers, J., Degryse, F., Maes, A., Smolders, E. (2008b) Modelling the effects of ageing on Cd, Zn, Ni and Cu solubility in soils using an assemblage model. *Eur. J. Soil Sci.* **59**, 1160-1170.

Buekers, J., Van Laer, L., Amery, F., Van Buggenhout, S., Maes, A., Smolders, E. (2007) Role of soil constituents in fixation of soluble Zn, Cu, Ni and Cd added to soils. *Eur. J. Soil Sci.* **58**, 1514-1524.

Burba, P. (1994) Labile/inert metal species in aquatic humic substances - an ion-exchange study. *Fresenius J. Anal. Chem.* **348**, 301-311.

Cabaniss, S.E. (1990) pH and ionic-strength effects on nickel fulvic-acid dissociation kinetics. *Environ. Sci. Technol.* **24**, 583-588.

Cances, B., Ponthieu, M., Castrec-Rouelle, M., Aubry, E., Benedetti, M.F. (2003) Metal ions speciation in a soil and its solution: experimental data and model results. *Geoderma* **113**, 341-355.

Charlatchka, R., Cambier, P. (2000) Influence of reducing conditions on solubility of trace metals in contaminated soils. *Water Air and Soil Pollution* **118**, 143-167.

Chenery, S.R., Izquierdo, M., Marzouk, E., Klinck, B., Palumbo-Roe, B., Tye, A.M. (2012) Soil-plant interactions and the uptake of Pb at abandoned mining sites in the Rookhope catchment of the N. Pennines, UK - A Pb isotope study. *Sci. Total Environ.* **433**, 547-560.

Cheng, T., De Schamphelaere, K., Lofts, S., Janssen, C., Allen, H.E. (2005) Measurement and computation of zinc binding to natural dissolved organic matter in European surface waters. *Anal. Chim. Acta* **542**, 230-239.

Chilimba, A.D.C., Young, S.D., Black, C.R., Meacham, M.C., Lammel, J., Broadley, M.R. (2012) Assessing residual availability of selenium applied to maize crops in Malawi. *Field Crops Research* **134**, 11-18.

Chin, Y.P., Gschwend, P.M. (1991) The abundance, distribution, and configuration of porewater organic colloids in recent sediments. *Geochimica Et Cosmochimica Acta* **55**, 1309-1317.

Choppin, G.R., Clark, S.B. (1991) The kinetic interactions of metal-ions with humic acids. *Mar. Chem.* **36**, 27-38.

Clark, M.W., Davies-McConchie, F., McConchie, D., Birch, G.F. (2000) Selective chemical extraction and grainsize normalisation for environmental assessment of anoxic sediments: validation of an integrated procedure. *Sci. Total Environ.* **258**, 149-170.

Collins, R.N. (2004) Separation of low-molecular mass organic acid-metal complexes by high-performance liquid chromatography. *J. Chromatogr. A* **1059**, 1-12.

Collins, R.N., Merrington, G., McLaughlin, M.J., Morel, J.L. (2003) Organic ligand and pH effects on isotopically exchangeable cadmium in polluted soils. *Soil Sci. Soc. Am. J.* **67**, 112-121.

Crout, N.M.J., Tye, A.M., Zhang, H., McGrath, S.P., Young, S.D. (2006) Kinetics of metal fixation in soils: Measurement and modeling by isotopic dilution. *Environ. Toxicol. Chem.* **25**, 659-663.

Daviescolley, R.J., Nelson, P.O., Williamson, K.J. (1984) Copper and cadmium uptake by estuarine sedimentary phases. *Environ. Sci. Technol.* **18**, 491-499.

Davis, J.A., Fuller, C.C., Cook, A.D. (1987) A model for trace-metal sorption processes at the calcite surface - adsorption of Cd^{2+} and subsequent solid-solution formation. *Geochimica Et Cosmochimica Acta* **51**, 1477-1490.

de Vries, W., Romkens, P., Bonten, L.T.C. (2008) Spatially explicit integrated risk assessment of present soil concentrations of cadmium, lead, copper and zinc in the Netherlands. *Water Air and Soil Pollution* **191**, 199-215.

Debievre, P., Barnes, I.L. (1985) Table of the isotopic composition of the elements as determined by mass-spectrometry. *Int. J. Mass Spectrom. Ion Process.* **65**, 211-230.

Degryse, F., Broos, K., Smolders, E., Merckx, R. (2003) Soil solution concentration of Cd and Zn can be predicted with a $CaCl_2$ soil extract. *Eur. J. Soil Sci.* **54**, 149-157.

Degryse, F., Buekers, J., Smolders, E. (2004) Radio-labile cadmium and zinc in soils as affected by pH and source of contamination. *Eur. J. Soil Sci.* **55**, 113-121.

Degryse, F., Smolders, E., Parker, D.R. (2009) Partitioning of metals (Cd, Co, Cu, Ni, Pb, Zn) in soils: concepts, methodologies, prediction and applications - a review. *Eur. J. Soil Sci.* **60**, 590-612.

Degryse, F., Voegelin, A., Jacquat, O., Kretzschmar, R., Smolders, E. (2011) Characterization of zinc in contaminated soils: complementary insights from isotopic exchange, batch extractions and XAFS spectroscopy. *Eur. J. Soil Sci.* **62**, 318-330.

Degryse, F., Waegeneers, N., Smolders, E. (2007) Labile lead in polluted soils measured by stable isotope dilution. *Eur. J. Soil Sci.* **58**, 1-7.

Dermatas, D., Chrysochoou, M., Grubb, D.G., Xu, X.F. (2008) Phosphate treatment of firing range soils: Lead fixation or phosphorus release? *J. Environ. Qual.* **37**, 47-56.

Di Toro, D.M., Allen, H.E., Bergman, H.L., Meyer, J.S., Paquin, P.R., Santore, R.C. (2001) Biotic ligand model of the acute toxicity of metals. 1. Technical basis. *Environ. Toxicol. Chem.* **20**, 2383-2396.

Dong, D.M., Hua, X.Y., Li, Y., Zhang, J.J., Yan, D.X. (2003) Cd adsorption properties of components in different freshwater surface coatings: The important role of ferromanganese oxides. *Environ. Sci. Technol.* **37**, 4106-4112.

Du Laing, G., Rinklebe, J., Vandecasteele, B., Meers, E., Tack, F.M.G. (2009) Trace metal behaviour in estuarine and riverine floodplain soils and sediments: A review. *Sci. Total Environ.* **407**, 3972-3985.

Elfalaky, A.A., Aboulroos, S.A., Lindsay, W.L. (1991) Measurement of Cadmium activities in slightly acidic to alkaline soils. *Soil Sci. Soc. Am. J.* **55**, 974-979.

