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BIOREMEDIATION OF SOILS POLLUTED BY HEAVY METALS USING ORGANIC ACIDS

by

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A thesis submitted to the Faculty of Graduate
Studies and Research in partial fulfillment
of the requirements for the degree of
DOCTOR OF PHILOSOPHY

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ABSTRACT

Weak organic acids and/or their salts were tested as soil washing or flushing agents for the ex- or in-situ remediation of soils polluted by heavy metals. Three soils naturally contaminated with heavy metals were used for the test.

The three soils were characterized as a clay loam, loam and sandy clay loam. Their organic matter, pH, saturated hydraulic conductivity, cation exchange capacity, particle density and heavy metal contents were also characterized. The different retention forms of heavy metals in all 3 soils were studied by sequential extraction. The clay loam was contaminated with Cr, Hg, Mn and Pb while the loam and sandy clay loam were contaminated with Cd, Pb, Cu and Zn. Weak organic acids and/or their salts and chelating agents (EDTA and DTPA) were used at different pH, levels of concentration and leaching time in batch experiments to establish optimum conditions for maximum removal of heavy metals from the three soils. Citrate and tartarate were found to be quite effective in leaching heavy metals from these soils. The rate of leaching of heavy metals from soils with citrate, tartarate and EDTA was modeled using two-reaction model at a constant pH and temperature.

Three contaminated soils of different textures were flushed in a column at optimum pH with a salt of weak organic acids, namely, citrate, tartarate, citrate+oxalate or a chelating agent such as EDTA and DTPA. The citrate and tartarate (ammonium salts) were found to be quite effective in removing heavy metals from the three contaminated soils while leaching little macronutrients and improving the soil's structure. An in-situ soil remediation simulation was also successfully tested using the sandy clay loam at large scale level in a tub (plastic container) using citrate as a flushing liquid. EDTA and DTPA were effective in removing the heavy metals except for Hg, but these strong chelating agents extracted important quantities of macronutrients from the soil. These chelating agents are also known to pollute the soil by being adsorbed on the soil particles.

A bioremediation process was developed using the fungus *Aspergillus niger* to produce weak organic acids (mainly citrate and partly oxalate depending on pH) for

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the leaching of heavy metals from contaminated soils. The fungus was cultivated on the surface of the three contaminated soils for 15 days at 30 °C and a pH $\leq$ 4 to enhance the production of citric acid rather than oxalic acid which hinders Pb leaching. By extrapolating the result, the three contaminated soils were expected to be sufficiently remediated to meet the A category (Quebec clean up criteria for cleaning soils contaminated by heavy metals) after 20 to 25 days of leaching using this technique.

Finally, the leachate collected following the soil remediation using weak organic acids and/or their salts, EDTA and DTPA was treated effectively using granular activated carbon.

RESUMÉ

Les acides organiques et/ou leur sel furent testées comme agent de lessivage des sols contaminés par les métaux lourds. Trois sols, contaminés naturellement par le lexiviat d' un site d' enfouissement sanitaire japonais, furent utilisés pour les essais.

Les trois sols expérimentaux furent caractérisés. Leur texture était d'argile limoneuse, de limon et de sable argilo-limoneux, respectivement. La matière organique, le pH, la conductivité hydraulique saturée, la capacité d'échange cationique, la densité des particules et leur teneur en métaux lourds de ces trois sols furent déterminés. Les différentes formes de rétention des métaux lourds, dans ces trois sols, furent aussi déterminées par extraction séquentielle. L'argile limoneuse était contaminés de Cr, Mn, Hg et Pb alors que le limon et le sable argilo-limoneux étaient contaminés de Cd, Pb, Cu et Zn. Les acides organiques et/ou leur sel et deux agents chélateurs (EDTA et DTPA) furent testés a différents pH, différentes concentrations d'agent de lessivage et pendant différentes périodes de lessivage. Ces essais ont établi les modalités optimumes de performance des acides organiques testées. Le citrate et le tartarate furent les deux acides organiques qui ont le mieux performé pendant les essais. Le taux de lessivage des métaux lourds avec le citrate,le tartarate et le EDTA fut modèlisé en utilisant un modèle à deux équations et en assumant un pH constant.

Les trois sols de texture différente furent lessivés dans des colonnes sous un pH optimum et avec les acides organiques les plus performantes, soit le citrate, le tartarate et le citrate-tartarate ainsi que deux agents chélateurs, le EDTA et le DTPA. Le citrate et le tartarate (sel d' ammonium) furent très efficaces et ont lessivé la majeur partie des métaux lourds pour les trois sols expérimentaux. Ces acides ont aussi lessivé peut de macro-éléments tels le Ca et le Mg, tout en améliorant la structure des sols contaminés. Un essai insitu fut réalisé dans un bassin et avec l'acide citrate comme agent de lessivage. EDTA et DTPA lessivaient avec efficacité tous les métaux lourds sauf le Hg. Mais, ces agents chélateurs lessivaient aussi des quantités importantes de macro-éléments. Ces agents chélateurs sont aussi des contaminants de sol puisqu'ils sont absorbés par les particules de sol et sont dégradés très lentement.

RESUMÉ SUITE

Un procédé de bio-rémédiation fut développé en utilisant le champigon *Aspergillus niger* comme source d'acide, et surtout de citrate avec une petite quantité d'oxalate. Le champignon produit du citrate à un pH de 4.0 et plutôt de l'oxalate à un pH inférieur à 4.0. Le champignon était cultivé à la surface des trois sols expérimentaux pendant 15 jours à 30C et à un pH au-dessus de 4. Avec un pH au-dessus de 4.0, le champignon produisait plutôt du citrate plus performant comme agent de lessivage que l'oxalate. De plus, l'oxalate lessive très peu de Pb. L' extrapolation des résultats de lessivage indiquait que le champignon pouvait améliorer la teneur en métaux lourds des trois sols contaminés de facon à rencontrer le plus haut critère québécois de rémédiation, soit le critère A, après 25 jours de traitement.

Enfin, le lixiviat lessivé des sols fut traité effectivement avec du charbon activé granulaire.

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LIST OF SYMBOLS AND ABBREVIATIONS

A°	Angstrom (1 A° = 10^{-10} m)
$BaSO_4$	Barium sulfate
$BaCl_2$	Barium chloride
Cd	Cadmium
Cu	Copper
Cr	Chromium
$CaCO_3$	Calcium carbonate
$CaCl_2$	Calcium chloride
C_e	Equilibrium concentration of metal chelates (mg/l)
CEC [cmol(+)/kg]	Cation exchange capacity (centimole (+) per kilogram of soil)
$Ca(NO_3)_2$	Calcium nitrate
$CuSO_4$	Copper sulfate
C_{ka}, C_{kb}	Heavy metal concentrations in the soil reacted according to reaction (1) and (2) in mg/kg
C_kt, C_k0	Heavy metal concentration in soil at time t and at time t = 0 in mg/kg
C_ℓ	Heavy metal concentration in leachate in mg/l (milligram per liter)
DTPA	Diethylenetriaminepentaacetic acid
d	Residual heavy metal concentration in the soil
EC (mS/cm)	Electrical conductivity (millisiemens per centimeter)
EDTA	Ethylenediaminetetraacetic acid
FCS	Ferric chloride solution
GAC	Granular activated carbon
GAA	Granular activated alumina
HAc	Acetic acid
HCl	Hydrochloric acid
$HClO_4$	Perchloric acid

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H_2S	Hydrogen sulfide
H_2O_2	Hydrogen peroxide
HNO_3	Nitric acid
H_2SO_4	Sulfuric acid
H_3PO_4	Phosphoric acid
$HgSO_4$	Mercury sulfate
Hg	Mercury
ICP-AES	Inductively Coupled Plasma-Atomic Emission Spectroscopy
k_s (cm/s)	Saturated hydraulic conductivity (centimeter per second)
k_1, k_2, k_3	Leaching or reaction rate constants [k_1 (per hour), k_2 (per hour) and k_3 (kilogram liquid per kilogram of soil per hour)]
k_{ad}	Adsorption rate constant (min^{-1})
$KMnO_4$	Potassium permanganate
$K_2S_2O_8$	Potassium persulfate
Mn	Manganese
M_{k0}, $M_{\ell 0}$	Mass of soil and extraction liquid at time t =0
$M_k(t)$, $M_\ell(t)$	Mass of soil and extraction liquid at time t
$MgCl_2$	Magnesium chloride
m_1	Quasi-equilibrium constant
$MgCO_3$	Magnesium carbonate
NaOH	Sodium hydroxide
$NaNO_3$	Sodium nitrate
NH_4Cl	Ammonium chloride
NH_4-OAc/NH_4Ac	Ammonium acetate
$NH_2OH.HCl$	Hyroxylamine hydrochloride
NaCl	Sodium chloride
$PbSO_4$	Lead sulfate
Pb	Lead

LIST OF SYMBOLS AND ABBREVIATIONS CONTD.

Q°, b	Langmuir constants related to the capacity (mg/g) and energy of adsorption (l/mg), respectively
q_e, q	Amount of metal chelates (mg/l) adsorbed at equilibrium and at time t (min)
$-R_a$, $-R_b$ (mg/kg/h)	Rate of leaching according to reaction (6.1) and (6.2)
t	Time (hour)
Zn	Zinc
α_0	Parameter for a slow extractable metals

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CHAPTER 1

INTRODUCTION

1.1 General Introduction

Heavy metal pollution of soils has become one of the most important environmental problems in developed as well as developing countries. There is a growing concern about soil pollution by heavy metals in Canada, USA, Japan and Europe mainly due to the metal mining and smelting industry (Asami 1988). Soils also receive large quantities of heavy metals from a variety of industrial wastes and municipal sewage sludge. In most cases, soil polluted by heavy metals such as Cd, Zn, Pb, Cu, As and Hg is disposed in a secured landfill. But those systems are always accompanied by a risk of secondary pollution.

One of the negative environmental impacts arising from the heavy metal pollution of soil is surface and groundwater contamination due to rainwater run off or its percolation through the soil, slowly extracting metals which eventually reach aquifers. Acidification of the soil by acid rains accelerates this process (Matschullat et al. 1992). In Quebec, about 50% of groundwater contamination sites result from landfills built in sand dunes without liners or from the cracking of lining materials such as bentonite (NRCC 1987). The metals contained in the leachate may be highly toxic to humans, animals and aquatic life. Conventional remediation techniques are costly when the soil is polluted by heavy metals because the metals are tightly bound to various soil fractions. It has been estimated that the clean-up cost for one hectare over a depth of one meter ranges between $ 0.6 to 3 million depending of the pollution load on the soil (Moffat 1995).

Surface water and sediment pollution have become serious problems in recent years. This pollution results from either discharge of industrial wastes, run off from contaminated sites, or metal deposition through emission from metal smelter, resulting from metal mines, metal smelter, refineries, as well as metal processing industries. For example, the sediments from the Lachine Canal (Montreal) were polluted by heavy metals and organic pollutants (Tecsult Inc. 1993). Estimates of the costs of extracting and treating the contaminated Lachine Canal sediments (122 m^3) vary between Can $ 30 to 43 million.

Superfund (1980) required the US Environmental Protection Agency to prepare a National Priority List (NPL) for the remediation of polluted sites to prevent groundwater pollution, which is a major source of water supply in USA (about 50%). At that time, the number of sites reached 1250 and it is still growing. Heavy metal pollution is found on 18% of the sites, whereas organic and heavy metal contamination is found to occur at 37% of the sites. The rest of the sites (45%), are contaminated with organic chemicals. The estimated cleanup cost of such contaminated sites is about US $ 150 billion. The estimated total cleanup cost including other sites runs close to one tenth of the gross national product (GNP) of the nation (Acra 1995).

In third world countries, less than 5% of the solid waste management budget is typically allocated to waste disposal, which may create problems such as soil and groundwater pollution as well as surface water and sediment pollution. In contrast, industrialized countries contribute 25-30% of such budgets to waste disposal (Bartone and Bernstein 1992). Wastes generated in industrialized countries are more toxic and produced in greater quantity than in developing countries. Therefore, more effective control systems have been implemented in industrialized countries.

Soils are complex porous aggregates usually possessing a high metal-binding capacity, primarily due to their small, high surface active components such as clay particles, organic materials and hydrous metal oxides. Heavy metals may be retained in the soil as exchangeable metals, carbonates, hydroxides, Fe/Mn-oxides, or may be bound to organic matter. In most cases, heavy metals are retained in the upper horizon of soils (< 0.5 m), depending on local environmental conditions. The scientific and technological challenges are to:

i) understand the interactions between soil and heavy metals,

ii) establish a better scientific basis for deciding:
    - what remediation approach to take at a given site
    - which heavy metal forms in the soil are removable

iii) develop new cleanup technologies or enhance existing ones

iv) develop a bioremediation process for heavy metals which is cost effective and environmentally friendly.

At a particular site, it is therefore important to know how the contaminants are held in the soil before designing a remediation process. Sequential extraction is thought to be the most suitable method to study how heavy metals are retained in different components of the soil and to evaluate the relative binding strength and hence the solubility, mobility and bioavailability of heavy metals in contaminated soils (Liu and Dezfulian 1995).

1.2 Available Remediation Technology

Remediation of polluted soils can be carried out either by ex-situ or in-situ leaching and in-situ immobilization (fixation) of heavy metals. Some soil remediation options are illustrated in Figure 1.1.

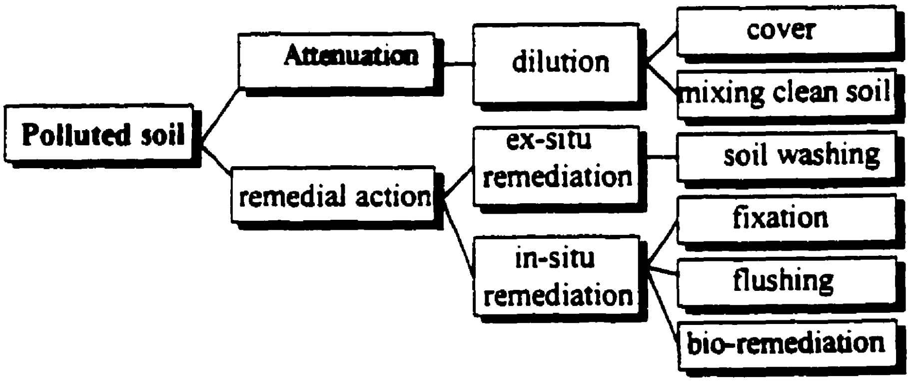

Figure 1.1 Options for remediation of polluted soils

1.2.1 Contaminated soil dilution and cap/cover

This method of treating soils polluted by heavy metals consists in covering the soil with unpolluted clay soils or to dilute them by mixing with unpolluted soils until the concentration of heavy metals is lowered to an acceptable limit.

1.2.2 Ex-situ remediation

The soil is first excavated (dug out) from the site and then submitted to soil washing; one of the most suitable processes for removing heavy metals from the soil. Heavy metals are mainly retained within few cms of surface soil depending on the soil condition, which can be excavated and treated by soil washing process to avoid in-situ remediation through drainage system. The washing solution may contain acids, bases, or chelating agents (Benschoten et al. 1994). Generally, a soil washing process consists of four basic steps:

i) pretreatment of the contaminated soil (sieving, hydrocyclonation),
ii) extraction of heavy metals from the soil by washing,
iii) separation of the cleaned soil particles from the liquid phase,
iv) cleaning and recycling of the extracting (washing) agent.

The contaminants in the extraction solution can be treated by conventional methods (Cushnie 1985, Tels 1987) and the extractant may be reused. This washing process has the disadvantage of using minerai acids (HCl, HNO_3) which physically, chemically and biologically alter soil properties.

1.2.3 In-situ immobilization

In-situ immobilization or fixation of heavy metals in soil is another remediation process. Different immobilization agents (such as chelate ion exchange resins, portland cement/silicate, natural materials-clays, molecular sieves, greensand and other additives like hydrated lime, silylated silica gel, insoluble starch xanthate, ferrous sulfate) can be added to the soil to reduce the mobility of heavy metals (Liu and Dezfulian 1995; Czupyrua et al. 1989). This process may create future problems such as the leaching of heavy metals into the groundwater, as a result of subsequent biological activity or acidification of the soil.

1.2.4 In-situ soil flushing

Soil washing can also be carried out in-situ (without digging out the soil). The flushing (washing) solution for contaminated sites may be the same as for ex-situ remediation. This process is the most appealing because it generally minimizes surface damage, requires a minimal amount of facilities, reduces the potential for human exposure to contaminants, is less expensive and, when effective, reduces or removes contaminants.

However, this remediation process is limited to permeable soils and cannot be applied easily to clay soils.

1.2.5 In-situ or ex-situ bioremediation

An increase in heavy metal mobility can result from microbial oxidation or metal reduction. This process can be exploited for remedial purposes. For example, an insoluble metal sulfide may be oxidized to form a soluble metal sulfate. The solubilized form of the metal when released from the matrix, can be collected for subsequent treatment. Using this type of biological activity to increase the mobility would be analogous to soil washing or a chemical leaching processes (Wildeman *et al.* 1994). This process is only suitable where all metals are present in the form of sulfide minerals; like in the case of metal mining, under anaerobic conditions such as sewage sludge or marine sediments. Subsequently, the process may create another problem such as the production of a strong acid (H_2SO_4) which will disturb soil properties.

The microbial reduction of sulfate under reduced conditions produces insoluble metal sulfide precipitates, thereby reducing the mobility of heavy metals. It is impossible to remove all metals from the soil by using such microbial processes because some heavy metals will form insoluble metal sulfates like $PbSO_4$ and $HgSO_4$. Increasingly, bioremediation is considered to meet the environmental requirements such as reduction of contaminants to permissible level. This process is particularly useful for wastewater treatment and is often included in remediation either as the sole technique or in combination with other processes such as soil washing and chemical extraction.

1.3 Standards

Soils cleaned by means of a specific remediation technology, should meet the following standard (Table 1.1). Washed soils are considered to be sufficiently clean if the heavy metal concentrations have been reduced to meet A specifications (Ministry of Environment and Forest, Quebec(MEFQ) 1994). Soil can be disposed on land when it meets A level requirements.

Some organic acids such as acetic, citric and oxalic acids have been tested at a single pH value to treat soils artificially contaminated with heavy metals (Farrah and Pickering 1978). The advantages associated with the use of such organic acids are:

i) low cost
ii) biodegradability, therefore environment friendly
iii) less destructive to soil structure than mineral acid
iv) more specific to heavy metals and less likely to leach soil macronutrients
v) improve soil properties.

Table 1.1 Standard for heavy metals in soil (mg/kg)

Soil	Cu	Pb	Cr	Ni	Zn	Cd	Hg
A-value	50	50	75	50	100	1.5	0.2
B-value	100	150	250	100	500	5.0	2.0
C-value	500	600	800	500	1500	20.0	10.0

B and C values means action is required (MEFQ 1994).

Similar guidelines were also used in The Netherlands published by the Ministry of Housing, Physical Planning and Environment based on three category of soils A, B and C values (Assink 1986), whereas USEPA standard for soil are based on leachability of metals from the soil.

1.4 Objectives

The objectives of this study are:

1. To characterize soil samples polluted by heavy metals and to measure the relative proportion of heavy metals retained by each soil fraction (exchangeable, oxides and organic matter).

2. To identify the best organic acids and/or their salts used for heavy metal leaching. To establish their capability to chelate heavy metals and form soluble metal complexes which

can be leached from the contaminated soil. Synthetic chelating agents (EDTA and DTPA) were used as bases of comparison.

3. To establish optimum heavy metal leaching at different pH and acid concentrations.
4. To establish the kinetics of such weak organic acids in chelating and leaching heavy metals from contaminated soils using batch and column set ups.
5. To model heavy metal leaching using a two-reaction model, the kinetic data obtained in batch experiments will be utilized.
6. To establish the capability of fungus *(Aspergillus niger)* to produce weak organic acids for in-situ and ex-situ bioremediation by means of a column study.
7. To demonstrate that granular activated carbon can treat the leachate originating from the remediation of soils using weak organic acids.

1.5 Scope of the Study

The remediation method developed applies to in-situ remediation techniques except the clay soils if soil is contaminated to a grater depth by constructing drainage facilities instead of recovery well extraction. For clay soils, ex-situ remediation may be used.

The experimental procedure is limited to three soils from soils (clay loam, loam and sandy clay loam) contaminated by heavy metals over a period of three years. The experiments were carried out in laboratory in Japan. The tests are limited to laboratory batch and column set ups using 3.5 to 8.5 g of contaminated soil as well as a tub set up to simulate field conditions using 7.7 kg of contaminated soil. The experiment tested the following weak organic acids against 2 synthetic organic chelating agents, EDTA and DTPA:

1.	Citric acid and/or its salt	$HOC(CH_2COOH)_2COOH$
2.	Tartaric acid and/or its salt	$HOOCCH(OH)CH(OH)COOH$
3.	Oxalic acid and/or its salt	$(COOH)_2$
4.	Pyruvic acid and/or its salt	$CH_3COCOOH$
5.	Fumaric acid and/or its salt	$HOOCCH{:}CHCOOH$
6.	Itaconic acid and/or its salt	$C_5H_6O_4$

7.	Formic acid and/or its salt	$HCOOH$
8.	Acetic acid and/or its salt	CH_3COOH
9.	Succinic acid and/or its salt	$HOOCCH_2CH_2COOH$
10.	Phthalic acid and/or its salt	$C_6H_4(COO)_2$
11.	Control using water, NaOH and/or HCl.	

The process developed improves soil structure and may enhance the in-situ bioremediation of soils with a low permeability.

A bioremediation process using *Aspergillus niger* was also set up for cleaning these soils in a column experiment. This process is economical and cost effective and limited to sandy clay loam, but also applied for clay loam and loam through a very slow leachate collection system or in ex-situ remediation.

Granular activated carbon (GAC), ferric chloride solution (FCS) and granular activated alumina (GAA) were tested as treatment media for the leachate.

1.6 Contribution to Knowledge

The pollution of soils by heavy metals has become one of the most important environmental problems in industrialized and developing countries. Our knowledge toward clean up of soils contaminated by heavy metals are limited. These limitations are due mainly to the lack of conceptual understanding of the retention form of heavy metals in soils and the lack of environmentally friendly remediation processes for soils to be used for agricultural purposes after cleaning. Laboratory experiments were conducted on three contaminated soils. These soils were characterized for their retention form of heavy metals. These soils were remediated using weak organic acids and/or their salts and chelating agents and the level of soil cleaning was evaluated using Quebec's guidelines for soil contaminated by heavy metals. Based on the results obtained, this research dissertation offers the following major contributions to knowledge:

1. The demonstration of the quantity of heavy metals held under various retention forms in three contaminated soils. The sequential extraction of heavy metals from these soils was found to be a new approach to understand the interactions between different

fractions of soils (such as exchangeable, oxides, organic matter or silicate clay) and heavy metals in order to establish a better scientific basis for which fraction of total heavy metals are easy or difficult to remediate. The retention forms examined were exchangeable, Fe/Mn-oxides, organic matter and silicate clay minerals. This sequential procedure was applied for the first time to soils contaminated by heavy metals for remediation. Based on retention form of heavy metals, a new clean up technologies can be designed.

2. The demonstration that weak organic acids and/or their salts can be used as chelating agents in batch, column or tub studies to form soluble heavy metal complexes and to leach them from contaminated soils. This approach is new to clean the soil and this idea was generated thinking that those organic acids are already present in the soil as metabolites of the organisms and as a result humic acid is formed by the interaction of these organic acids. Out of ten organic acids, citrate and tartarate were quite effective to remove heavy metals from soils. This clean up technique meets soil clean up criteria A for Zn, Cr, B for Pb, Cd, Cu and Mn and C for Hg in 12 h of washing or flushing with ammonium citrate and tartarate solutions. With the further washing or flushing, the soils could reach the category A for all other metals.

3. A two-reaction model was tested using the measured kinetic data obtained in batch up to 36 h for heavy metals leaching with citrate, tartarate, EDTA and DTPA washing solutions. The model fits nicely to the kinetic data obtained in batch experiment which was carried out for up to 36 h of leaching time. The model was quite useful to understand the leaching rate of metals in order to design a treatment unit and its cost.

4. The demonstration that weak organic acids can be produced naturally by *Aspergillus niger* and can be produced in sufficient quantities to treat appreciable volumes of heavy metals contaminated soils and sediments. The approach is innovative and for the first time *Aspergillus niger* was used to clean soils contaminated by heavy metals. This technique is very economical and effective to clean soils in order to meet the soil clean up criteria.

5. Demonstrate the technique's less disturbing character. The properties of the soil is less disturbed by the present technique as compared to strong chelating agents such as EDTA

and DTPA as well as strong acids (HCl, HNO_3, H_2SO_4). Citrate leaches 4 to 5 times less macronutrients (Ca, Mg) as compared to EDTA.

6. The demonstration that the leachate from soil remediation using weak organic acids and/or their salts can be successfully treated using granular activated carbon. This is also a new way to treat leachate containing soluble heavy metals organic acids/and or strong chelating agents complexes. The granular activate carbon containing heavy metals can either be recycled or used in ore processing unit.

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CHAPTER 2

LITERATURE REVIEW

2.1 Use of Strong Acids, Chelating Agents and Neutral Salts for the Extraction of Soil Metals

Large amounts of lead (about 80%) were extracted from artificially polluted soil using chelating agents such as diethylenetriaminepentaacetic acid (DTPA) and ethylenediaminetetraacetic acid (EDTA) at a specific pH (Karamanos et al. 1976). Small amounts of soil Pb are exchangeable and the majority is combined with organic matter or present as insoluble precipitates. EDTA and DTPA form extremely stable complexes with heavy metals and are considered to give a measure of Pb organically bound although they extract some adsorbed or exchangeable Pb.

Kaolinite, smectite and corresponding organic materials in clay soils contaminated by Cr, Ag and Cu were remediated with HNO_3, $Ca(NO_3)_2$, EDTA and fulvic acid at several concentrations over 48 h and at room temperature (Flemming et al. 1990). The metal mobilization or resistance to solubilization depends on the physical properties of the organic and clay particle surfaces and on the concentration of the extractant. In general, the order of remobilization was $Cr < Ag < Cu$.

Seventeen different chemical solutions have been tested to evaluate their ability to leach heavy metals (Pb, Zn, Cu, Cd) from artificially contaminated clays such as kaolinite, illite and montmorillonite, at either pH 5 or 7 (Farrah and Pickering 1978). Of the reagents used, only EDTA (0.001 M, pH 7) quantitatively released all four metals from the three clays, while oxalic acid (0.1 M, pH 3.3) totally displaced at least three metals from each clay. Other reagents, such as ammonium oxalate (0.1 M), ammonium nitrate (0.01 M), nitric acid (0.1 M) and sodium citrate (0.01 M) effectively displaced one or more heavy metals from individual clays.

The selective removal of heavy metals and alkaline-earth metals adsorbed on kaolinite particles by four chelating agents such as the nitrilotriacetic acid (NTA), EDTA, ethyleneglycol-bis 2-aminoethylether tetreacetic acid (EGTA) and 1,2-

diaminocyclohexane N,N,N',N-tetraacetic acid (DCyTA), has been investigated (Hong and Pintauro 1996). There was a good selectivity for heavy metal removal by EGTA when Cd, Ca and Mg were adsorbed on kaolinite because only 5-10% of Ca and Mg were leached while > 80% of Cd was leached when the concentration of EGTA was 0.5 μmole. The leaching percentage of Cd and Ca increased to 100 and 40%, respectively, as EGTA concentration doubled and leaching of Ca and Mg increased further as concentration increased. Among the four chelators, NTA had the poorest removal selectivity between Cd and alkaline-earth metals. The chelating ability of DCyTA was excellent but its selectivity between Cd and the alkaline-earth metals was poor. EGTA appears to be the best chelator, based on its binding ability and selectivity for Cd. When the concentration of chelator in solution was insufficient to combine with all metals adsorbed on kaolinite, the order of metal removal was Cd > Cu > Pb (for EGTA), Cd > Pb > Cu (for EDTA and DCyTA) and Cu > Cd > Pb (for NTA).

EDTA and citric acid were used to extract heavy metals such as Cu, Zn, Cd, Ni, Pb and Cr from contaminated soils at a neutral pH (Leidmann et al. 1994). EDTA was able to mobilize up to 90% of Cd, Cu, Pb and Zn except for Cr and Ni (25-45%). Citric acid (0.26 M) could extract from 50 to 80% of Cu, Cd, Zn and Ni, whereas only 30 to 40% of Cr and Pb were extracted at a pH of 7.

When selecting a complexing ligand to mobilize heavy metals from the soil, the following should be considered (Peters and Shem 1992): i) the ligands should be able to form stable complexes with metals over a wide range of pH, ii) the biodegradability of metal-complex should be low to prevent release of metals into the soil pore water, iii) the resulting complexes should have low affinity towards the surface sites or should be non-adsorbable, iv) the ligand should have a low toxicity and a low negative impact on the environment, v) the reagents should be cost effective.

Several researchers have obtained better metal removal results from soils using EDTA as compared to HCl, while others have reported the opposite (Tuin and Tels 1990). Generally, metal extraction efficiencies for EDTA are similar to those obtained for strong acid extractions. Sandy and silty clays artificially polluted by Pb were investigated for the mobility of Pb in the presence of EDTA (Peters and Shem 1992).

Independent of pH between 4 and 12, Pb removal ranged from 58 to 64% for EDTA concentrations of 0.01 to 0.1 M. EDTA was also used for Cd adsorption on soils. EDTA decreases the adsorption of Cd on the soil particle thereby Cd mobilizes from the soil (Lo et al. 1992).

The distribution of heavy metals in contaminated soil has been studied by $CaCl_2$ extraction (Asami 1995). It was found that Cd and Zn are more soluble (45%) in the presence of $CaCl_2$ solution, whereas the solubility of other metals such as Cu and Pb was below 10%. The removal order of metals from soil fractions using $CaCl_2$ and acetic acid was Cd > Zn > Pb > Cu. Approximately 20% of the total Zn and Pb and 10% of the total Cd and Cu were present in the free oxide fractions. In soils and sediments, Hg was mainly bound to organic matter (Hempel et al. 1995) and mobilization often took place by degradation of the humic substance. Most of the Hg found in the first few centimeters of the contact surface was probably fixed on organic matter and clay minerals.

Chromium content in clay loam, sand and sludge samples was extracted in succession with distilled water, 1 M NH_4Cl, 0.1 M $CuSO_4$, 0.3 M ammonium oxalate and citrate-dithionite-bicarbonate (Grove and Ellis 1980). Extraction levels of 45% and 80% were reached with oxalate and dithionite extractants, respectively, in clay loam. The removal of Cr by other extractants was negligible. The removal of Cr from sand was 39 and 75 % by oxalate and dithionite, respectively. The percentage extraction of Cr with oxalate and dithionite was reduced when sands and clay loams were amended with sludge.

2.2 Use of Organic Acids for the Extraction of Metals from Soils

The effect of different concentrations of citric acid (common in freshwater) for the adsorption of mercury on kaolinite at various pH has been studied (Singh et al. 1996). Citric acid had a significant retarding effect on the adsorption process at a pH above 6 for Hg and had a significant positive effect on the adsorption process at a pH of 4. Low-molecular-weight (< 500) organic acids such as oxalic, tartaric, citric, acetic, fumaric and succinic acids are present in the top layer of soils and can form complexes with metals, therefore affecting the metal solubility and speciation (Fox and Comerford 1990). High concentrations of oxalic acid were found in the soil followed by formic, citric and acetic acids. Organic acids such as citric and oxalic acids form more stable complexes with metals than formic and lactic acids.

Biological compounds or organic molecules such as glycine, citric acid, tartaric acid and gluconic acid have chelating properties and can react with heavy metals (Lo et al. 1992). When the heavy metals form complexes with organic acid (humic acid), they form stable chelates and lower the mobility of metals. However, the metal mobility and bioavailability increases when heavy metals combine with other organic acids such as acetic acid.

Several kinds of organic acids such as formic acid, acetic acid, malonic acid, lactic acid and citric acid, were used for the extraction of rare earth metals (Matsuyama et al. 1996). In analytical procedures, acetic acid has been used frequently to replace the more strongly adsorbed cations of heavy metals and to extract the "available" Pb fraction from soils. This reagent can remove slightly higher amounts of Pb than the exchange reagents such as ammonium acetate (Karamanos et al. 1976).

2.3 Remediation Techniques

Soil washing of heavy metals from a contaminated sandy soil was studied to evaluate the effectiveness of conventional soil washing extractants such as HCl, HNO_3, H_2SO_4, EDTA and $NH_2OH.HCl$ (Benschoten et al. 1994). Application of the sequential extraction procedure showed that most of the metals were in the residual fraction implying that they were bound. Thus, soil surface coatings such as sandy soil coated with iron oxide may make metal extraction difficult. In the presence of such coating, hydrochloric acid (HCl) was more effective than HNO_3 or H_2SO_4 and approximately equal in effectiveness to EDTA. The extractant, hyroxylamine hydrochloride ($NH_2OH.HCl$) was tested to determine if reductive dissolution of iron oxides might also release other metals, but only small amounts were released. Soil washing using 2.0 M HCl for 24 h was successful in removing Pb, Zn and Cu. The kinetics were important in the washing processes. For a 2.0 M HCl, the removal of metals was increased with washing time. Soil exposed to 2.0 M HCl for 24 h in batch tests, resulted in 91, 31 and

45% removal of Pb, Zn and Cu, respectively while As and Hg were removed at 66 and 27%, respectively. Metal release was greater for HCl than with HNO_3 extraction, possibly due to the formation of soluble metal-chloride complexes (Tuin and Tels 1990). A disadvantage of using strong acids as extractants is the adverse effects on the soil's physical and chemical structure, such as mineral dissolution.