Ellam, R.M. (2010) The graphical presentation of lead isotope data for environmental source apportionment. *Sci. Total Environ.* **408**, 3490-3492.

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

Elliott, H.A., Denneny, C.M. (1982) Soil adsorption of cadmium from solutions containing organic ligands. *J. Environ. Qual.* **11**, 658-663.

Elzinga, E.J., van Grinsven, J.J.M., Swartjes, F.A. (1999) General purpose Freundlich isotherms for cadmium, copper and zinc in soils. *Eur. J. Soil Sci.* **50**, 139-149.

Emmanuel, S., Erel, Y. (2002) Implications from concentrations and isotopic data for Pb partitioning processes in soils. *Geochimica Et Cosmochimica Acta* **66**, 2517-2527.

Erel, Y., Axelrod, T., Veron, A., Mahrer, Y., Katsafados, P., Dayan, U. (2002) Transboundary atmospheric lead pollution. *Environ. Sci. Technol.* **36**, 3230-3233.

Erel, Y., Veron, A., Halicz, L. (1997) Tracing the transport of anthropogenic lead in the atmosphere and in soils using isotopic ratios. *Geochimica Et Cosmochimica Acta* **61**, 4495-4505.

Esnaola, M.V., Millan, E. (1998) Evaluation of heavy metal lability in polluted soils by a cation exchange batch procedure. *Environ. Pollut.* **99**, 79-86.

Eusterhues, K., Rumpel, C., Kogel-Knabner, I. (2005) Organo-mineral associations in sandy acid forest soils: importance of specific surface area, iron oxides and micropores. *Eur. J. Soil Sci.* **56**, 753-763.

Fairbrother, A., Glazebrook, P.W., Van Straalen, N., Tarazona, J.V. (1999). *Test Methods for Hazard Determination of Metals and Sparingly Soluble Metal*

Compounds in Soils: Summary of a SETAC Pellston Workshop. Society of Environmental Toxicology and Chemistry (SETAC), San Lorenzo de El Escorial, Spain.

Farmer, J.G., Broadway, A., Cave, M.R., Wragg, J., Fordyce, F.M., Graham, M.C., Ngwenya, B.T., Bewley, R.J.F. (2011) A lead isotopic study of the human bioaccessibility of lead in urban soils from Glasgow, Scotland. *Sci. Total Environ.* **409**, 4958-4965.

Farmer, J.G., Eades, L.J., Graham, M.C. (1999) The lead content and isotopic composition of British coals and their implications for past and present releases of lead to the UK environment. *Environmental Geochemistry and Health* **21**, 257-272.

Fischer, B.E., Haring, U.K., Tribolet, R., Sigel, H. (1979) Metal ion/buffer interactions. Stability of binary and ternary complexes containing 2-amino-2(hydroxymethyl)-1,3-propanediol (Tris) and adenosine 5'-triphosphate (ATP). *European Journal of Biochemistry* **94**, 523-530.

Fischer, L., Brummer, G.W., Barrow, N.J. (2007) Observations and modelling of the reactions of 10 metals with goethite: adsorption and diffusion processes. *Eur. J. Soil Sci.* **58**, 1304-1315.

Flament, P., Bertho, M.L., Deboudt, K., Puskaric, E. (1996) Changes in the lead content of atmospheric aerosols above the Eastern Channel between 1982/83 and 1994. *Sci. Total Environ.* **192**, 193-206.

Fujii, R., Corey, R.B. (1986) Estimation of isotopically exchangeable cadmium and zinc in soils. *Soil Sci. Soc. Am. J.* **50**, 306-308.

Fujiwara, F., Rebagliati, R.J., Marrero, J., Gomez, D., Smichowski, P. (2011) Antimony as a traffic-related element in size-fractionated road dust samples collected in Buenos Aires. *Microchemical Journal* **97**, 62-67.

Gabler, H.E., Bahr, A., Heidkamp, A., Utermann, J. (2007) Enriched stable isotopes for determining the isotopically exchangeable element content in soils. *Eur. J. Soil Sci.* **58**, 746-757.

Gambrell, R.P. (1994) Trace and toxic metals in wetlands - A review. *J. Environ. Qual.* **23**, 883-891.

GarciaDelgado, R.A., GarciaHerruzo, F., RodriguezMaroto, J.M., Vereda, C. (1996) Influence of soil carbonates in lead fixation. *Journal of Environmental Science and Health Part a-Environmental Science and Engineering & Toxic and Hazardous Substance Control* **31**, 2099-2109.

Garciamiragaya, J. (1984) Levels, chemical fractionation, and solubility of lead in roadside soils of caracas, venezuela. *Soil Science* **138**, 147-152.

Gerard, E., Echevarria, G., Sterckeman, T., Morel, J.L. (2000) Cadmium availability to three plant species varying in cadmium accumulation pattern. *J. Environ. Qual.* **29**, 1117-1123.

Gerritse, R.G., Vandriel, W. (1984) The relationship between adsorption of trace metals, organic matter, and pH in temerate soils. *J. Environ. Qual.* **13**, 197-204.

Graney, J.R., Halliday, A.N., Keeler, G.J., Nriagu, J.O., Robbins, J.A., Norton, S.A. (1995) Isotopic record of lead pollution in lake-sediments from the northeastern united-states. *Geochimica Et Cosmochimica Acta* **59**, 1715-1728.

Groenenberg, J.E., Dijkstra, J.J., Bonten, L.T.C., de Vries, W., Comans, R.N.J. (2012) Evaluation of the performance and limitations of empirical partition-relations and process based multisurface models to predict trace element solubility in soils. *Environ. Pollut.* **166**, 98-107.

Groenenberg, J.E., Romkens, P., Comans, R.N.J., Luster, J., Pampura, T., Shotbolt, L., Tipping, E., de Vries, W. (2010) Transfer functions for solid-solution partitioning of cadmium, copper, nickel, lead and zinc in soils: derivation of relationships for free metal ion activities and validation with independent data. *Eur. J. Soil Sci.* **61**, 58-73.

Guo, X.Y., Zhang, S.Z., Shan, X.Q., Luo, L., Pei, Z.G., Zhu, Y.G., Liu, T., Xie, Y.N., Gault, A. (2006) Characterization of Pb, Cu, and Cd adsorption on particulate organic matter in soil. *Environ. Toxicol. Chem.* **25**, 2366-2373.

Gupta, S.K., Aten, C. (1993) Comparison and evaluation of extraction media and their suitability in a simple model to predict the biological relevance of heavy metal concentrations in contaminated soils. *Int. J. Environ. Anal. Chem.* **51**, 25-46.

Gustafsson, J.P. (2006) Arsenate adsorption to soils: Modelling the competition from humic substances. *Geoderma* **136**, 320-330.

Gustafsson, J.P., Tiberg, C., Edkymish, A., Kleja, D.B. (2011) Modelling lead(II) sorption to ferrihydrite and soil organic matter. *Environ. Chem.* **8**, 485-492.

Gustafsson, J.P., van Schaik, J.W.J. (2003) Cation binding in a mor layer: batch experiments and modelling. *Eur. J. Soil Sci.* **54**, 295-310.

Hammer, D., Keller, C., McLaughlin, M.J., Hamon, R.E. (2006) Fixation of metals in soil constituents and potential remobilization by hyperaccumulating and non-hyperaccumulating plants: Results from an isotopic dilution study. *Environ. Pollut.* **143**, 407-415.

Hamon, R.E., McLaughlin, M.J. (2002) Interferences in the determination of isotopically exchangeable P in soils and a method to minimise them. *Aust. J. Soil Res.* **40**, 1383-1397.

Hamon, R.E., McLaughlin, M.J., Cozens, G. (2002) Mechanisms of attenuation of metal availability in in situ remediation treatments. *Environ. Sci. Technol.* **36**, 3991-3996.

Hamon, R.E., Parker, D.R., Lombi, E. (2008). Advances in isotopic dilution techniques in trace element research: a review of methodologies, benefits, and limitations, in: Sparks, D.L. (Ed.), *Advances in Agronomy, Vol 99*, pp. 289-343.

Heidmann, I., Christl, I., Kretzschmar, R. (2005) Sorption of Cu and Pb to kaolinite-fulvic acid colloids: Assessment of sorbent interactions. *Geochimica Et Cosmochimica Acta* **69**, 1675-1686.

Hooda, P.S., Alloway, B.J. (1994) Sorption of Cd and Pb by selected temperate and semi-arid soils: Effects of sludge application and aging of sludges soils. *Water Air and Soil Pollution* **74**, 235-250.

Hough, R.L., Tye, A.M., Crout, N.M.J., McGrath, S.P., Zhang, H., Young, S.D. (2005) Evaluating a 'Free Ion Activity Model' applied to metal uptake by Lolium perenne L. grown in contaminated soils. *Plant and Soil* **270**, 1-12.

Huang, X.D., Olmez, I., Aras, N.K., Gordon, G.E. (1994) Emissions of trace-elements from moto vehicles - potential marker elements and source composition profile. *Atmospheric Environment* **28**, 1385-1391.

Huang, Z.Y., Chen, T., Yu, J.A., Zeng, X.C., Huang, Y.F. (2011) Labile Cd and Pb in vegetable-growing soils estimated with isotope dilution and chemical extractants. *Geoderma* **160**, 400-407.

Hughes, J.C., Noble, A.D. (1991) Extraction of chromium, nickel and iron and the availability of chromium and nickel to plants from some serpentinite ‐ derived soils from the eastern Transvaal as revealed by various single and sequential extraction techniques. *Communications in Soil Science and Plant Analysis* **22**, 1753-1766.

Imperato, M., Adamo, P., Naimo, D., Arienzo, M., Stanzione, D., Violante, P. (2003) Spatial distribution of heavy metals in urban soils of Naples city (Italy). *Environ. Pollut.* **124**, 247-256.

Ivezic, V., Almas, A.R., Singh, B.R. (2012) Predicting the solubility of Cd, Cu, Pb and Zn in uncontaminated Croatian soils under different land uses by applying established regression models. *Geoderma* **170**, 89-95.

Izquierdo, M., Tye, A.M., Chenery, S.R. (2012) Sources, lability and solubility of Pb in alluvial soils of the River Trent catchment, UK. *Sci. Total Environ.* **433**, 110-122.

Izquierdo, M., Tye, A.M., Chenery, S.R. (2013) Lability, solubility and speciation of Cd, Pb and Zn in alluvial soils of the River Trent catchment UK. *Environmental Science: Processes & Impacts* **in press**.

Jackson, B.P., Ranville, J.F., Bertsch, P.M., Sowder, A.G. (2005) Characterization of colloidal and humic-bound Ni and U in the "dissolved" fraction of contaminated sediment extracts. *Environ. Sci. Technol.* **39**, 2478-2485.

Jacquat, O., Voegelin, A., Kretzschmar, R. (2009) Local coordination of Zn in hydroxy-interlayered minerals and implications for Zn retention in soils. *Geochimica Et Cosmochimica Acta* **73**, 348-363.

Johnson, C.C., Breward, N., Ander, E.L., Ault, L. (2005) G-BASE: baseline geochemical mapping of great Britain and northern Ireland. *Geochemistry-Exploration Environment Analysis* **5**, 347-357.

Johnson, C.E., Siccama, T.G., Driscoll, C.T., Likens, G.E., Moeller, R.E. (1995) Changes in lead biogeochemistry in response to decreasing atmospheric inputs. *Ecological Applications* **5**, 813-822.

Karathanasis, A.D. (1999) Subsurface migration of copper and zinc mediated by soil colloids. *Soil Sci. Soc. Am. J.* **63**, 830-838.

Kaste, J.M., Bostick, B.C., Friedland, A.J., Schroth, A.W., Siccama, T.G. (2006) Fate and speciation of gasoline-derived lead in organic horizons of the northeastern USA. *Soil Sci. Soc. Am. J.* **70**, 1688-1698.

Kayhanian, M. (2012) Trend and concentrations of legacy lead (Pb) in highway runoff. *Environ. Pollut.* **160**, 169-177.

Kiekens, L. (1995). Zinc, in: Alloway, B.J. (Ed.), *Heavy metals in soils*, 2nd ed ed. Blackie Academic & Professional, London, pp. xiv, 368 p.

Kim, N.D., Fergusson, J.E. (1991) Effictiveness of a commonly used sequential extraction technique in determining the speciation of cadmium in soils. *Sci. Total Environ.* **105**, 191-209.

Kinniburgh, D.G., van Riemsdijk, W.H., Koopal, L.K., Borkovec, M., Benedetti, M.F., Avena, M.J. (1999) Ion binding to natural organic matter: competition, heterogeneity, stoichiometry and thermodynamic consistency. *Colloids and Surfaces a-Physicochemical and Engineering Aspects* **151**, 147-166.

Knight, B.P., Chaudri, A.M., McGrath, S.P., Giller, K.E. (1998) Determination of chemical availability of cadmium and zinc in soils using inert soil moisture samplers. *Environ. Pollut.* **99**, 293-298.