Soil washing or flushing in columns of a sandy loam artificially polluted by Pb was investigated using solutions of HCl, EDTA and $CaCl_2$ (Reed et al. 1996). Lead removal efficiencies for HCl, EDTA and $CaCl_2$, were 85%,100% and 78%, respectively. The HCl at final soil pH ($\approx$ 1) increases Pb mobility but decreases soil productivity because of the mineral dissolution. Final soil pH for EDTA and $CaCl_2$ ranged between 4.9 and 5.2. Several washing chemicals such as HCl, EDTA, DTPA and chlorine were applied at various concentrations to a soil artificially contaminated with Zn (Davis and Singh 1995). The effects of ionic strength, flow rate and form of Zn contamination were also determined. The most efficient washing occurred using organic chelating agents. Reducing flow rate had little effects and washing efficiency was found to be strongly dependent on the retention form of Zn in the contaminated soil.

Remediation of soils polluted by heavy metals has been studied by the counter-current extraction process (Masscheleyn et al. 1996). The process involved decarbonation (removal of carbonates bound metals), acid solubilization of metal, washing and liming. The removal of Cu and Pb was limited to approximately 15%, whereas Cd and Zn were removed at levels of 85 and 25%, respectively.

In Japan, agricultural soils have been polluted by heavy metals such as Cd, Cu and As mainly because of acid mine drainage carried away by flooding to the lower portion of river basins. Some sites extending over as many as 7140 ha were polluted this way (Tokunaga 1996). Cadmium was the common pollutant while other contaminants such as Cu and As were much less frequent. In most cases, these sites were located in volcanic regions or downstream from a mining area. These soils were remediated by soil-covering or by the dilution method. The polluted soils were covered with a layer of non-polluted soil (20-25 cm thick) for safe crop production. The dilution method involved mixing polluted soil with unpolluted soil to reduce the concentration of heavy metals to acceptable levels. Cadmium polluted soils were covered with unpolluted soils so that the rice grown should not contain more than 1 mg Cd/kg of rice. Cadmium in agricultural land was the main reason for the itai-itai disease, a world famous disease due to soil pollution. Urban soil pollution in Japan is mainly due to inorganic pollutants (62%) such as Pb, Cr, Hg, Cd, As, CN and Zn. Technologies used to treat these polluted soils were containment and stabilization or solidification.

The in-situ remediation of a silt loam contaminated by Pb was carried out using electrokinetic soil flushing (Reed et al. 1995). Acetic acid (HAc), HCl and EDTA were used as conditioning agents around the electrodes to enhance the transport of Pb to the cathode. By adding 1 M HAc to the cathode reservoir, 65% of the Pb was transported to the cathode. When HCl was added to the anode and HAc to the cathode, more than 75% of the Pb was deposited on the cathode. Approximately 90% of the Pb was removed in the EDTA experiment as compared to the HCl-HAc experiment (65%).

There are 12 commercially available soil-washing processes for site remediation, six from USA and six from Europe. The processes were evaluated to clean up soils contaminated with heavy metals from abandoned chemical and pesticide plants in Italy to develop the area for residential and commercial complexes (Dennis et al. 1992). These processes were mostly developed for organic pollutants, however some of them have been used for both organic and inorganic pollutants. The removal of both organic and inorganic pollutants required studies of bench-scale treatability in order to provide the reassurance of system performance and scale-up data. Approximately 125,000 m^3 of contaminated soil have been estimated for cleaning operation at an estimated total cost of $22 million. This cost excluded excavation, debris removal, treatment and disposal of residual solids and effluents, site restoration, utilities and bench- and pilot-scale studies costs.

2.4 Bioremediation of Soil Polluted by Heavy Metals

Microbial oxidation or reduction of metals can be used to remediate soils contaminated by heavy metals. For example insoluble metal sulfides may be oxidized to form soluble metal sulfates by the microorganism *Thiabacillus ferrooxidans:*

$$ZnS + 2.5\,O_2 + 2H^+ \rightarrow Zn^{2+} + SO_4^{2-} + H_2O \qquad (2.1)$$

In natural environment, iron is generally associated with the leaching system of sulfide minerals where the bacteria oxidize pyrite (FeS_2) by the following reaction:

$$2FeS_2 + 7.5O_2 + H_2O \rightarrow 2Fe^{3+} + 4SO_4^{2-} + 2H^+ \qquad (2.2)$$

Except for Pb and Hg, the metal sulfate dissolved from the matrix can be collected for subsequent treatment. The biological activity used for mobilization of heavy metals is analogous to soil washing or chemical leaching (Wildeman et al. 1994). The bioleaching process is suitable when metals are present in soils as sulfide minerals such as in metal mining or soil under anaerobic conditions following the application of sewage sludge or sediments. However, this process produces sulfuric acid which disturb soil properties.

Under anaerobic conditions, the microbial reduction of sulfate produces insoluble metal sulfides, thereby reducing the mobility of heavy metals in soil. Therefore, it is difficult to remove metals from the soil by such microbial processes. Because, metals can be solubilized and then immobilized by sulfur as they are exposed in sequences, to aerobic and anaerobic conditions.

Several microorganisms such as yeast and several fungi are able to accumulate considerable amounts of organic acids such as citric, oxalic, malic, tartaric, fumaric and pyruvic acids (Rehm and Reed 1983). Some organisms have high rate of production of organic acids ranging up to 90% of the carbon source supplied. Citric acid is a common metabolite present in most living organisms. General nutritional requirements for the citric acid production by *Aspergillus niger* in surface and submerged cultivation are incorporated in Table 2.1. These two surface and submerged systems differ considerably from the effect of nitrogen, phosphorus, trace metals, pH and sugar concentration. The nitrogen and phosphorus are the important factors determining citric acid accumulation particularly in the case of submerged system (Table 2.1). The production of citric acid has not been influenced by the presence of these two nutrients in the case of surface cultivation (Steinbock et al. 1991).

Table 2.1 Nutrient parameters influencing citric acid accumulation by *Aspergillus niger* (+ influence and - no influence)

Parameter	Culture	
	Surface	Subsurface
Sugar type	+	+
Sugar concentration	+	+
Nitrogen source and concentration	-	+
Phosphate concentration	-	+
Metal ions (Mn, Fe, Zn) concentration	-	+
pH	+	+

(Source: Steinbock et al. 1991)

The effect of pH on the production of organic acids is demonstrated in Table 2.2. If the cultivation of *Aspergillus niger* is carried out at a pH above 4, gluconic and oxalic acids are formed at the expense of citrate, depending on the availability of nitrogen and phosphorus. Nitrogen, phosphorus, zinc, manganese and iron have little effect on the production of citric acid when surface cultivation was used. The production of citric acid was drastically reduced in the case of submerged cultivation in the presence of trace metals.

The parameters of major importance to citric acid accumulation are pH as well as the concentration and type of sugar (Xu et al. 1989). The optimal initial pH was around 4 under surface and submerged conditions (Figure 2.1A). The most important finding was that *Aspergillus niger* could grow well at pH values around 2.5 to 3.5 and that citric acid had abundantly produced at pH values lower than 2.0. Most striking fact, however, was the finding that yields of over 60% could be achieved within 1 to 2 weeks. At pH values below 3, citric acid is the major byproduct, whereas at higher pH values, substantial amounts of oxalic and gluconic acids may be produced.

Table 2.2 **Relationship of pH, nitrogen and phosphate on the accumulation of organic acids by *Aspergillus niger* (+ indicates the presence of nutrient)**

pH	Concentration		Organic acid accumulated
	N	P	
2	+	+	Citric acid
2	+	-	Citric acid
2	-	-	Citric acid
5	+	+	Oxalic acid
5	+	-	Oxalic acid and gluconic acid
5	-	-	Gluconic acid

(Source: Steinbock et al. 1991)

Maximum citric acid accumulation was observed at 10% sucrose concentration for both surface and subsurface cultivations (Figure 2.1B). Only few carbohydrates give

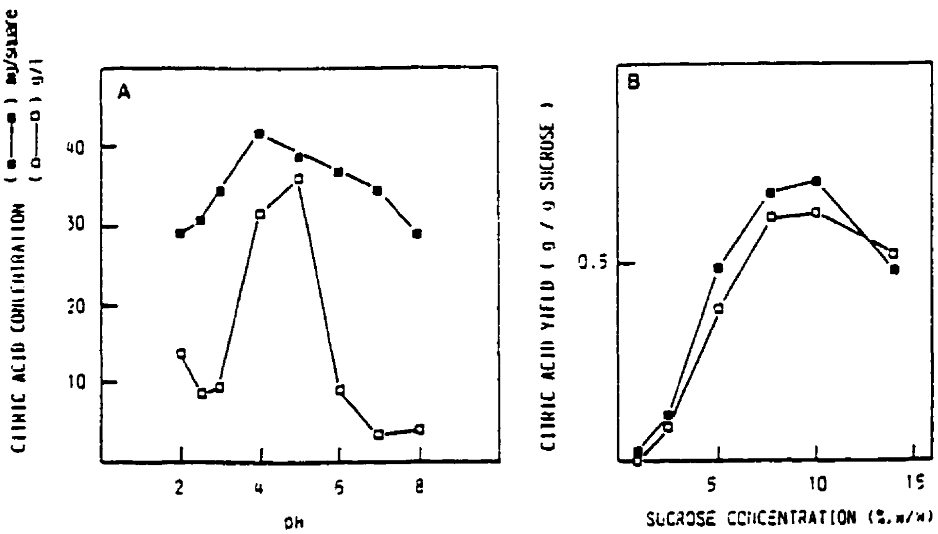

Figure 2.1 **Citric acid production by *A. niger* in submerged cultivation (□) and in filter paper surface cultivation (■). A: effect of initial pH, B: effect of initial sucrose concentration (Steinbock et al. 1991).**

optimum yields of citric acid even at high concentrations. The amount of citric acid extracted from the surface cultivation was significantly lower than that secreted under the same conditions in submerged cultivation. Aeration is extremely critical for submerged system because the production of citric acid increases with aeration. This organism (*A. niger*) has been used for the commercial production of citric and oxalic acids (Rehm and Reed 1983).

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CONNECTING STATEMENT

The next 7 chapters described seven successive experiments carried out to: i) provide an overview of the retention form of heavy metals in the three experimental and naturally polluted soils, ii) evaluate the efficiency of various organic acids to remediate the experimental clay loam polluted by heavy metals, iii) evaluate the efficiency of various organic salts to remediate the experimental loam and sandy clay loam polluted by heavy metals iv) Study the kinetics of heavy metal leaching from the three experimental soils (clay loam, loam and sandy clay loam) using citrate, tartarate and EDTA, v) evaluate organic acids for the in-situ remediation of soils polluted by heavy metals by soil flushing in columns, vi) study the bioremediation of soils polluted by heavy metals using *Aspergillus niger* a fungus producing citric and oxalic acids and vii) study the treatment of leachate from the soil washing process using weak organic acids.

The last chapter (chapter 10) presents the summary and conclusion, while the complete reference list is presented in chapter 11.

Chapter 3 deals with the retention form of heavy metals in three experimental and naturally polluted soils. The physical and chemical properties were initially measured for these three soils. Then, the relative proportion of heavy metals held by each soil retention form was measured through sequential extraction. Soils characteristic and retention form of heavy metals are important to understand how heavy metals are bound in different fractions of soils such as exchangeable, Mn/Fe oxides, organic matter or residual. This relationship between soil and heavy metals, establishes a better scientific basis for deciding what remediation approach to take at a given site and to help in developing new clean up technology.

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CHAPTER 3

RETENTION FORM OF HEAVY METALS IN THREE POLLUTED SOILS

3.1 Abstract

Three soils contaminated by heavy metals were characterized as a clay loam, loam and sandy clay loam. Their organic matter, pH and saturated hydraulic conductivity (k_s) ranged from 5.4 to 9.9%, 4.2 to 6.3 and 1.1 to 4.0×10^{-4} cm/s, respectively. The metal content (in mg/kg) were: i) Cr :840, Mn: 868, Hg: 1280 and Pb: 1664 for the clay loam, ii) Cd: 1821 and Pb: 2797 for the loam and iii) Cd: 794, Cu: 926, Pb: 1036 and Zn: 1029 for the sandy clay loam. As opposed to $MgCl_2$ and $CaCl_2$, $BaCl_2$ was found to better exchange the heavy metals in all 3 soils. With $BaCl_2$ 31 to 70% of Cd and Zn were exchanged, while only 17 to 20% of Hg and Pb were exchanged. By sequential extraction on all 3 soils, most of the heavy metals were found to be bound to Fe/Mn oxides and the organic fraction (30 to 40 %) except for Cd and Zn which were mainly present on the exchangeable fraction (63 to 87 %).

Key Words: Soil contaminated by heavy metals, characterization, sequential extraction, metal retention in the soil.

3.2 Introduction

The contamination of soils and groundwater by heavy metals is an ongoing problem in North America, Japan and Europe because of the current anthropogenic practices of mining and garbage landfilling (Asami 1988). Subsoils polluted by heavy metals are subjected to leaching leading to the contamination of groundwater reservoirs and drinking water supplies. Furthermore, leaching can be accelerated by acid rains (Matschullat *et al.* 1992). When accumulated in surface soils, heavy metals such as lead, cadmium, copper, zinc, nickel, chromium and mercury can be present in concentrations toxic to plants, animals, humans and aquatic life (Martin *et al.* 1976, Dowdy and Volk 1983). To be successful on a specific site, the remediation technique must be selected according to the form of retention of the heavy metal on the soil particles.

Heavy metals in soils can be associated with a number of geochemical phases or forms of retention such as exchangeable on clay minerals, bound on iron and magnesium oxides, adsorbed by organic matter and bound by carbonates (Asami *et al.* 1995; Yarlagadda *et al.* 1995). The exchangeable ionic form is easiest to remove from a contaminated soil using standard ion-exchange extractants such as ammonium acetate (NH_4-OAc), dilute HCl, NaCl and $MgCl_2$ (Engler *et al.* 1977). The Mn and Fe oxides, either single or in combination, play an important role in scavenging heavy metals from polluted leachate. Mn and Fe oxides are relatively stable in soils under aerobic conditions but can come into solution under anaerobic conditions generated most often by the oxidation of organic matter by microorganisms. Therefore, Mn and Fe oxides should undoubtedly be considered when assessing the risk of heavy metal leaching into groundwater. Soil organic matter can loosely chelate heavy metals or adsorb them tightly within its humus fraction. The decomposition of organic matter mineralizes the heavy metals. Also, organic matter under anoxic conditions normally can generate H_2S forming highly insoluble sulfides when in contact with heavy metals. Increasing the soil/sediment redox potential releases the metals from the sulfides. Metals can also react with solid carbonates and be incorporated into their solid lattice rather than purely adsorbed on their surface. Zinc is adsorbed more strongly on $MgCO_3$ than on calcite because the Mg atom has an ionic radius that more closely matches that of Zn. Cadmium is preferentially adsorbed on calcite and diffuses inside the matrix of $CaCO_3$ (Yong et al. 1992).

Sequential extraction of heavy metals from contaminated soils or sediments is used to identify the forms of heavy metals retained in the soil. This technique consists in submitting the soil to the successive action of a series of reagents with different chemical properties. Being capable of either creating weak acidic conditions, changing the redox potential of the solution or oxidizing the complexing agents, these agents extract a fraction of the heavy metals linked to a specific form of retention. In recent years, sequential extraction procedures have been used increasingly for environmental studies estimating the relative bonding strengths of metals in different forms of retention. The knowledge obtained from such extractions indicates the mobility and the solubility of the metals in water (Lake *et al.* 1984, Tessier *et al.* 1979, Rule and Alden 1992, Engler *et al.* 1977, Fiedler *et al.* 1994, Legret *et al.* 1988, Dudka and Chlopecka 1990).