Komarek, M., Ettler, V., Chrastny, V., Mihaljevic, M. (2008) Lead isotopes in environmental sciences: A review. *Environ. Int.* **34**, 562-577.

Kosmulski, M. (2009) pH-dependent surface charging and points of zero charge. IV. Update and new approach. *Journal of Colloid and Interface Science* **337**, 439-448.

Kozai, N., Ohnuki, T., Iwatsuki, T. (2013) Characterization of saline groundwater at Horonobe, Hokkaido, Japan by SEC-UV-ICP-MS: Speciation of uranium and iodine. *Water Res.* **47**, 1570-1584.

Krishnamurti, G.S.R., Huang, P.M., Vanrees, K.C.J., Kozak, L.M., Rostad, H.P.W. (1995) Speciation of particulate-bound cadmium of soils and its bioavailability. *Analyst* **120**, 659-665.

Laborda, F., Ruiz-Begueria, S., Bolea, E., Castillo, J.R. (2009) Functional speciation of metal-dissolved organic matter complexes by size exclusion chromatography coupled to inductively coupled plasma mass spectrometry and deconvolution analysis. *Spectrochimica Acta Part B-Atomic Spectroscopy* **64**, 392-398.

Laborda, F., Ruiz-Begueria, S., Bolea, E., Castillo, J.R. (2011) Study of the size-based environmental availability of metals associated to natural organic matter by stable isotope exchange and quadrupole inductively coupled plasma mass spectrometry coupled to asymmetrical flow field flow fractionation. *J. Chromatogr. A* **1218**, 4199-4205.

Laing, G.D. (2010a). Chapter 4. Analysis and Fractionation of Trace Elements in Soils, in: Hooda, P.S. (Ed.), *Trace elements in soils*. Wiley, Chichester, pp. xix, 596 p.

Laing, G.D. (2010b). Chapter 5. Fractionation and Speciation of Trace Elements in Soil Solution, in: Hooda, P.S. (Ed.), *Trace elements in soils*. Wiley, Chichester, pp. xix, 596 p.

Lamy, I., Bourgeois, S., Bermond, A. (1993) Soil cadmium mobility as a consequence of sewage-sludge disposal. *J. Environ. Qual.* **22**, 731-737.

Lang, F., Kaupenjohann, M. (2003) Effect of dissolved organic matter on the precipitation and mobility of the lead compound chloropyromorphite in solution. *Eur. J. Soil Sci.* **54**, 139-147.

Ledin, M., KrantzRulcker, C., Allard, B. (1996) Zn, Cd and Hg accumulation by microorganisms, organic and inorganic soil components in multi-compartment systems. *Soil Biol. Biochem.* **28**, 791-799.

Lee, M.H., Choi, S.Y., Chung, K.H., Moon, H. (1993) Complexation of cadmium (II) with humic acids - effects fo pH and humic-acid origin. *Bull. Korean Chem. Soc.* **14**, 726-732.

Li, H.B., Yu, S., Li, G.L., Deng, H., Luo, X.S. (2011) Contamination and source differentiation of Pb in park soils along an urban-rural gradient in Shanghai. *Environ. Pollut.* **159**, 3536-3544.

Li, X.D., Coles, B.J., Ramsey, M.H., Thornton, I. (1995) Sequential extraction of soils for multielement analysis by ICP-AES. *Chemical Geology* **124**, 109-123.

Li, X.D., Thornton, I. (2001) Chemical partitioning of trace and major elements in soils contaminated by mining and smelting activities. *Appl. Geochem.* **16**, 1693-1706.

Linde, M., Oborn, I., Gustafsson, J.P. (2007) Effects of changed soil conditions on the mobility of trace metals in moderately contaminated urban soils. *Water Air and Soil Pollution* **183**, 69-83.

Lindsay, W.L., Norvell, W.A. (1978) Development of a DTPA soil test for zinc, iron, manganese, and copper. *Soil Sci. Soc. Am. J.* **42**, 421-428.

Liu, G.L., Cai, Y. (2010) Complexation of arsenite with dissolved organic matter Conditional distribution coefficients and apparent stability constants. *Chemosphere* **81**, 890-896.

Liu, G.L., Fernandez, A., Cai, Y. (2011) Complexation of Arsenite with Humic Acid in the Presence of Ferric Iron. *Environ. Sci. Technol.* **45**, 3210-3216.

Liu, W.X., Li, X.D., Shen, Z.G., Wang, D.C., Wai, O.W.H., Li, Y.S. (2003) Multivariate statistical study of heavy metal enrichment in sediments of the Pearl River Estuary. *Environ. Pollut.* **121**, 377-388.

Lock, K., Janssen, C.R. (2001) Ecotoxicity of zinc in spiked artificial soils versus contaminated field soils. *Environ. Sci. Technol.* **35**, 4295-4300.

Lofts, S., Tipping, E. (1998) An assemblage model for cation binding by natural particulate matter. *Geochimica Et Cosmochimica Acta* **62**, 2609-2625.

Lofts, S., Tipping, E. (2011) Assessing WHAM/Model VII against field measurements of free metal ion concentrations: model performance and the role of uncertainty in parameters and inputs. *Environ. Chem.* **8**, 501-516.

Lofts, S., Woof, C., Tipping, E., Clarke, N., Mulder, J. (2001) Modelling pH buffering and aluminium solubility in European forest soils. *Eur. J. Soil Sci.* **52**, 189-204.

Lombi, E., Hamon, R.E., McGrath, S.P., McLaughlin, M.J. (2003) Lability of Cd, Cu, and Zn in polluted soils treated with lime, beringite, and red mud and identification of a non-labile colloidal fraction of metals using isotopic techniques. *Environ. Sci. Technol.* **37**, 979-984.

Lopez, P.L., Graham, E.R. (1972) Labile pool and plant uptake of micronutrients: 1. Determination of labile pool of Mn, Fe, Zn, Co, and Cu in deficient soils by isotopic exchange. *Soil Science* **114**, 295-&.

Ma, H.Z., Kim, S.D., Cha, D.K., Allen, H.E. (1999) Effect of kinetics of complexation by humic acid on toxicity of copper to Ceriodaphnia dubia. *Environ. Toxicol. Chem.* **18**, 828-837.

Ma, L.Q., Rao, G.N. (1997) Chemical fractionation of cadmium, copper, nickel, and zinc in contaminated soils. *J. Environ. Qual.* **26**, 259-264.

Ma, Q.Y., Lindsay, W.L. (1993) Measurements of free zinc^{2+} activity in uncontaminated and contaminated soils using chelation. *Soil Sci. Soc. Am. J.* **57**, 963-967.

Ma, Y.B., Lombi, E., Nolan, A.L., McLaughlin, M.J. (2006a) Determination of labile Cu in soils and isotopic exchangeability of colloidal Cu complexes. *Eur. J. Soil Sci.* **57**, 147-153.

Ma, Y.B., Lombi, E., Nolan, A.L., McLaughlin, M.J. (2006b) Short-term natural attenuation of copper in soils: Effects of time, temperature, and soil characteristics. *Environ. Toxicol. Chem.* **25**, 652-658.

MacDonald, J.D., Hendershot, W.H. (2006) Modelling trace metal partitioning in forest floors of northern soils near metal smelters. *Environ. Pollut.* **143**, 228-240.

MAFF. (1993). *Code of good agricultural practice for the protection of soil*. Ministry of Agriculture Fisheries and Food, Welsh Office Agriculture Department, London.

Makino, T., Sugahara, K., Sakurai, Y., Takano, H., Kamiya, T., Sasaki, K., Itou, T., Sekiya, N. (2006) Remediation of cadmium contamination in paddy soils by washing with chemicals: Selection of washing chemicals. *Environ. Pollut.* **144**, 2-10.

Malinovsky, D., Dahlqvist, R., Baxter, D.C., Ingri, J., Rodushkin, I. (2005) Performance of diffusive gradients in thin films for measurement of the isotopic composition of soluble Zn. *Anal. Chim. Acta* **537**, 401-405.

Manceau, A., Schlegel, M.L., Musso, M., Sole, V.A., Gauthier, C., Petit, P.E., Trolard, F. (2000) Crystal chemistry of trace elements in natural and synthetic goethite. *Geochimica Et Cosmochimica Acta* **64**, 3643-3661.

Markiewiez-Patkowska, J., Hursthouse, A., Przybyla-Kij, H. (2005) The interaction of heavy metals with urban soils: sorption behaviour of Cd, Cu, Cr, Pb and Zn with a typical mixed brownfield deposit. *Environ. Int.* **31**, 513-521.

Marshall, S.J., Young, S.D., Gregson, K. (1995) Humic-acid proton equilibria - a comparison of two models and assessment of titration error. *Eur. J. Soil Sci.* **46**, 471-480.

Martinez, C.E., Motto, H.L. (2000) Solubility of lead, zinc and copper added to mineral soils. *Environ. Pollut.* **107**, 153-158.

Marzouk, E.R. (2012). Using multi-element stable isotope dilution to quantify metal reactivity in soil. University of Nottingham, United Kingdom.

Marzouk, E.R., Chenery, S.R., Young, S.D. (2013) Measuring reactive metal in soil: a comparison of multi-element isotopic dilution and chemical extraction. *Eur. J. Soil Sci.* **64**, 526-536.

McBride, M., Sauve, S., Hendershot, W. (1997) Solubility control of Cu, Zn, Cd and Pb in contaminated soils. *Eur. J. Soil Sci.* **48**, 337-346.

McBride, M.B. (1994). *Environmental chemistry of soils*. Oxford University Press, New York ; Oxford.

McCarthy, J.F., Roberson, L.E., Burrus, L.W. (1989) Association of benzo(a)pyrene with dissolved organic matter: Prediction of Kdom from structural and chemical properties of the organic matter. *Chemosphere* **19**, 1911-1920.

McGill, R.A.R., Pearce, J.M., Fortey, N.J., Watt, J., Ault, L., Parrish, R.R. (2003) Contaminant source apportionment by PIMMS lead isotope analysis and SEM-image analysis. *Environmental Geochemistry and Health* **25**, 25-32.

McLaren, R.G., Crawford, D.V. (1973) Studies on soil copper. 1. Fractionation of copper in soils. *Journal of Soil Science* **24**, 172-181.

McLaren, R.G., Crawford, D.V. (1974) Studies on soil copper 3. Isotopically exchangeable copper in soils. *Journal of Soil Science* **25**, 111-119.

McLaughlin, M.J., Smolders, E., Merckx, R. (1998). Soil-root interface: physicochemical processes, in: Huang, P.M., Adriano, D.C., Logan, T.J., Checkai, R.T. (Eds.), *Soil chemistry and ecosystem health*. Soil Science Society of America Inc., Madison, US, pp. 233-277.

McLean, J.E., Bledsoe, B.E. (1992). Behavior of metals in soils, Ground Water Issue.

Meers, E., Du Laing, G., Unamuno, V., Ruttens, A., Vangronsveld, J., Tack, F.M.G., Verloo, M.G. (2007) Comparison of cadmium extractability from soils by commonly used single extraction protocols. *Geoderma* **141**, 247-259.

Meers, E., Unamuno, V.R., Du Laing, G., Vangronsveld, J., Vanbroekhoven, K., Samson, R., Diels, L., Geebelen, W., Ruttens, A., Vandegehuchte, M., Tack, F.M.G. (2006) Zn in the soil solution of unpolluted and polluted soils as affected by soil characteristics. *Geoderma* **136**, 107-119.

Meeussen, J.C.L. (2003) ORCHESTRA: An object-oriented framework for implementing chemical equilibrium models. *Environ. Sci. Technol.* **37**, 1175-1182.

Midwood, A.J. (2007). Stable isotope methods for estimating the labile metal content of soils, in: Willey, N. (Ed.), *Phytoremediation : methods and reviews*. Humana Press, Totowa, N.J.

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

Monna, F., Dominik, J., Loizeau, J.L., Pardos, M., Arpagaus, P. (1999) Origin and evolution of Pb in sediments of Lake Geneva (Switzerland-France). Establishing a stable Pb record. *Environ. Sci. Technol.* **33**, 2850-2857.

Morin, G., Ostergren, J.D., Juillot, F., Ildefonse, P., Calas, G., Brown, G.E. (1999) XAFS determination of the chemical form of lead in smelter-contaminated soils and mine tailings: Importance of adsorption processes. *American Mineralogist* **84**, 420-434.

Nakhone, L.N., Young, S.D. (1993) The significance of (radio-)labile cadmium pools in soil. *Environ. Pollut.* **82**, 73-77.

Noble, S.R., Horstwood, M.S.A., Davy, P., Pashley, V., Spiro, B., Smith, S. (2008) Evolving Pb isotope signatures of London airborne particulate matter (PM(10)) - constraints from on-filter and solution-mode MC-ICP-MS. *Journal of Environmental Monitoring* **10**, 830-836.

Nolan, A.L., Ma, Y.B., Lombi, E., McLaughlin, M.J. (2004) Measurement of labile Cu in soil using stable isotope dilution and isotope ratio analysis by ICP-MS. *Anal. Bioanal. Chem.* **380**, 789-797.