Although there were initially some doubts as to the performance of the sequential extraction technique for heavy metals, it is now an accepted procedure. The selectivity of the chemical agents and the re-adsorption of trace metals during sequential extraction were the main aspects initially raising doubts (Nirel and Morel 1990, Tessier and Campbell 1991). Despite these problems, the use of sequential extraction as a procedure for the determination of how heavy metals are retained in a soil has continued to be recognized as a valuable tool (Lopez-Sanchez *et al.* 1993, Tessier *et al.* 1979). The results obtained have been helpful in identifying the retention mechanisms of heavy metals under different soil buffering conditions. In 1987, the European Community Bureau Reference (BCR now Measuring and Testing Program) started a program to harmonize the methodology, using single and sequential extractions schemes for the determination of extractable metals from soils and sediments. A single three step scheme for the sequential extraction of heavy metals from soils and sediments was proposed and accepted by European experts for inter-comparison in the BCR program (Lopez-Sanchez and Rauret 1993, Fiedler *et al.* 1994).

The objective of the present study was therefore to use sequential extraction procedure to characterize the forms of retention of various heavy metals in contaminated soils. The results of this study can be quite useful in designing a remediation process for a site contaminated by heavy metals.

3.3 Materials and Methods

3.3.1 The experimental soils and their preparation

Soil samples were obtained from a site contaminated by various heavy metals at least for three years. The moist soil samples were air dried and sieved to remove the particles greater than 2 mm. The soil samples were homogenized and kept in plastic bags for further analyses.

3.3.2 Physical and chemical characterization of the soil

The experimental soils were characterized physically by determining their particle size (hydrometer and field method), bulk density (core method for disturbed soil), particle density (pycnometer method), porosity and saturated hydraulic conductivity, k_s (constant head method) (Klute 1986). The experimental soils were also analyzed chemically for:

i) water and buffer pH,
ii) electrical conductivity,
iii) cation exchange capacity (CEC),
iv) total sulfur, SO_4^{2-} and S^{2-},
v) organic matter and
vi) heavy metal content.

The pH and buffer pH of the soils were measured by soaking them in deionized water in the H_2O:soil ratio of 2:1 and in the same ratio of a $CaCl_2$ (0.01 M) solution. The electrical conductivity (EC: mS/cm) of the soil's supernatant in deionized water was also measured using an electrical conductivity meter. The cation exchange capacity (CEC in cmol(+)/kg) was calculated by exchanging heavy metals and other cations present in the contaminated soils with $BaCl_2$ solution (Hendershot *et al.* 1993). Barium does not interfere with sulfate ion to form $BaSO_4$ because of Pb or Hg which forms very stable sulfate compounds if soluble sulfate is present. All metals except Na and K were determined by ICP-AES (ICPS-1000 II, Shimadzu Corp., Kyoto, Japan). Sodium and K were determined by flame emission spectrophotometry (Perkin Elmer).

Total sulfur, sulfur as SO_4^{2-} and S^{2-}, were determined by the triple acid digestion method using HNO_3-$HClO_4$-H_3PO_4 (Page *et al.* 1982). The sulfur content present in soil as sulfate was determined by digesting the soil sample (2 g) in a 6 mol/l HC1 solution overnight with continuous stirring at 60 °C. Sulfur present as sulfide was calculated as the difference between total and sulfate sulfur (Fiedler *et al.* 1994). The sulfur concentration in the solution was determined by ICP-AES.

The organic matter was measured by a wet dichromate oxidation technique (Jackson 1958).

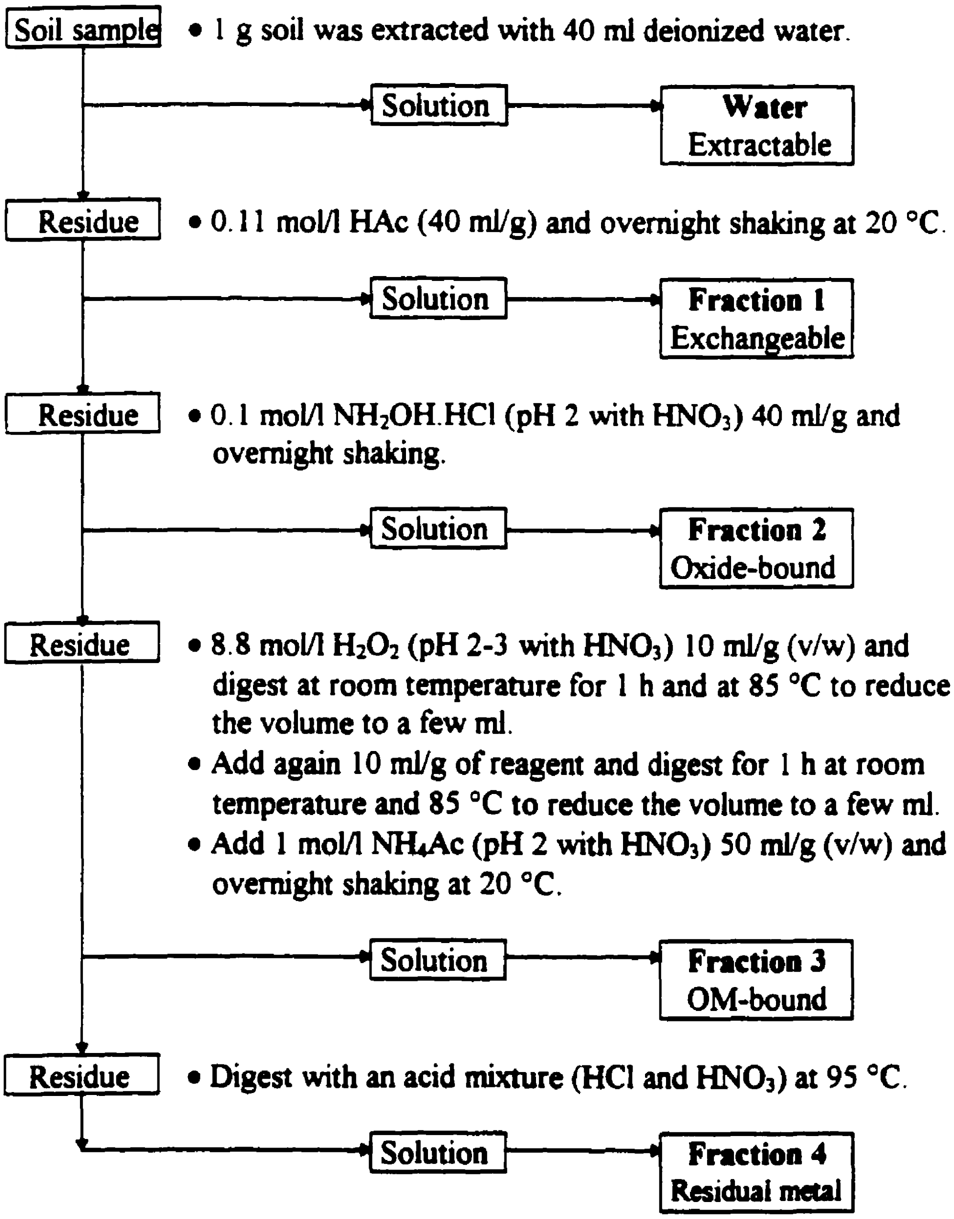
Except for mercury, heavy metals in the soil samples were extracted by the hot acid digestion procedure according to the ASTM D3974-81 A (ASTM 1993). The hot acid digestion procedure was performed in a Erlenmeyer flask containing 1 g soil to determine the content of the most abundant metals in the soil, using a dilute HNO_3/HCl mixture at 95 °C. To extract Hg, the soils were digested cautiously with 25 ml of aqua regia prepared just prior to use by mixing 1 part of concentrated HNO_3 to 3 parts of concentrated HCl. The mixture was heated for 1 min. and 25 ml of 5% $KMnO_4$ and 2 ml of 5% $K_2S_2O_8$ were added carefully. The resulting mixture was allowed to stand for 16 h. The residual $KMnO_4$ was reduced with 20% hydroxylamine hydrochloride solution to a faint pink color. The mixture was filtered and diluted using a 100 ml volumetric flask (Jacobs and Keeney 1974). All metals extracted from the soil samples such as Cr, Mn, Hg, Pb, Cd, Cu and Zn were determined by ICP-AES.

3.3.3 Heavy metals exchangeable with $MgCl_2$, $CaCl_2$ and $BaCl_2$

One gram of each experimental soil was suspended in 25 ml of 0.1 M $MgCl_2$, $CaCl_2$ and $BaCl_2$ separately in 35 ml polycarbonate tubes. The pH of each solution was adjusted between 4 to 6 using dilute HCl. The resulting samples were shaken for 2 h at 20 °C and then centrifuged to separate the soil and liquid phases at 10,000 rpm for 20 min. The supernatant was collected and analyzed for heavy metals. Similarly, the soil samples were also extracted in distilled water without the addition of $MgCl_2$, $CaCl_2$ and $BaCl_2$ solution as a control experiment. All experiments were carried out in triplicates.

3.3.4 Sequential extraction of heavy metals

The heavy metals bound on different fractions of the contaminated soils were determined by the most recently accepted sequential extraction procedure (Fiedler *et al.* 1994). The portion of heavy metals bound in four different fractions, HAc-extractable, Fe-Mn oxides, organic matter and silicate minerals, were determined by extracting in sequence using four solutions and triplicate soil samples (Figure 3.1). The heavy metals bound with HAc-extractable, Mn-Fe oxides and organic matter as well as silicate minerals (residual) were extracted in fraction 1, 2, 3 and 4 respectively. In the extractable fraction,



- 1 g soil was extracted with 40 ml deionized water.
- 0.11 mol/l HAc (40 ml/g) and overnight shaking at 20 °C.
- 0.1 mol/l $NH_2OH.HCl$ (pH 2 with HNO_3) 40 ml/g and overnight shaking.
- 8.8 mol/l H_2O_2 (pH 2-3 with HNO_3) 10 ml/g (v/w) and digest at room temperature for 1 h and at 85 °C to reduce the volume to a few ml.
- Add again 10 ml/g of reagent and digest for 1 h at room temperature and 85 °C to reduce the volume to a few ml.
- Add 1 mol/l NH_4Ac (pH 2 with HNO_3) 50 ml/g (v/w) and overnight shaking at 20 °C.
- Digest with an acid mixture (HCl and HNO_3) at 95 °C.

Figure 3.1 Schematic diagram of the sequential extraction procedure for the determination of heavy metals bound to different fraction of contaminated soil.

metals precipitated or co-precipitated as natural sulfate or sulfide or as oxides were released by the application of a weak acid such as acetic acid (HAc). The metals attached to amorphous or poorly crystallized Fe/Mn oxides were extracted using acidic solution of hydroxylamine hydrochloride ($NH_2OH.HCl$) which does not attack either the silicate minerals nor the organic matter. The release of the metals bound to the organic matter was carried out by oxidation of the organic matter by hydrogen peroxide (H_2O_2) under acidic condition. All four fractions including residual fraction (treated with strong acids such as HCl-NHO_3) were analyzed by ICP-AES to quantify these levels of heavy metals. All experiments were carried out in triplicate.

3.4 Results and Discussion

3.4.1 Physical and chemical characterization of the soil

Both the hydrometer and field methods for soil particle size distribution indicated that the texture of the three experimental soils were clay loam, loam and sandy clay loam, respectively. The low particle density of all three soils ranged from 2.0 to 2.1 g/cm^3 and was explained in part by their high organic matter content ranging from 5.4 to 9.9%. The k_s of each soil ranged from 1.1 to 4.0 x 10^{-4} cm/s and increased in parallel with their porosity and texture. All these soils were acidic in nature with a pH (in water) of 4.2, 5.1 and 6.3, respectively. Their EC decreased with pH probably as a result of soil particles binding strongly the metals and salts as soil pH increased. The results of the physical and chemical characterization of the contaminated soils are incorporated in Table 3.1.

The clay loam, loam and sandy clay loam contained 1088, 846 and 1467 mg/kg of total sulfur, of which 9.9, 13.6 and 51.8% was sulfide, respectively. The CEC varied from, 2.0 to 12.2 cmol (+)/kg for the clay loam to the sandy clay loam. Clay loam has low CEC, may be because of exchangeable sites on soil surface is tightly occupied by heavy metals which is difficult to exchange with $BaCl_2$ solution. The soil's heavy metal content was: i) for the clay loam, 840, 868, 1280, 1664 mg/kg of Cr, Mn, Hg and Pb, respectively, ii) for the loam, 2797 and 1821 mg/kg of Pb and Cd, respectively and iii) for the sandy clay loam, 1036, 794, 926 and 1029 me/kg of Pb, Cd, Cu and Zn, respectively. All three experimental soils were found to contain high levels of heavy metals exceeding the permissible levels (MEFQ 1994). The fraction of heavy metals held by either form of sulfur is an indication of their fixation. Sulfates are insoluble when bound to Pb and Hg, but soluble when bound to Cu, Zn and Cd. Thus, Pb and Hg are less mobile in the presence of sulfates. Metals have a strong affinity for sulfides and are therefore precipitated in their presence. Metal sulfides such as PbS, CuS, NiS, ZnS and

Table 3.1. physical and chemical characterization of contaminated soils.

Soil characteristic	Clay loam	Loam	Sandy clay loam
Particle size			
% sand	29.0±0.6	49.5±0.5	53.5±0.9
%Silt	36.0±0.4	30.0±1.2	24.0±1.6
% clay	35.0±1.0	20.7±0.7	22.7±0.7
Bulk density (g/cm^3)	0.8±0.0	0.9±0.0	0.9±0.0
Particle density (g/cm^3)	2.0±0.0	2.0±0.0	2.1±0.1
Total porosity (%)	45.4±0.1	44.1±0.2	48.0±0.4
k_s(cm/s)	1.1±0.1x10^{-4}	1.3±0.1x10^{-4}	4.0±0.2x10^{-4}
EC (mS/cm)-water	9.2±0.2	8.0±0.2	2.3±0.2
pH-water	4.2±0.0	5.1±0.1	6.3±0.1
-$CaCl_2$	4.3±0.1	5.2±0.1	6.0±0.0
CEC (cmol(+)/kg)	2.0±0.2	4.7±0.0	12.2±0.2
Sulfur (mg/kg)-total	1088.0±2.4	846.0±2.0	1467.0±19.5
-as SO_4^{2-}	980.0±4.8	731.0±4.8	705.6±10.5
-as S^{2-}	108.2±7.2	115.0±2.8	761.5±9.0
Organic matter (%)	9.9±0.1	5.4±0.1	7.1±0.1
Heavy metals (mg/kg)			
Cr (Total)	840	-	-
Mn	868	-	-
Hg	1280	-	-
Pb	1664	2797	1036
Cd	-	1821	794
Cu	-	-	926
Zn	-	-	1029

Ag_2S, are stable under anaerobic condition, but they become unstable and oxidize when exposed to air at the soil surface. The reaction under aerobic conditions produces sulfuric acid which increases the solubility of heavy metals and promotes their mobility (Yong *et al.* 1992).

3.4.2 Sequential extraction

The results of the sequential extraction of the three contaminated soils are incorporated in Table 3.2. Heavy metals retained in different fraction of soils are shown in Figure 3.2.

3.4.2.1 Water soluble

For all 3 experimental soils, the water soluble fraction was generally small or almost nonexistent for Zn, Cu. Pb, Cr and Hg. Water extractable Mn in the clay loam was found to be 21.1 %, whereas for all other metals it was less than 3.7 %. 15.2% of Cd in the loam was water extractable, whereas Pb was found to be negligible (0.8%). Water extractable Pb, Cd, Cu and Zn from the sandy clay loam in water medium was found to be 4.5, 0.70, 1.9 and 0.95%, respectively (Table 3.2 and Figure 3.2).

3.4.2.2 Acetic acid-extractable

In the clay loam, the extractable fraction of Pb was found to be 25% of the Pb, but less then 8% of the other metals. With the loam, Cd was mainly bound in extractable (or exchangeable) fraction (63%), whereas 26% of the Pb was found in extractable fraction. The percentage of Cd, Cu. Pb and Zn in the sandy clay loam, bound to the extractable soil particle site was 87, 28, 9 and 67%, respectively (Figure 3.2). Because Cd and Zn are chemically similar, their adsorption on the soil particles were similar, as expected (Mat *et al.* 1994).

3.4.2.3 Metal oxide fractions

In the clay loam, the Fe/Mn oxides accounted for a major portion of the Cr, Mn, Hg and Pb, which were calculated to be 23, 45, 21 and 31%, respectively. In the loam, 40% of the Pb was found bound to Fe/Mn oxides, whereas only 12 % of the Cd was found on

Table 3.2 Heavy metal concentrations in the geochemical phases of soil

Soil	Geochemical phase (mg/kg)					Total
	Water	Ex.	Fe-Mn oxides	Organic matter	Residual	
Clay loam						
Cr	2.1±0.5	67.0±7.0	190.6±7.0	486.9±7.2	84.0±3.5	832.2±13.0
Mn	183.0±6.6	10.0±6.4	392.2±12.0	198.9±12.3	84.3±2.8	868.1±2.6
Hg	48.0±1.5	71.7±6.0	266.0±10.1	766.0±8.4	123.0±2.0	1273.3±21.7
Pb	36.0±2.0	411.6±2.0	509.0±17.8	670.0±9.2	54.4±10.1	1624.3±26.1
Loam						
Cd	276±10.1	1144.0±19.2	222.4±7.0	80.0±1.6	13.2±1.8	1736.0±28.0
Pb	22.4±1.5	731.5±9.5	1138.0±13.3	536.2±14.3	284.0±6.0	2721.4±32.5
Sandy clay loam						
Cd	5.5±0.3	687.6±7.5	81.4±2.8	12.2±3.2	ND	786.8±3.2
Cu	17.5±1.2	256.1±6.6	273.2±8.5	287.0±13.5	78.2±3.8	913.0±24.3
Pb	46.6±1.9	96.1±10.3	431.8±16.0	415.1±6.2	8.7±2.0	998.4±21.4
Zn	9.8±0.3	687.0±8.1	178.2±6.4	42.0±8.5	97.0±7.6	1013.4±25.6

Water (W): water soluble, Ex.: Acetic acid (HAc)-extractable, the values are the average of the triplicates.

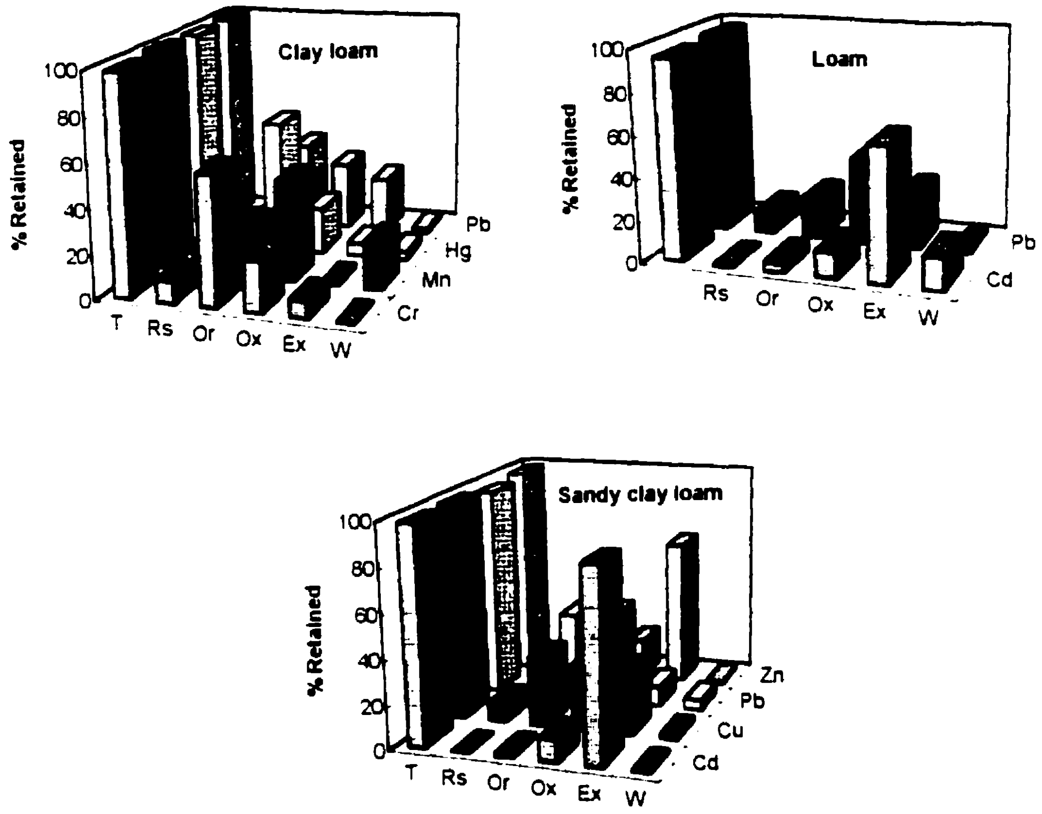


Figure 3.2 Heavy metals in various fractions of polluted soil (water extractable -W, HAc-extractable (loosely bound)-Ex, Mn-Fe oxides -Ox, organic matter -Or, residual -Rs and total-T).

this fraction. In the sandy clay loam, the Fe/Mn oxides had bound 30 and 40% of the Cu and Pb, whereas 15 and 17% of the Cd and Zn had been bound by this form (Table 3.2). Lead tends to have a high affinity for the Mn/Fe oxides in partially anaerobic condition (Mat *et al.* 1994). The oxides/hydroxides of Fe and Mn and organic fractions are considered to be the most important geochemical components for retaining heavy metals in soils and sediments (Tessier *et al.* 1985, Rule and Alden 1992, Young and Harvey 1992). However, the adsorption of metals by these fractions is largely dependent on their overall abundance relative to each other (Young and Harvey 1992).

3.4.2.4 Organic fraction

High levels of heavy metals such as Cr, Mn, Hg and Pb were found on the organic fractions. In the clay loam, these levels were 58, 23, 60 and 40%, respectively. In the loam, the organic fraction held 32% of the Pb, but only 4% of the Cd. This fraction retained significant amounts of Cu (31%) and Pb (40%) in the sandy clay loam, but insignificant levels of Cd and Zn (Figure 3.2, Table 3.2). Most metals in anaerobic sediments are expected to be associated with the organic/sulfide fraction (Rule and Alden 1992).

The presence of high organic matter (5.4 to 9.9%), may increase anoxic conditions in sediments (Ahmad 1993). Soils of this nature normally have high levels of hydrogen sulfides forming insoluble metal sulfides except for Mn and Fe, which are not easily exchangeable (Rule and Alden 1992). For the three experimental soils, the organic/sulfide fraction contain a significantly high levels of Cu (31 %), Pb (40%), Hg (60%) and Cr (58%), reflecting its high affinity for heavy metals because of its ligands or groups that can form chelates with the metals (Yong *et al.* 1992). The relevant functional groups present in organic matter are amino, imino, carboxyl, phenolic, alcoholic, carbonyl and sulfhydryl. With increasing pH, the stability of the complexes are likely to increase because of the increased ionization of the functional groups. Nevertheless, Cu will maintain stable complexes over a wide range of pH (Jones and Jarvis 1981). The order for the stability of heavy metals complexes is as follows: $Cu^{2+} > Fe^{2+} > Pb^{2+} > Ni^{2+} > Co^{2+} > Mn^{2+} > Zn^{2+}$. Heavy metals associated with sulfides can be available into reducible, carbonate and exchangeable fractions after an increase in the redox potential (Rule and Alden 1992).

3.4.2.5 Residual fraction

Metals in this fraction are generally considered to be bound within the lattice of silicate minerals and can become available only after digestion with strong acids at elevated temperature. This fraction is not considered to be significantly large and is important only in completing mass balance calculations. Heavy metals in the residual fraction were found to be less than 10% in all three soils (Figure 3.2). Cadmium was found to be undetectable, whereas Pb was found at levels of 10% (means 150-250 mg/kg Pb bound in silicate mineral lattices that are still higher than clean up criteria of Pb for soil, which is

50 mg/kg). In the Figure 3.2, total heavy metals is slightly less than 100% which may be due to loss in sequential extraction of different fraction of soils.

3.4.2.6 Exchangeable fraction

Heavy metals in the exchangeable fractions are weakly bound and expected to be easily leached out to the groundwater or absorbed by aquatic organisms. To extract the exchangeable fraction of heavy metals, ammonium acetate, sodium acetate and magnesium chloride have been used (Tessier *et al.* 1979). Ammonium acetate has been used extensively in the analysis of soil and sediment to extract the carbonate bound heavy metals. Salts such as $MgCl_2$, $CaCl_2$, and $NaNO_3$ are commonly used as ion-displacing extractants to promote the release of metal ions physically bound by electrostatic attraction to the negatively charged sites on the soil particle surfaces. Metal ions present in the soil in this form are considered to be the most "available" to plants (Soon and Bates 1982). Magnesium chloride, $CaCl_2$ and $BaCl_2$ were used for exchangeable heavy metals from polluted soils at varying pH to predict the availability to plants and groundwater contamination (Figure 3.3a and 3.3b).

In all 3 experimental soils, a lesser amount of exchangeable heavy metals was found with $MgCl_2$ as compared to $CaCl_2$. A high amount of exchangeable heavy metals was found when using $BaCl_2$ because Ba has a larger ionic size (1.34 A°) than Ca (0.99 A°) and Mg (0.66 A°). Same valence with increasing ionic size gives greater replacing power (Yong *et al.* 1992). The concentration of exchangeable metals decreases as pH increases because metals especially Pb, become more tightly bound to the soil surface. Therefore, almost negligible amounts of Pb were found in the sandy clay loam, whereas about 17% Pb was found to be exchangeable with $BaCl_2$ in both the clay loam and the loam with a more acidic nature (pH 4.2-5.1). The concentration of Pb as well as those of other metals decreased as the pH increased (Figure 3.3a and 3.3b). For a soil solution pH between 4 and 5, Pb was retained in an exchangeable form, but its adsorption decreases as the pH differed from this limit, to the benefits of the hydroxides and carbonates (Yong *et al.* 1992). A measure of this retention capacity is the cation exchange capacity.

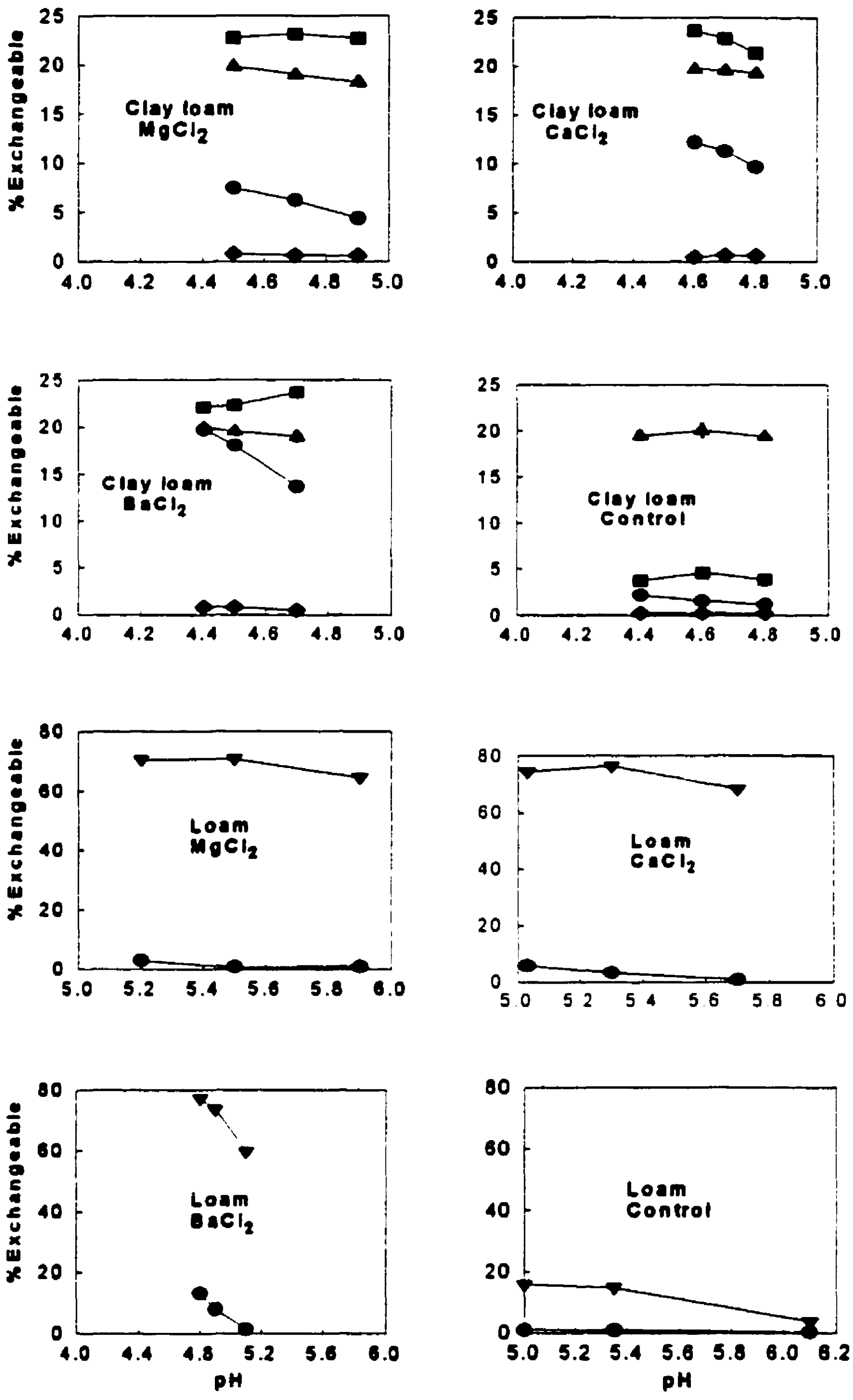


Figure 3.3a Exchangeable heavy metals from the soil with $MgCl_2$, $CaCl_2$ and $BaCl_2$ at different pH. Clay loam: ◆ Cr, ■ Hg, ▲ Mn and ● Pb, Loam: ▼ Cd and ● Pb.

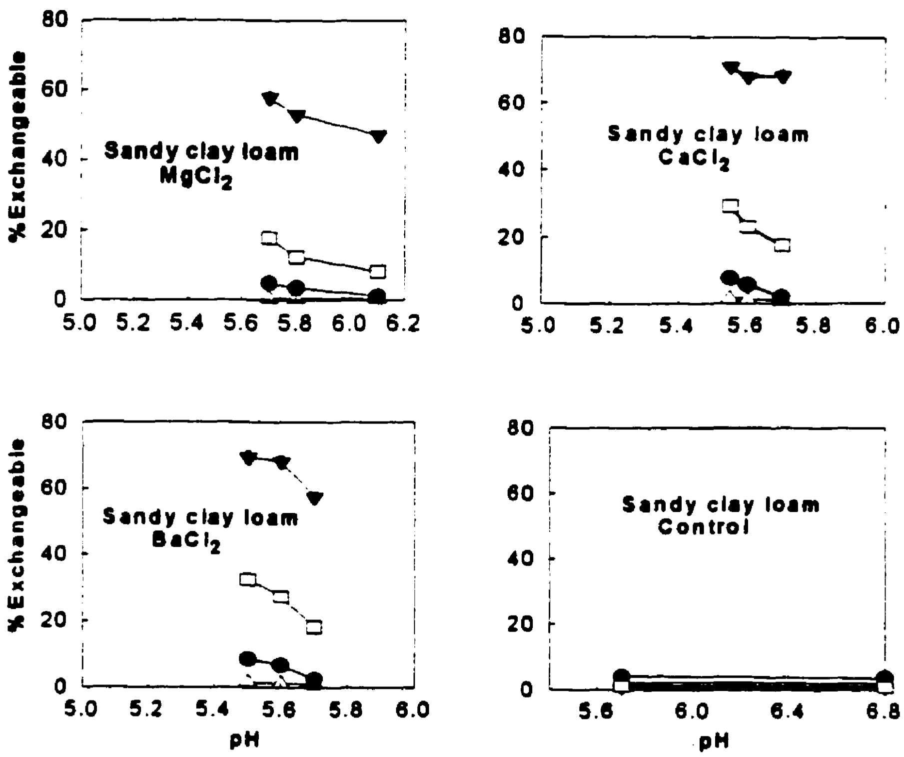


Figure 3.3b Exchangeable heavy metals from the sandy clay loam with $MgCl_2$, $CaCl_2$ and $BaCl_2$ at different pH. ▼ Cd, □ Zn, Δ Cu and ● Pb.

3.5 Summary and Conclusions

To evaluate the possible remediation process, three textures of soils polluted by heavy metals were tested for forms of retention. The soils were initially characterized for particle size, bulk and particle densities, porosity, k_s, electrical conductivity, pH, sulfur, organic matter and heavy metals. The soils were characterized as a clay loam, a loam and a sandy clay loam with an organic matter content of 9.9±0.1, 5.4±0.1 and 7.1±0.1%, respectively. These soils were acidic, with a pH between 4.2 and 6.3. The saturated hydraulic conductivity varied between 1.1 to 4.0×10^{-4} cm/s. Heavy metal content in the contaminated soils were (mg/kg):

- Clay loam. Cr :840, Mn: 868, Hg: 1280 and Pb: 1664.
- Loam. Cd: 1821 and Pb: 2797.
- Sandy clay loam. Cd: 794, Cu: 926, Pb: 1036 and Zn: 1029.