Nolan, A.L., Ma, Y.B., Lombi, E., McLaughlin, M.J. (2009) Speciation and Isotopic Exchangeability of Nickel in Soil Solution. *J. Environ. Qual.* **38**, 485-492.

Novozamsky, I., Lexmond, T.M., Houba, V.J.G. (1993) A single extraction procedure of soil for evaluation of uptake of some heavy-metals by plants. *Int. J. Environ. Anal. Chem.* **51**, 47-58.

Nriagu, J.O. (1996) A history of global metal pollution. *Science* **272**, 223-224.

Pedrot, M., Dia, A., Davranche, M. (2009) Double pH control on humic substance-borne trace elements distribution in soil waters as inferred from ultrafiltration. *Journal of Colloid and Interface Science* **339**, 390-403.

Peijnenburg, W., Posthuma, L., Zweers, P., Baerselman, R., de Groot, A.C., Van Veen, R.P.M., Jager, T. (1999) Prediction of metal bioavailability in Dutch field soils for the oligochaete Enchytraeus crypticus. *Ecotoxicology and Environmental Safety* **43**, 170-186.

Pelekani, C., Newcombe, G., Snoeyink, V.L., Hepplewhite, C., Assemi, S., Beckett, R. (1999) Characterization of natural organic matter using high performance size exclusion chromatography. *Environ. Sci. Technol.* **33**, 2807-2813.

Piper, C.S. (1944). *Soil and plant analysis : a laboratory manual of methods for the examination of soils and the determination of the inorganic constituents of plants.* University of Adelaide, Adelaide.

Puchelt, H., Kramar, U., Cumming, G.L., Krstic, D., Noltner, T., Schottle, M., Schweikle, V. (1993) Anthropogenic pb contamination of soils, southwest germany. *Applied Geochemistry, Supplementary Issue No 2, January 1993: Environmental Geochemistry*, 71-73.

Rate, A.W., McLaren, R.G., Swift, R.S. (1993) Response of copper (II) humic-acid dissociation kinetics to factors influencing complex stability and macromolecular conformation. *Environ. Sci. Technol.* **27**, 1408-1414.

Rawlins, B.G., McGrath, S.P., Scheib, A.J., Breward, N., Cave, M., Lister, T.R., Ingham, M., Gowing, C., Carter, S. (2012). The advanced soil geochemical atlas of England and Wales. British Geological Survey, Nottingham, UK, p. 227.

Rieuwerts, J.S., Ashmore, M.R., Farago, M.E., Thornton, I. (2006) The influence of soil characteristics on the extractability of Cd, Pb and Zn in upland and moorland soils. *Sci. Total Environ.* **366**, 864-875.

Rigol, A., Roig, M., Vidal, M., Rauret, G. (1999) Sequential extractions for the study of radiocesium and radiostrontium dynamics in mineral and organic soils from Western Europe and Chernobyl areas. *Environ. Sci. Technol.* **33**, 887-895.

Rodrigues, S.M., Henriques, B., da Silva, E.F., Pereira, M.E., Duarte, A.C., Romkens, P. (2010) Evaluation of an approach for the characterization of reactive and available pools of twenty potentially toxic elements in soils: Part I - The role of key soil properties in the variation of contaminants' reactivity. *Chemosphere* **81**, 1549-1559.

Rohl, B.M. (1996) Lead isotope data from the Isotrace Laboratory, Oxford: Archaeometry data base 2, galena from Britain and Ireland. *Archaeometry* **38**, 165-180.

Romkens, P.F., Bril, J., Salomons, W. (1996) Interaction between Ca^{2+} and dissolved organic carbon: Implications for metal mobilization. *Appl. Geochem.* **11**, 109-115.

Rosenbaum, M.S., McMillan, A.A., Powell, J.H., Cooper, A.H., Culshaw, M.G., Northmore, K.J. (2003) Classification of artificial (man-made) ground. *Engineering Geology* **69**, 399-409.

Rowell, D.L. (1994). *Soil science : methods and applications*. Longman Scientific & Technical, Harlow.

Ruby, M.V., Davis, A., Schoof, R., Eberle, S., Sellstone, C.M. (1996) Estimation of lead and arsenic bioavailability using a physiologically based extraction test. *Environ. Sci. Technol.* **30**, 422-430.

Sahu, S., Banerjee, D.K. (1990) Complexation Properties of Typical Soil and Peat Humic Acids with Copper(II) and Cadmium(II). *Int. J. Environ. Anal. Chem.* **42**, 35-44.

Salam, A.K., Helmke, P.A. (1998) The pH dependence of free ionic activities and total dissolved concentrations of copper and cadmium in soil solution. *Geoderma* **83**, 281-291.

Sarret, G., Balesdent, J., Bouziri, L., Garnier, J.M., Marcus, M.A., Geoffroy, N., Panfili, F., Manceau, A. (2004) Zn speciation in the organic horizon of a contaminated soil by micro-x-ray fluorescence, micro- and powder-EXAFS spectroscopy, and isotopic dilution. *Environ. Sci. Technol.* **38**, 2792-2801.

Sauve, S., McBride, M.B., Hendershot, W.H. (1997) Speciation of lead in contaminated soils. *Environ. Pollut.* **98**, 149-155.

Sauve, S., Norvell, W.A., McBride, M., Hendershot, W. (2000) Speciation and complexation of cadmium in extracted soil solutions. *Environ. Sci. Technol.* **34**, 291-296.

Sawhney, B.L. (1972) Selective sorption and fixation of cations by clay-minerals - review. *Clays and Clay Minerals* **20**, 93-&.

Schmitt, D., Muller, M.B., Frimmel, F.H. (2001) Metal distribution in different size fractions of natural organic matter. *Acta Hydrochim. Hydrobiol.* **28**, 400-410.

Schnitzer, M., Khan, S.U. (1972). *Humic substances in the environment*. M. Dekker, New York.

Schramel, O., Michalke, B., Kettrup, A. (2000) Study of the copper distribution in contaminated soils of hop fields by single and sequential extraction procedures. *Sci. Total Environ.* **263**, 11-22.

Schroder, T.J., Hiemstra, T., Vink, J.P.M., van der Zee, S. (2005) Modeling of the solid-solution partitioning of heavy metals and arsenic in embanked flood plain soils of the rivers Rhine and Meuse. *Environ. Sci. Technol.* **39**, 7176-7184.

Schulin, R., Johnson, A., Frossard, E. (2010). Trace Element-Deficient Soils, in: Hooda, P.S. (Ed.), *Trace elements in soils*. Wiley, Chichester, pp. 175-197.

Shetaya, W.H., Young, S.D., Watts, M.J., Ander, E.L., Bailey, E.H. (2012) Iodine dynamics in soils. *Geochimica Et Cosmochimica Acta* **77**, 457-473.