For each of the 3 soils, the heavy metals were extracted from each geochemical phase (soil fraction) through selective means using the appropriate reagents such as inert electrolytes ($MgCl_2$, $CaCl_2$ and $BaCl_2$) for exchangeable metals, acetic acid for the extractable fraction, acidic hydroxyleamine hydrochloride for the Fe/Mn oxides, acidic hydrogen peroxide for the organic fraction and strong acids (HCl-HNO_3) for the residual fraction. From 30 to 40% of the heavy metals were bound to the Fe/Mn oxides and the organic fractions except for Cd and Zn which were mainly present as exchangeable fractions. The heavy metals in the residual fraction were found to be less than oxides and organic matter.

Magnesium chloride exchanged Hg and Pb in the clay loam at levels of 20 and 5%, respectively, whereas Cr and Mn were not exchanged. The exchangeable Hg and Pb in the clay loam were found to be 20 and 10% with $CaCl_2$ and 20 and 17% with $BaCl_2$, respectively. Both Cr and Mn were not exchangeable with $CaCl_2$ and $BaCl_2$. In the loam, 55 to 62% of Cd was exchanged and only 2.4 to 12.6% of Pb with either electrolytes, $BaCl_2$, $CaCl_2$ and $MgCl_2$. In the sandy loam, 57 to 70% and 16 to 31.5% of Cd and Zn, respectively, were exchanged with these electrolytes. Magnesium chloride was less capable of exchanging Cd than $CaCl_2$ and $BaCl_2$. A higher amount of Zn and Cd was exchanged by $BaCl_2$ than $CaCl_2$ while the least amount was exchanged with $MgCl_2$.

The sequential extraction of heavy metals proved a useful tool in understanding the retention mechanism of heavy metals in the soil. For designing the optimum remediation technique for a specific site, this method provides a means of assessing how strongly the contaminants are held in the soil.

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CONNECTING STATEMENT

After characterizing the three experimental soils and their form of heavy metals retention, the next chapter deals with the remediation of clay loam using various weak organic acids and/or their salts as compared to the chelating agents (EDTA and DTPA). Chapter 4 deals with the experimental clay loam and chapter 5 deals with the experimental loam and sandy clay loam. The remediation was carried out in batch experiments at various pH levels, concentration of weak organic acids or chelating agents and at different time intervals. The results were compared with the Quebec clean up criteria of the soil.

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CHAPTER 4

ORGANIC ACIDS TO REMEDIATE A CLAY LOAM POLLUTED BY HEAVY METALS

4.1 Abstract

Weak organic acids have the capabilities of washing heavy metals without seriously deteriorating the soil properties. A clay loam naturally contaminated by heavy metals was characterized physically and analyzed for its content in Cr, Hg, Mn and Pb. At various pH, concentration and reaction time, several weak organic acids and their salts were used to leach out heavy metals. The optimum pH ranged from 5 to 7 for citrate, 2 to 3 for tartarate in the case of Cr, Hg and Mn and 2 to 7 in the case of Pb. More than 85% of Pb was removed with both citrate and tartarate at a concentration of 0.12 and 0.2 M, respectively within 24 h. From 86 to 99% of Hg was removed by tartarate and citrate whereas up to 60% of Cr was removed. EDTA and DTPA were found to be quite effective in removing Pb but not the other metals. Four citrate or tartarate extraction are required to remediate the experimental soil to a category A except Hg. For Hg, remediation criteria are so strict that only met the C criteria after two extractions, citrate and tartarate left levels of 32 and 62 mg/kg, respectively, when a category A of soil clean up criteria requires no more than 0.5 mg/kg.

4.2 Introduction

Soil pollution by heavy metals is a growing concern in Canada, USA, Japan and Europe as well as developing countries. It is mainly due to metal mining, smelting industries (Asami 1988), and the production of large quantities of industrial wastes and municipal wastewater sludge. In most cases, wastes containing heavy metals are disposed in secured landfills, where there is always a risk of secondary pollution due to leakage. One of the negative environmental impacts, arising from landfills (if not lined properly) is surface and groundwater contamination due to run off and percolation of rainwater through the soil, slowly extracting metals which eventually reach aquifers. Acidification of the soil from acid rain accelerates this process by desorption of heavy metals from contaminated soil (Matschullat et al. 1992). Metal mining industries dominate in Quebec where about 50% of the groundwater contamination sites result from landfills built in sand dunes without liners or lined with materials, such as bentonite, which are subject to cracking (NRCC 1987). The metals in the leachate are highly toxic to humans, animals and aquatic life. Humans are exposed to heavy metals in the soil through food chain transfer. Chronic food chain exposure of heavy metal are more numerous as compared to acute toxicity in humans, some farmers in Japan suffered painful bone losses after the long-term consumption of Cd enriched rice. The rice has been grown in paddies soil polluted by mining and smelting activities (Pierzynski et al. 1994). The conventional remediation is costly when the soil is polluted by heavy metals, because they are tightly bound to various fractions of the soil. It has been estimated that the cleanup cost by current technology (ex-situ or excavation and reburial) for one hectare of soil over a depth of one meter (10, 1000 m^3) can range between $ 0.6 to 3 million depending on the pollution load (Moffat 1995).

Soil polluted by heavy metals can be cleaned up using an ex-situ remediation process. This remediation process requires the excavation of the soil from the site followed by its washing. It is one of the most suitable processes for removing heavy metals because mostly heavy metals are retained on the surface of the soil depending on the property of the soil. The washing solution contains either acids or chelating agents (Benschoten et al. 1994, Tuin and Tels 1990). The contaminants in the extraction solution can be treated for reuse by conventional methods (Eckenfelder 1989, Tels 1987). The problem with this washing process, especially with mineral acids (HCl, HNO_3) and strong chelating agents (EDTA and DTPA), is that the physical, chemical and biological properties of the soil are disturbed. Commercial plants using soil washing techniques for the remediation of soils contaminated with heavy metals and pesticides require preliminary bench scale treatability studies to select the extractant (Dennis et al. 1992).

EDTA and citric acid have been used to extract heavy metals such as Cu, Zn, Cd, Ni, Pb and Cr from contaminated soils at a neutral pH (Leidmann et al. 1994). EDTA can extract up to 90% of Cd, Cu, Pb and Zn, and 25 to 45% of Cr and Ni. Citric acid (0.26 M) can extract from 50 to 80% of Cu, Cd, Zn and Ni, but only 30 to 40% of Cr and Pb at a single neutral pH. The ability of seventeen different chemical solutions has been examined to displace heavy metals (Pb, Zn, Cu, Cd) from artificially contaminated clays such as kaolinite, illite and montmorillonite at either pH of 5 or 7 (Farrah and Pickering 1978). Of the reagents used, only EDTA (0.001 M, pH 7) quantitatively released all four metals from the three clays, while oxalic acid (0.1 M, pH 3.3) totally displaced three metals (Cu, Cd and Zn) from each clay. Other reagents such as ammonium oxalate (0.1 M), ammonium nitrate (0.01 M), nitric acid (0.1 M) and sodium citrate (0.01 M) effectively displaced one or more heavy metals (Cd and/or Zn or Cu) from clays.

Some organic acids such as acetic, citric and oxalic acids have been tested for the remediation of soils artificially contaminated with heavy metals at a single pH value (Farrah and Pickering 1978). Biological compounds such as glycine, citric acid, tartaric acid and gluconic acids are known to have chelating properties binding heavy metals and to occur naturally in soils (Lo et al. 1992, Huang and Schnitzer 1986). Heavy metals are less mobile when they form stable complexes with humic acid. On the contrary, the metal mobility and bioavailability increases when heavy metals combine with organic acids such as acetic acid. These organic acids are of low cost, biodegradable and less destructive to soil structure than strong mineral acids and strong chelating agents.

The present study was designed to: i) identify the best weak organic acids and/or their salts and to establish their potential in a batch experiment for the leaching of heavy metals from a naturally polluted soil and, ii) compare the performance of the best organic acids and their salts to the synthetic chelating agents, EDTA and DTPA.

4.3 Materials and Methods

4.3.1 Experimental soil, organic acids and chelating agents

The experimental soil was obtained from a 3 year old polluted site in Japan. The wet soil sample was air dried and sieved to remove particles greater than 2 mm (No. 10 sieve). The soil sample was homogenized and kept in a plastic bag for further analysis.

The following organic acids and their salts were used to study the remediation of the polluted soil by dissolving in distilled water: citric acid monohydrate (0.2 M) and ammonium citrate dibasic (0.2 M), oxalic acid dihydrate (0.2 M) and ammonium oxalate monohydrate (0.2 M), tartaric acid (0.5 M) and ammonium tartarate (0.5 M), acetic acid (0.11 M) and ammonium acetate (0.5 M), succinic acid (0.1 M) and sodium succinate (0.1 M), formic acid (0.2 M), phthalic acid (0.01 M), itaconic acid (0.2 M), formic acid (0.2 M), pyruvic acid (0.1M) and sodium pyruvate (0.1 M). The chelating agents used as a base for comparison were EDTA (ethylenediaminetetraacetic acid) disodium salt (0.1 M) and DTPA (diethlenetriaminepentaacetic acid, dissolved in sodium hydroxide solution) (0.1 M). Different concentrations of chelating agents were used because of different chelating strength. Dilute solutions of hydrochloric acid (0.1 M) and sodium hydroxide (0.1 M) were used for pH adjustment.

4.3.2 Procedure

The contaminated soil was characterized for texture, saturated hydraulic conductivity (k_s), pH, organic matter content, cation exchange capacity (CEC) and heavy metal contents (Klute 1986, Jackson 1958, ASTM 1993) (Table 4.1). Despite its high organic matter and clay particle content, the soil CEC was low because the exchangeable sites were occupied by heavy metals. Retention form of heavy metals such as water soluble, exchangeable, bound to organic matter, Fe/Mn oxides and residual in the soil was studied using a sequential extraction procedure (Wasay et al. 1998a) (Table 4.2). Heavy metals are mainly present in Fe/Mn oxides and organic matter of the soil which are difficult to remediate as compared to exchangeable form.

As control, two series of plastic tubes containing 25 ml of distilled water were prepared and their pH was adjusted in the range from 1.9 to 5.0 with 0.1 M HCl and/or NaOH. Another two series of 25 ml solutions was prepared using one of the organic acids or their salts or EDTA and DTPA. The pH values were adjusted either with the appropriate mixture of organic acids and their salts (in most cases) or by addition of a small amount of 0.1 M HCl and/or NaOH. The volume of the solution was maintained at

25 ml. One gram of contaminated soil was added to each tube and each series served as a duplicate. The suspension was shaken for 24 h at 20 °C, centrifuged (10,000 rpm for 20 min.) and the supernatant withdrawn. The residue was extracted once more with 25 ml of extracting organic acid solution, chelating agent or distilled water, as used initially. The supernatants were analyzed for metals by the ICP-AES (ICPS 1000 II, Shimadzu Corp., Kyoto, Japan).

To test the effect of extractant concentrations on heavy metal leaching, another series of 25 ml solutions was prepared but a various concentrations of one of the organic

Table 4.1 Physical and chemical characteristics of contaminated soil

Soil characteristic	Clay loam
Particle size	
% sand	29.0±0.6
% silt	36.0±0.4
% clay	35.0±1.0
Bulk density (kg/m^3)	0.8±0.0
Particle density (kg/m^3)	2.0±0.0
Total porosity (%)	45.4±0.1
k_s (m/s)	1.1±0.1x10^{-6}
pH-water	4.2±0.0
CEC (cmol(+)/kg)	2.0±0.2
Organic matter (%)	9.9±0.1
Heavy metals (mg/kg)	
Cr	840
Mn	868
Hg	1280
Pb	1664

Table 4.2 Retention form of heavy metals in clay loam

Soil	Geochemical phase (mg/kg)					Total
	Water	Ex.	Fe-Mn oxides	Organic matter	Residual	
Clay loam						
Cr	2.1±0.5	67.0±1.5	190.6±7.0	486.9±7.2	84.0±3.5	832.2±13.0
Mn	183.0±6.6	10.0±6.4	392.2±12.0	198.9±12.3	84.3±2.8	868.1±2.6
Hg	48.0±1.5	71.7±6.0	266.0±10.1	766.0±8.4	123.0±2.0	1273.3±21.7
Pb	36.0±2.0	411.6±2.0	509.0±17.8	670.0±9.2	54.4±10.1	1624.3±26.1

salts or the chelating agents. Only the pH of the chelating agents was adjusted using either HCl or NaOH in solution at 0.1 M. One gram of the soil was added to each tube and then shaken for 24 h at 20 °C before centrifugation. The supernatant was subjected to ICP-AES for heavy metal analysis.

At optimum pH, determined from the previous experiment (4.3.2), a kinetic study was carried out for the removal of heavy metals from the soil as a function of time. A series of 25 ml solution was prepared in plastic tubes containing either one of the organic salts or one of the chelating agents. One gram of the soil was added to each tube and then shaken at 20 °C for 10 min to 36 h. Then, each soil sample was filtered and the supernatant was collected for heavy metals quantification by ICP-AES.

4.3.3 Statistical analysis

The SAS procedures used are: i) One-way analysis of variance or randomized complete block design (RCBD) for pH effect. ii) repeated measures analysis of variance for the rate of leaching study with time (SAS 1990).

4.4 Results and Discussion

4.4.1 pH effect on leachability

The solubility and behavior of heavy metals was significantly affected by soil pH particularly in the case of organic acids and/or salts (Figure 4.1a and 4.1b) but not in the case of EDTA and DTPA, and particularly for Pb (Figure 4.1a). The maximum removal of Cr, Hg, Mn and Pb was found to be 54, 96, 69 and 85%, respectively in the presence of citrate when the pH ranged between 5 to 7. The pH range leads to the dissociation of the carboxyl groups of citrate (Huang and Schnitzer 1986).

The leachability of heavy metals with tartarate was also pH dependent except for Pb. The removal efficiency of Cr, Hg and Mn decreased as pH increased, whereas that of Pb remained constant at 86% in the wide pH range of 2 to 7. The removal of Cr, Mn and Hg was 41, 54 and 84% at pH 4.3, respectively. The dissociation of the carboxyl groups in tartarate takes place at a pH between 3 to 5 whereas dissociation of the phenolic groups takes place at a pH above 5. Therefore, Pb reacts more with the dissociated

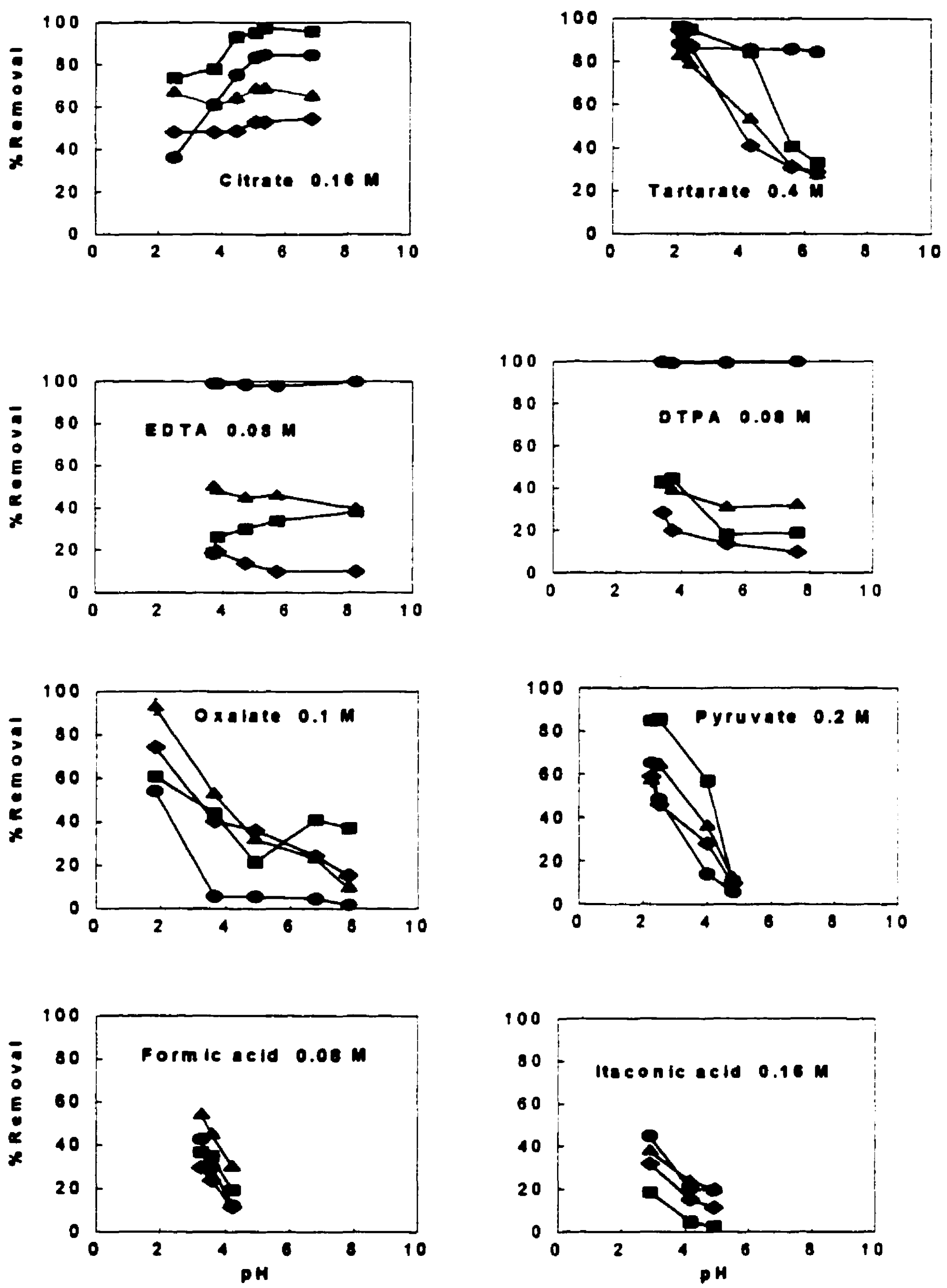

Figure 4.1a Effect of pH on the removal of heavy metals from the clay loam in a batch experiment after 24 h of retention time. Symbol: ◆ Cr, ■ Hg, ▲ Mn and ● Pb.

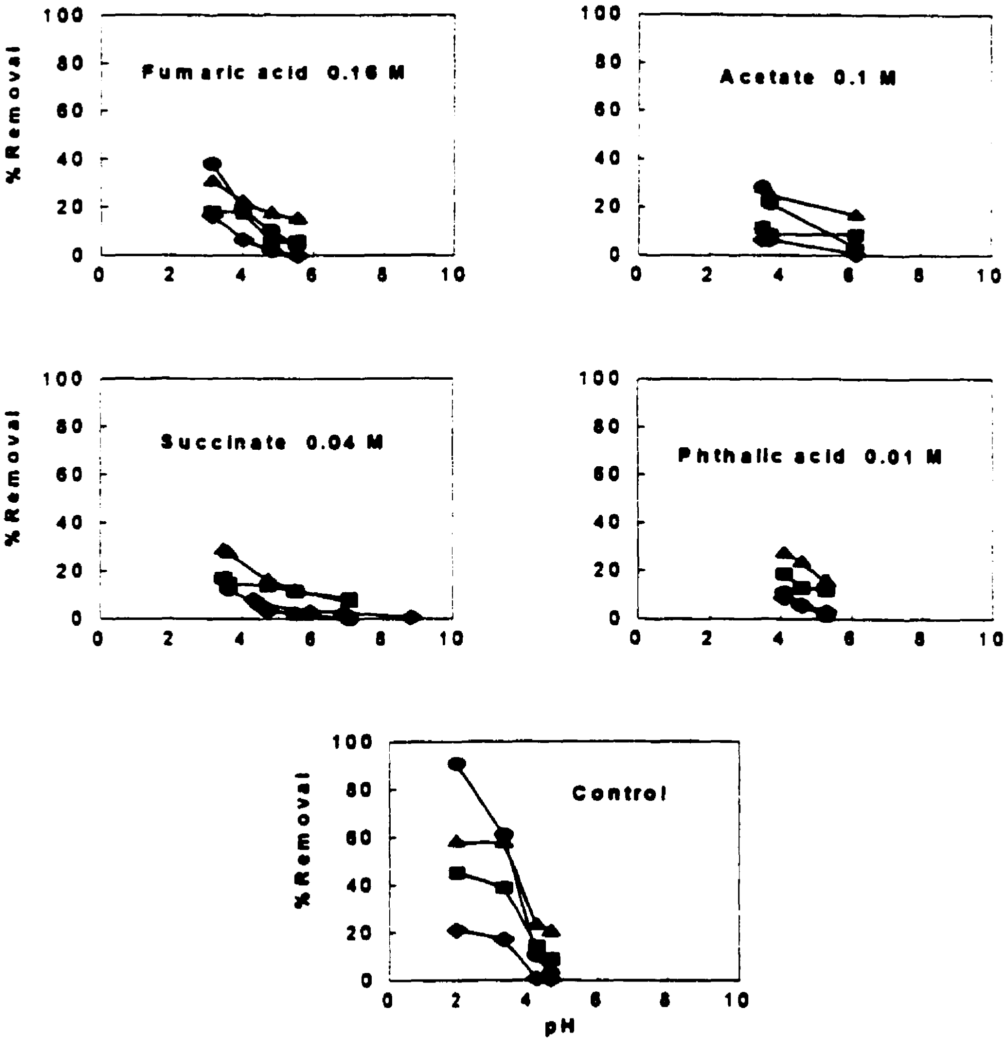

Figure 4.1b Effect of pH on the removal of heavy metals from the clay loam in a batch experiment after 24 h of retention time. Symbol: ◆ Cr, ■ Hg, ▲ Mn and ● Pb.

molecules of tartarate than other metals.

The removal of heavy metals by oxalate at a different pH was not promising and decreased with an increasing pH. At a pH of 3.7, the removal of Cr, Hg, Mn and Pb was 41, 43, 54 and 5.6 %, respectively. The negligible removal of Pb resulted from the formation of insoluble Pb-oxalate complexes on the soil surface.

The removal efficiency of metals by other organic acids such as itaconic, fumaric, pyruvic, acetic, succinic, formic and phthalic acids and/or their salts was low and decreased as the pH increased (Figure 4.1a and 4.1b). The removal of Cr, Hg, Mn and Pb in the presence of pyruvate was 28, 57, 36 and 14%, respectively. These organic acids and their salts did not form strong chelates as compared to citrate and tartarate. The removal of metals in the control experiment using water also decreased with pH. The percentage removal of Cr, Hg, Mn and Pb was found to be 0.3, 10.6, 24 and 4.6%, respectively at pH 4.4.

EDTA and DTPA had removed 99% of Pb (Figure 4.1a). The removal of other metals was pH dependent. EDTA and DTPA are strong chelating agents which can extract macronutrients such as Ca, Mg along with heavy metals due to their high stability constants as compared to organic acids and/or their salts (Anderegg 1977, Perrin 1979). Chromium was the least leachable followed by Hg and Mn. At pH 4.7, the maximum removal of Cr, Hg and Mn by EDTA was 14, 30 and 45%, respectively. The removal of Cr, Hg and Mn by DTPA was 13.5, 18 and 13% at pH 5.4, respectively. As compared to citrate and tartarate, EDTA and DTPA were less effective in removing Cr, Hg and Mn and were incapable of removing extremely toxic Hg. All extractions were carried out in the soil:extractant ratio of 1:25. The extraction may be affected if soil and liquid ratio is changed provided the concentration of extractant remain the same.

4.4.2 Remediation effect on soil properties

It was visually observed that washing with organic acids and/or their salts improved soil structure and the stability of the granular aggregate, as compared to the original soil. The weak organic acids must have directly linked the soil particles according to the soil stabilization theory (Emerson 1959). The heavy metal reacted with organic acids and/or their salts, formed negative charge complexes repelled by the negative charges on the surface of the soil particles, and as a consequence were leached with the liquid fraction.

The soil was acidic in nature (Table 4.1), probably as a result of leaching of macronutrients and their replacement by heavy metals. The soil also demonstrated a low CEC because its exchangeable sites were occupied by heavy metals which are difficult to exchange using $BaCl_2$ solution (Wasay et al. 1998a). However, Ca, Mg and Fe were detected in the leachates of the soil after extraction with citrate and EDTA, but EDTA was found to leach 4 to 5 times more macronutrients than citrate (Table 4.3).

Table 4.3 Soil mineral extraction with remediation using citrate and EDTA

Chemical	Drop in macronutrient content (mg/kg)		
	Ca	Mg	Fe
Citrate	2.00	0.50	0.25
EDTA	10.00	2.50	1.00

4.4.3 Concentration effect on leachability

The removal of heavy metals increased slightly as citrate concentration was increased from 0.04 to 0.1 M (Figure 4.2). Heavy metals removal was found constant with tartarate concentration above 0.2 M. Other organic acids were not tested because of their low removal efficiency. EDTA concentration, varied between 0.006 to 0.08 M, had almost no effect on heavy metal removal rate. This may be due to very strong chelating behavior towards metals. The DTPA removal increased with concentrations varied from 0.008 to 0.08 M for Cr, Hg, Mn and Pb.

4.4.4 The kinetic study

At optimum pH, most of the heavy metals bound to the soil were released in the liquid phase within 24 h and in a single extraction. Residual metals in the soil (C_kt) was plotted as a function of time (Figure 4.3). Reaction rate was determined using two-reaction model for soil (Wasay et al. 1998b). Citrate removed Hg and Pb within 24 h at a level of 89 and 60%, respectively. The removal efficiency of Cr and Mn was only 38 and 48%, respectively, probably because Cr(III) is tightly bound on organic matter and chemisorbed on oxides and silicate clays even at a low pH (McBride 1994). Oxidation of

Cr(III) to Cr(VI) is a difficult operation to carry out in the soil but can occur slowly under flooding condition (Pardue and Patrick 1995). A small fraction of Cr(III) in soils can also be oxidized to chromate at high pH which can be further promoted by Mn oxides (McBride 1994). The removal of Cr, Hg, Mn and Pb by tartarate was found to be 29, 70, 42 and 66% within 12 h, respectively. The maximum release of these metals was obtained within 12 h by EDTA and was 18, 20, 37 and 90%, respectively, but EDTA showed a low affinity for Cr, Mn and Hg. Similarly, the removal efficiency of DTPA was not very good for these metals except for Pb. The maximum removal was reached within 15 h and was found to be 20, 40, 32 and 97% for Cr, Hg, Mn and Pb, respectively, in a single extraction.

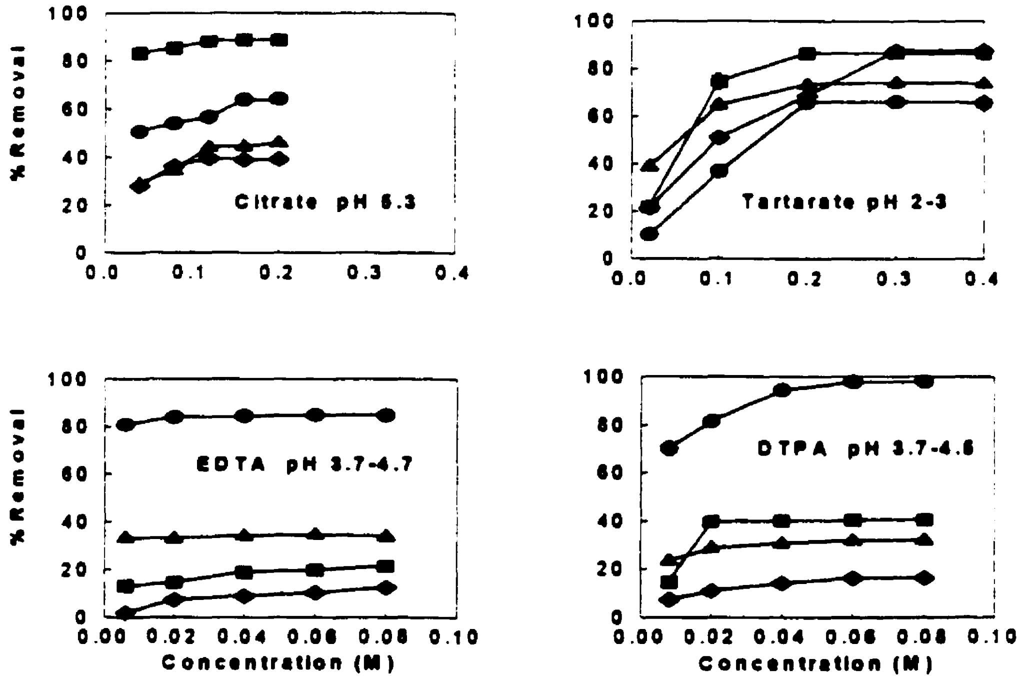

Figure 4.2 Effect of concentration on the removal of heavy metals from the clay loam in a batch experiment after 24 h of retention time. Symbol: ◆ Cr, ■ Hg, ▲ Mn and ● Pb.

4.4.5 Soil clean up criteria

Quebec has guidelines for the use of remediated soil based on three categories, A, B and C (MEFQ 1994). Remediated soil should meet category A criteria before disposal on agricultural land.

After two extractions with tartarate and citrate, the experimental soil respected

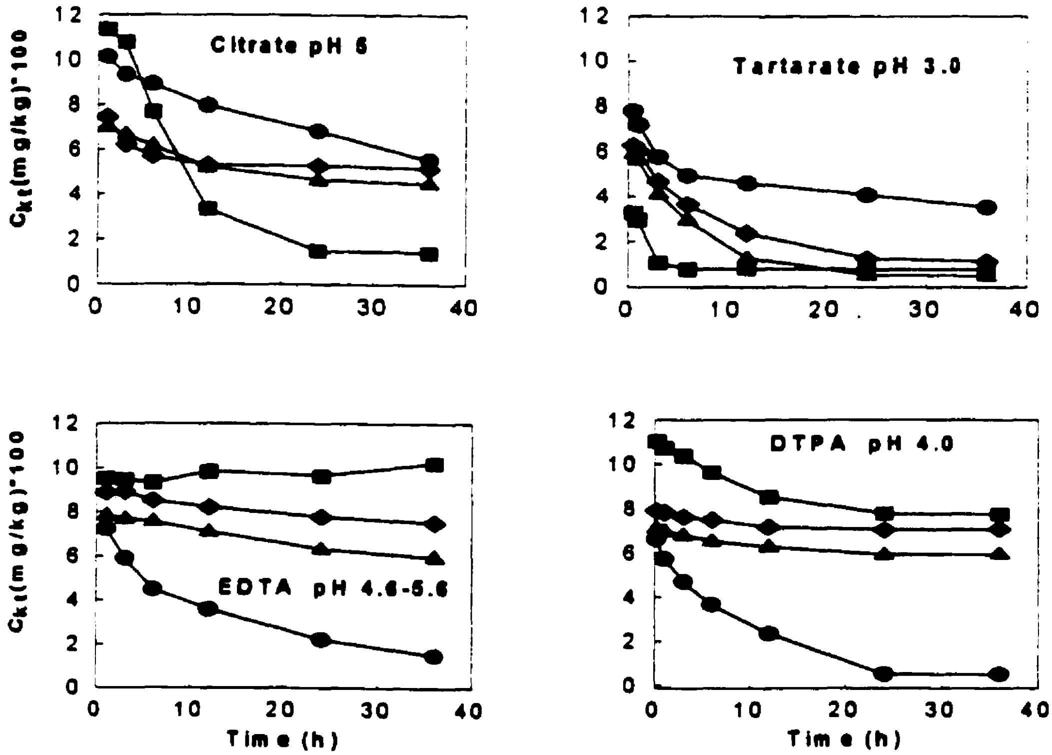

Figure 4.3 Kinetic study for leaching of heavy metals from the clay loam in a batch experiment at different intervals of time. Symbol: ◆ Cr, ■ Hg, ▲ Mn and ● Pb.

the Cr levels required for category B and C, respectively, while the Pb levels respected the category B in both cases. A remediated soil of category A could probably be met for all heavy metals except Hg, after additional two extractions with citrate or tartarate. After two extractions using citrate and tartarate, Hg levels were still at 32 and 62 mg/kg, levels far above the 0.5 mg/kg required to meet category A. EDTA and DTPA only removed enough Pb after two extractions to respect the category A requirements. EDTA and DTPA were ineffective in removing the other heavy metals.

4.4.6 Mechanism of soil and leachate clean up

Aqueous ligands, such as citrate, tartarate, EDTA or DTPA, compete against soil particles for heavy metals absorption. The partitioning of heavy metals between the soil surface groups and a soluble complexing ligand largely depends upon the stability constants described by the following reactions, where $\equiv$S stands for the soil:

$$\equiv\text{S-Metal} \quad + \quad \text{Ligand} \rightleftharpoons \text{Ligand-Metal complex} \quad + \quad \equiv\text{S}^-$$

(Contaminated soil) (Soluble chelate) (Soil surface site)

A chelate (i.e. EDTA) is a complexing ligand that has two or more electron groups which form more than one bond between the metal and the ligand. The multiple bonds form a tight ring structure around the metal ion enabling the chelate to solubilize the heavy metals and leach them from contaminated soils. EDTA and DTPA are sexadentate and octadentate molecules, meaning that they have six and eight donor atoms (z) which can coordinate to a single metal ion to produce five and seven (z-1) chelate rings (Bell 1977). Two and three donor sites are located on the nitrogen atom of EDTA and DTPA, respectively, while the remaining four and five donor sites are negatively charged carboxyl groups (-COOH) which react with metal cations when these sites become deprotonated ($-COOH \rightarrow -COO^- + H^+$). Speciation (release of protons) of EDTA and DTPA depends on pH, with $H_2[EDTA]^{2-}$ or $H_2[DTPA]^{3-}$ being the dominant form in the pH range of 3 to 6 where most heavy metals exist as divalent cations. Chromium, Hg and Mn are so tightly bound to soil surface sites and these chelating agents are not able to form complex with them.