Shi, Z.Q., Di Toro, D.M., Allen, H.E., Sparks, D.L. (2008) A WHAM-based kinetics model for Zn adsorption and desorption to soils. *Environ. Sci. Technol.* **42**, 5630-5636.

Shirahata, H., Elias, R.W., Patterson, C.C., Koide, M. (1980) Chronological variations in concentrations and isotopic compositions of anthropogenic atmospheric lead in sediments of a remote subalpine pond. *Geochimica Et Cosmochimica Acta* **44**, 149-162.

Shuman, L.M. (1991). Chemical forms of micronutrients in soils, in: Mortvedt, J.J., F.R. Cox, L.M., Shuman, R.M., Welch (Eds.), *Micronutrients in agriculture- Second Edition*. Soil Science Society of Americ, Seiten, US, pp. 113-144.

Sinaj, S., Dubois, A., Frossard, E. (2004) Soil isotopically exchangeable zinc: A comparison between E and L values. *Plant and Soil* **261**, 17-28.

Sinaj, S., Machler, F., Frossard, E. (1999) Assessment of isotopically exchangeable zinc in polluted and nonpolluted soils. *Soil Sci. Soc. Am. J.* **63**, 1618-1625.

Sinitsyn, V.A., Aja, S.U., Kulik, D.A., Wood, S.A. (2000) Acid-base surface chemistry and sorption of some lanthanides on K^+-saturated marblehead illite: I. Results of an experimental investigation. *Geochimica Et Cosmochimica Acta* **64**, 185-194.

Smith, E.J., Rey-Castro, C., Longworth, H., Lofts, S., Lawlor, A.J., Tipping, E. (2004) Cation binding by acid-washed peat, interpreted with humic ion-binding model VI-FD. *Eur. J. Soil Sci.* **55**, 433-447.

Smolders, E., Brans, K., Foldi, A., Merckx, R. (1999) Cadmium fixation in soils measured by isotopic dilution. *Soil Sci. Soc. Am. J.* **63**, 78-85.

Sposito, G. (2008). *The chemistry of soils,* 2nd ed. Oxford University Press, New York ; Oxford.

Stacey, S., Merrington, G., McLaughlin, M.J. (2001) The effect of aging biosolids on the availability of cadmium and zinc in soil. *Eur. J. Soil Sci.* **52**, 313-321.

Stalikas, C.D., Pilidis, G.A., Tzouwara-Karayanni, S.M. (1999) Use of a sequential extraction scheme with data normalisation to assess the metal distribution in agricultural soils irrigated by lake water. *Sci. Total Environ.* **236**, 7-18.

Stenberg, A., Andren, H., Malinovsky, D., Engstrom, E., Rodushkin, I., Baxter, D.C. (2004) Isotopic variations of Zn in biological materials. *Anal. Chem.* **76**, 3971-3978.

Sterckeman, T., Carignan, J., Srayeddin, I., Baize, D., Cloquet, C. (2009) Availability of soil cadmium using stable and radioactive isotope dilution. *Geoderma* **153**, 372-378.

Sterckeman, T., Douay, F., Proix, N., Fourrier, H. (2000) Vertical distribution of Cd, Pb and Zn in soils near smelters in the North of France. *Environ. Pollut.* **107**, 377-389.

Streck, T., Richter, J. (1997a) Heavy metal displacement in a sandy soil at the field scale .1. Measurements and parameterization of sorption. *J. Environ. Qual.* **26**, 49-56.

Streck, T., Richter, J. (1997b) Heavy metal displacement in a sandy soil at the field scale .2. Modeling. *J. Environ. Qual.* **26**, 56-62.

Tack, F.M.G. (2010). Trace Elements: General Soil Chemistry, Principles and Processes, in: Hooda, P.S. (Ed.), *Trace elements in soils*. Wiley, Chichester, pp. 9-39.

Tessier, A., Campbell, P.G.C., Bisson, M. (1979) Sequential extraction procedure for the speciation of particulate trace metals. *Anal. Chem.* **51**, 844-851.

Teutsch, N., Erel, Y., Halicz, L., Banin, A. (2001) Distribution of natural and anthropogenic lead in Mediterranean soils. *Geochimica Et Cosmochimica Acta* **65**, 2853-2864.

Thakali, S., Allen, H.E., Di Toro, D.M., Ponizovsky, A.A., Rooney, C.P., Zhao, F.J., McGrath, S.P. (2006) A Terrestrial Biotic Ligand Model. 1. Development and application to Cu and Ni toxicities to barley root elongation in soils. *Environ. Sci. Technol.* **40**, 7085-7093.

Thornton, I., Farago, M.E., Thums, C.R., Parrish, R.R., McGill, R.A.R., Breward, N., Fortey, N.J., Simpson, P., Young, S.D., Tye, A.M., Crout, N.M.J., Hough, R.L., Watt, J. (2008) Urban geochemistry: research strategies to assist risk assessment and remediation of brownfield sites in urban areas. *Environmental Geochemistry and Health* **30**, 565-576.

Thums, C.R., Farago, M.E., Thornton, I. (2008) Bioavailability of trace metals in brownfield soils in an urban area in the UK. *Environmental Geochemistry and Health* **30**, 549-563.

Tiller, K.G., Honeyset.Jl, Devries, M.P.C. (1972a) Soil zinc and its uptake by plants II. soil chemistry in relation to prediction of availability. *Aust. J. Soil Res.* **10**, 165-182.

Tiller, K.G., Honeyset.Jl, Devries, M.P.C. (1972b) Soil zinc and its uptake by plants. I. isotopic exchagne equilibria and application of tracer techniques. *Aust. J. Soil Res.* **10**, 151-164.

Tipping, E. (1994) WHAM - a chemical equilibrium model and computer code for waters, sediments, and soils incorporating a discrete site/electrostatic model of ion-binding by humic substances. *Computers & Geosciences* **20**, 973-1023.

Tipping, E. (1998) Humic ion-binding model VI: An improved description of the interactions of protons and metal ions with humic substances. *Aquat. Geochem.* **4**, 3-48.

Tipping, E. (2002). *Cation binding by humic substances*. Cambridge, Cambridge.

Tipping, E., Lawlor, A.J., Lofts, S., Shotbolt, L. (2006) Simulating the long-term chemistry of an upland UK catchment: Heavy metals. *Environ. Pollut.* **141**, 139-150.

Tipping, E., Lofts, S., Sonke, J.E. (2011) Humic Ion-Binding Model VII: a revised parameterisation of cation-binding by humic substances. *Environ. Chem.* **8**, 225-235.

Tipping, E., Rieuwerts, J., Pan, G., Ashmore, M.R., Lofts, S., Hill, M.T.R., Farago, M.E., Thornton, I. (2003) The solid-solution partitioning of heavy metals (Cu, Zn, Cd, Pb) in upland soils of England and Wales. *Environ. Pollut.* **125**, 213-225.

Tongtavee, N., Shiowatana, J., McLaren, R.G., Gray, C.W. (2005) Assessment of lead availability in contaminated soil using isotope dilution techniques. *Sci. Total Environ.* **348**, 244-256.

Trivedi, P., Axe, L. (2001) Predicting divalent metal sorption to hydrous Al, Fe, and Mn oxides. *Environ. Sci. Technol.* **35**, 1779-1784.

Tye, A.M., Young, S., Crout, N.M.J., Zhang, H., Preston, S., Zhao, F.J., McGrath, S.P. (2004) Speciation and solubility of Cu, Ni and Pb in contaminated soils. *Eur. J. Soil Sci.* **55**, 579-590.

Tye, A.M., Young, S.D., Crout, N.M.J., Zhang, H., Preston, S., Barbosa-Jefferson, V.L., Davison, W., McGrath, S.P., Paton, G.I., Kilham, K., Resende, L. (2003) Predicting the activity of Cd^{2+} and Zn^{2+} in soil pore water from the radio-labile metal fraction. *Geochimica Et Cosmochimica Acta* **67**, 375-385.

Ure, A.M. (1996) Single extraction schemes for soil analysis and related applications. *Sci. Total Environ.* **178**, 3-10.

Ure, A.M., Quevauviller, P., Muntau, H., Griepink, B. (1993) Speciation of heavy metals in soils and sediment - an account of the improvement and harmonization of extraction techniques undertaken under the auspices of the BCR of the commission of the European communities. *Int. J. Environ. Anal. Chem.* **51**, 135-151.

Vane, C.H., Chenery, S.R., Harrison, I., Kim, A.W., Moss-Hayes, V., Jones, D.G. (2011) Chemical signatures of the Anthropocene in the Clyde estuary, UK: sediment-hosted Pb, Pb-207/206, total petroleum hydrocarbon, polyaromatic hydrocarbon and polychlorinated biphenyl pollution records. *Philosophical Transactions of the Royal Society a-Mathematical Physical and Engineering Sciences* **369**, 1085-1111.

Watmough, S.A., Hutchinson, T.C. (2004) The quantification and distribution of pollution Pb at a woodland in rural south central Ontario, Canada. *Environ. Pollut.* **128**, 419-428.

Weckwerth, G. (2001) Verification of traffic emitted aerosol components in the ambient air of Cologne (Germany). *Atmospheric Environment* **35**, 5525-5536.

Wei, S., Xiang, W. (2013) Adsorption removal of Pb(II) from aqueous solution by fulvic acid-coated ferrihydrite. *Journal of Food Agriculture & Environment* **11**, 1376-1380.

Welp, G., Brummer, G.W. (1999) Adsorption and solubility of ten metals in soil samples of different composition. *J. Plant Nutr. Soil Sci.-Z. Pflanzenernahr. Bodenkd.* **162**, 155-161.

Weltje, L. (1998) Mixture toxicity and tissue interactions of Cd, Cu, Pb and Zn in earthworms (Oligochaeta) in laboratory and field soils: A critical evaluation of data. *Chemosphere* **36**, 2643-2660.

Weng, L.P., Temminghoff, E.J.M., Lofts, S., Tipping, E., Van Riemsdijk, W.H. (2002) Complexation with dissolved organic matter and solubility control of heavy metals in a sandy soil. *Environ. Sci. Technol.* **36**, 4804-4810.

Weng, L.P., Temminghoff, E.J.M., van Riemsdijk, W.H. (2001) Contribution of individual sorhents to the control of heavy metal activity in sandy soil. *Environ. Sci. Technol.* **35**, 4436-4443.

Weng, L.P., Van Riemsdijk, W.H., Hiemstra, T. (2009) Effects of Fulvic and Humic Acids on Arsenate Adsorption to Goethite: Experiments and Modeling. *Environ. Sci. Technol.* **43**, 7198-7204.

Weng, L.P., Wolthoorn, A., Lexmond, T.M., Temminghoff, E.J.M., Van Riemsdijk, W.H. (2004) Understanding the effects of soil characteristics on phytotoxicity and bioavailability of nickel using speciation models. *Environ. Sci. Technol.* **38**, 156-162.

Wixson, B.G., Davies, B.E. (1994) Guideline for lead in soil proposal of the society for environmental geochemistry and health. *Environ. Sci. Technol.* **28**, A26-A31.

Xu, Y., Boonfueng, T., Axe, L., Maeng, S., Tyson, T. (2006) Surface complexation of Pb(II) on amorphous iron oxide and manganese oxide: Spectroscopic and time studies. *Journal of Colloid and Interface Science* **299**, 28-40.

Young, S., Crout, N., Hutchinson, J., Tye, A., Tandy, S., Nakhone, L. (2007). Techniques for measuring attenuation: Isotopic dilution methods, in: Hamon, R., McLaughlin, M.J., Lombi, E., SETAC (Society) (Eds.), *Natural attenuation of trace element availability in soils*. CRC, Boca Raton, Fla. ; London, p. 236 p.

Young, S.D., Tye, A., Carstensen, A., Resende, L., Crout, N. (2000) Methods for determining labile cadmium and zinc in soil. *Eur. J. Soil Sci.* **51**, 129-136.

Young, S.D., Zhang, H., Tye, A.M., Maxted, A., Thums, C., Thornton, I. (2005) Characterizing the availability of metals in contaminated soils. I. The solid phase: sequential extraction and isotopic dilution. *Soil Use and Management* **21**, 450-458.

Zhang, H., Young, S.D. (2005) Characterizing the availability of metals in contaminated soils. II. The soil solution. *Soil Use and Management* **21**, 459-467.

Zhang, H., Zhao, F.J., Sun, B., Davison, W., McGrath, S.P. (2001) A new method to measure effective soil solution concentration predicts copper availability to plants. *Environ. Sci. Technol.* **35**, 2602-2607.

Zimdahl, R.L., Skogerboe, R.K. (1977) Behavior of lead in soil. *Environ. Sci. Technol.* **11**, 1202-1207.

Zwolsman, J.J.G., Berger, G.W., Vaneck, G.T.M. (1993) Sediment accumulation rates, historical input, postdepositional mobility and retention of major elements and trace metals in salt marsh sediments of the Scheldt estuary, SW Netherlands. *Mar. Chem.* **44**, 73-94.