Citrate and tartarate carry three and two negatively charged carboxyl groups ($-COO^-H^+$) which have stronger coordination power (form chelate with metals) than their respective alcoholic OH group [citrate, $HOC(CH_2COOH)_2COOH$] and [tartarate, HOOCCH(OH)CH(OH)COOH]. The coordination of citrate to a metal ion involves two carboxyl groups and one hydroxyl group or alcoholic group (OH), whereas coordination with tartarate occurs between one carboxyl group and two hydroxyl groups. The observed order of effectiveness in chelation is: citrate > tartarate > other organic acid anions. Metal-citrate complexes are more stable than those of the other acid anions, partly because citrate carries three negative charge.

The soil leachate was treated effectively with granular activated carbon (Wasay et al. 1997) and the recovered heavy metal can be recycled in ore processing units.

4.4.7 **Statistical analysis of the results**

The SAS procedures indicated that pH had a highly significant effect ($P < 0.0001$) on remediation level except for Pb. When the salts of weak organic acids were used (particularly for citric acid), higher pH values gave significantly better removal levels. Tartaric acid gave significantly better removal levels at lower pH values except for Pb which gave good removal levels at all pH values. The coefficients of variance (CV) remained under 1.5% for all removal levels for citrate and between 2 to 3% for tartarate.

The contrast between reaction time and metal removal from the soil was significant ($P < 0.05$). Among the heavy metals, Pb residues were significantly higher for all the reaction times during the extraction with salts of organic acids. However, Hg showed the lowest residual of all heavy metals after 12 h of leaching with both citrate and tartarate, whereas residual values for Mn and Cr were intermediate up to 36 h. Lowest residual values after 24 h were obtained for Cr and Hg with tartarate and citrate, respectively.

4.5 Conclusions

The following conclusions can be drawn:

1. Citrate showed high removal capabilities at a pH about 5, as it leached 54, 69, 85 and 96% of Cr, Mn, Pb and Hg, respectively. Tartarate removed 86% of the Pb under a wide pH range of 2 to 7, whereas it removed 41, 54 and 84% of Cr, Mn and Hg, respectively

at a pH 4.3. Both citrate and tartarate were very effective in removing extremely toxic metals such as Hg and Pb.

2. EDTA and DTPA were only effective in removing Pb (99%), but not the other metals, particularly for Hg and Cr (< 30%). Because Hg is extremely toxic even at low concentration, EDTA and DTPA were not capable of adequately detoxifying the contaminated soil.

3. Weak organic acids and/or their salts were found to offer an environmentally friendly remediation technique because an improved soil structure was observed after treatment. Furthermore, they leached significantly less macronutrients, such as Ca and Mg. Strong acids and chelating agents are known to disturb the soil properties by extracting nutrients, destroying their structure and disturbing their biological activity.

4. Four extractions (solid:extractant ratio is 1:25) with citrate and tartarate would allow the polluted soil to meet the category A requirements, for its disposal on agricultural land.

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CONNECTING STATEMENT

The next chapter deals with the remediation of the experimental loam and sandy clay loam polluted by heavy metals in a batch experiments using salts of weak organic acids, EDTA and DTPA. Various parameters such as effect of pH, concentration and time intervals were studied for setting up optimum conditions. Citrate and tartarate as well as oxalate in combination with citrate are very effective in remediating these two experimental soils and comparable with EDTA and DTPA. The results were compared with the Quebec clean up criteria of soils polluted by heavy metals.

This paper has been published in the ***Environmental Technology*****. 1998. 19:369-380 Authors: Wasay, S. A., Barrington, S. and Tokunaga, S.** The contribution of authors are: i) First author carried out entire experimental work and write up of manuscript ii) Second author supervised and technical correction of the work iii) third author provided his analytical laboratory facilities and manuscript correction.

CHAPTER 5

REMEDIATION OF SOILS POLLUTED BY HEAVY METALS USING SALTS OF ORGANIC ACIDS AND CHELATING AGENTS

5.1 Abstract

Various weak organic acids and/or their salts were tested for the remediation of a loam and a sandy clay loam naturally polluted by heavy metals for over three years against two strong synthetic chelating agents (EDTA and DTPA). Among 7 weak organic acids and/or their salts, citrate, tartarate and oxalate were found to effectively remove Cd, Cu, Pb and Zn from the two soils in double extractions, at a wide range of pH. Citrate removed 80 to 99.9% of all four metals within 24 h at pH from 2.3 to 7.5. Tartarate removed 84 to 99.9% of all metals from both soils within 24 h at pH from 2.1 to 6.7. Oxalate alone is not effective in removing metals particularly Pb but with ammonium citrate (1:1 ratio) removed effectively 82, 70, 99.9, and 99.9% for Cd, Pb, Cu and Zn, respectively at pH from 2.6 to 5.8. Zinc met the Quebec A level soil clean up criteria after two extractions whereas Cu and Pb reached the B level. Probably, two more extractions with these weak organic acids and/or their salts are required to meet the A level for the soil clean up criteria for Cd, Cu and Pb. EDTA and DTPA removed 75 to 99.9% of the heavy metals within 24 h at pH from 3.5 to 9.0 for both soils. Citrate and tartarate were found to remove heavy metals from two contaminated soils, as effectively as EDTA and DTPA. But as opposed to EDTA and DTPA, they were removed 80% less macronutrients from the soil and improved its structure. Weak organic acids and/or their salts can therefore be used as an environmentally friendly remediation technique.

Key words: Soil remediation, heavy metals, salts of weak organic acids, EDTA, DTPA.

5.2 Introduction

The technology of soil washing employed to remediate polluted soils and sediments, uses two basic approaches. The first approach immobilizes the heavy metals to minimize their migration. Immobilization is achieved with complexing agents, lime and cementing materials (Anderson and Dee 1994, Czupyrna et al. 1989). This technique has the negative impact of fixing in the soil all essential nutrients besides the heavy metals and, moreover, the heavy metals may slowly leach out as a result of biological activity and/or by acidification of the soil. The second approach involves the transfer of the soil bound heavy metals to the liquid phase by desorption and solubilization. Generally strong acids such as hydrochloric, nitric and sulfuric acids are used as washing solutions, but these solutions can markedly alter the chemical, physical and biological properties of the soil.

Before washing, the soil particles are generally segregated because most of the heavy metals are bound to the finer particles. The soil binding mechanisms of heavy metals are adsorption on Fe/Mn oxides, clay particles, organic matter and $CaCO_3$, or retained as exchangeable ions (Assink 1986). The pH is an important parameter affecting the efficiency of any chemical agent to release heavy metals from the soil. The concentration required of these strong acids depends on the magnitude of contamination. At concentrations of 0.1 to 2.0 M, HCl can remove from 62 to 88% of the soil Pb within 0.5 h (Raghavan *et al.* 1989). In removing heavy metals, HCl is more effective than HNO_3 since it forms soluble metal-chloride complexes (Tuin and Tels 1990).

Synthetic organic chelating agents such as ethylenediaminetetraacetic acid (EDTA) and diethylenetriaminepentaacetic acid (DTPA) remove heavy metals as effectively as strong acids but with less impact on the physical and chemical properties of the soil (Tuin and Tels 1990). Strong chelating agents such as EDTA and DTPA have a greater affinity for soil macronutrients like Ca and Mg besides heavy metals and their metal complexes may be difficult to remove from the aqueous phase by lime precipitation because of the negative charges of metal complexes. Besides this problem, EDTA is stable and remains adsorbed in the soil after extraction, this can make the soil unfit for further use because residual EDTA can slowly leach nutrients from the soil. Therefore, it can also disturb the soil's physical, chemical and biological properties.

Soil organic matter consists of residues of plants, animals and organisms such as actinomycetes and insects. As opposed to the non-humified fraction (weak organic acids), the humified organic matter results from oxidative degradation and consist of compounds such as fulvic and humic acids. Plants and animal tissues contain a large number of acids, which may be released into the soil during decomposition. These can range from simple aliphatic acids to complex aromatic and heterocyclic acids (Huang and Schnitzer 1986). These biochemical compounds have chelating characteristics and have the ability to mobilize heavy metals in soil. Simple aliphatic acids are of special interest as natural chelators because of they are ubiquitous and because many of their hydroxy derivatives are effective solubilizers of metals. The organic acids most effective in forming stable chelate complexes with metal ions are those of the di- and tricarboxylic-hydroxy types, such as citric, tartaric and oxalic acids. Organic acids such as citric, fumaric, malic, malonic and succinic are most abundant in tree root exudates (Smith 1976). These organic ligands undoubtedly enhance the availability of metals for plants and in some instances, they reduce or eliminate the toxicity effect of the metal's ionic forms. The free (hydrated) metal ion is the most toxic, whereas metal-organic acid complexes are less toxic (Leeper 1978).

Organic compounds offer some advantages as extractants over synthetic chelating agents such as EDTA and DTPA or strong acids: i) The residual organic compounds in the soil after cleaning enhance the formation of water-stable aggregates because of the formation of humified organic matter, ii) Compared to EDTA and DTPA for example, organic compounds are less selective for alkaline-earth metals such as Ca, Mg and Fe present in soils. Therefore, organic compounds tend to have less impact on the soil, as the preferred remediation technology requires ligands with low toxicity and a limited impact on the environment.

The ideal complexing agent should compete effectively for heavy metals bound to soil particles. The metal partitioning between the soil surface and the organic complexing agent dependents largely upon the stability constant of the metal-organic complex.

The present study was designed to i) identify organic acids and/or their ammonium (NH_4^+) salts that can potentially be used to remediate soils polluted by heavy metals with limited environmental impact and ii) compare the performance of organic acids and/or their NH_4^+ salts to the synthetic chelating agents EDTA and DTPA.

5.3 Materials and Methods

5.3.1 The experimental soil

The soil samples were obtained from soil contaminated for at least three years. The wet soil samples were air dried, sieved to remove the particles > 2 mm, homogenized and kept in plastic bags for further study. The soils were characterized physically (Table 5.1) and the forms of heavy metal retention were determined using sequential extraction and exchangeable techniques. The sequential extraction was performed using a method described by Fiedler (Fiedler et al. 1994). The different forms of heavy metals retention in these soils and heavy metals bound to the soil surface in exchangeable form are presented elsewhere (Wasay et al 1998 or Chapter 3).

5..3.2 Organic acids and chelating agents

The organic acids and/or their salts and chelating agents used with distilled water were: citric acid monohydrate (0.2 M) and ammonium citrate dibasic (0.2 M), oxalic acid dihydrate (0.2 M) and ammonium oxalate monohydrate (0.2 M), tartaric acid (0.5 M) and ammonium tartarate (0.5 M), acetic acid (0.1 M) and ammonium acetate (0.5 M), itaconic acid (0.2 M), fumaric acid (0.2 M), pyruvic acid (0.1 M) and sodium pyruvate (0.1 M). The chelating agents used were ethylenediaminetetraacetic acid (EDTA) a disodium salt at 0.1 M and diethylenetriaminepentaacetic acid (DTPA) dissolved in a sodium hydroxide solution at 0.1 M.

Dilute solutions of hydrochloric acid (0.1 M) and sodium hydroxide (0.1 M) were used for pH adjustment. The available heavy metals were assessed in a batch test using $MgCl_2$, $CaCl_2$ or $BaCl_2$ (0.1 M), shaken with the soil for 2 h at 20 °C in a temperature controlled shaker (Wasay et al. 1998).

5.3.3 Experiment 1-removal of metals from soil as a function of pH

Remediation of heavy metals from soil was conducted in a batch experiment to find out maximum removal at optimum pH. The control consisted of four series of plastic (polycarbonate) tubes holding 25 ml of distilled water with pH adjusted at various levels between 1.9 and 5.0 with 0.1 M HCl, 0.1 M NaOH was used when the pH becomes too

Table 5.1 Physical and chemical properties of the soil.

Property	Loam	Sandy clay loam
Particle size		
% sand	49.5±.5	53.5±0.9
% silt	30.0±1.2	24.0±1.6
% clay	20.7±0.7	22.7±0.7
Bulk density (g/cm^3)	0.9±0.0	0.9±0.0
Particle density (g/cm^3)	2.0±0.0	2.1±0.1
Total porosity (%)	44.1±0.2	48.0±0.4
k$_s$(cm/s)	1.3±0.1x10^{-4}	4.0±0.2x10^{-4}
EC (mS/cm)-water	8.0±0.2	2.3±0.2
pH-water	5.1±0.1	6.3±0.1
-CaCl$_2$	5.2±0.1	6.0±0.1
CEC (cmol(+)/kg)	4.7±0.0	12.2±0.2
Organic matter (%)	5.4±0.1	7.1±0.1
Heavy metals (mg/kg)		
Pb	2797	1036
Cd	1821	794
Cu	-	926
Zn	-	1029

The ± values is the standard deviation due to experimental error; k$_s$ stands for saturated hydraulic conductivity; EC stands for electrical conductivity and CEC stands for cation exchange capacity.

acidic, otherwise, only HCl was used. Two series received 1 g of the loam while the second two series received 1 g of the sandy clay loam. The treatments consisted of four more series of plastic tubes holding 25 ml of a solution of one of the organic acids and/or its salts or one of the chelating agents with pH adjusted at various levels between 1.4 and 9.7. The pHs were adjusted by mixing different ratios of organic acids and their NH_4^+ salts (in most cases) or with small amounts of 0.1 M NaOH. For chelating agents, the pH levels were adjusted with 0.1 M NaOH and/or with HCl. Again, two series received 1 g of the loam while the second two series received 1 g of the sandy clay loam. The suspensions were shaken for 24 h at 20 °C and centrifuged at 7000 rpm for 20 minutes before withdrawing an aliquot of supernatant. The residue was extracted once more with 25 ml of the organic acids, and/or their NH_4^+ salts solutions, chelating agents or distilled water, as used initially. Both supernatant volumes were analyzed for heavy metals by ICP-AES (ICPS 1000 II, Shimadzu Corp., Kyoto, Japan). The extraction may be affected if soil and extractant ratio is changed provided the concentration of extractant remain the same.

5.3.4 Experiment 2-concentration effect of organic acids and chelating agents

Four series of plastic tubes (two for loam and two for sandy clay loam) were filled with a 25 ml solution containing various concentration of one of the organic acids and their salts or one of the chelating agents at optimum pH for heavy metal removal. Each tube received 1 g of soil and was shaken for 24 h at 20 °C. The mixture in the tube was centrifuged and the supernatant was analyzed by ICP-AES for heavy metals. A single extraction was performed.

5.3.5 Kinetic study

At optimum pH for maximum removal, determined by the previous test, the removal of heavy metals from the soils was measured at different time intervals. Four series of plastic tubes were filled with a 25 ml solution containing either one of the organic acids and/or their NH_4^+ salts or one of the chelating agents at optimum pH level. For four series, two series each received 1 g of loam while the other two series each received 1 g of sandy clay loam. The tubes were shaken at 20 °C between 10 min. to 36 h. Then, each soil sample in duplicate was filtered and the supernatant was analyzed for heavy metals by ICP-AES.

5.3.6 Statistical analysis

The SAS procedures used are: i) One-way analysis of variance or randomized complete block design (RCBD) for pH effect. ii) repeated measures analysis of variance for the rate of leaching study with time (SAS 1990). Heavy metal leaching at different pH was analyzed by SAS for the different treatments or combination of organic acids and/or their salts and water. The SAS analysis of the raw data gives the optimum pH for maximum leaching of heavy metals. The kinetic study of heavy metal leaching was also analyzed by repeated measurement at different time intervals. The SAS analysis of the raw data gives the optimum level of organic acids and/or their salts and water (at optimum pH) for the maximum leaching of the individual metals.

5.4 Results and Discussion

5.4.1 Removal of metals from the soil as a function of pH

The heavy metal removal efficiency was determined at various pHs, for the 7 organic acids and 2 chelating agents (Figure 5.la, 5.lb and 5.1c). The extraction efficiency was highly pH dependent for the organic acids and independent for EDTA and DTPA. For the loam, citrate removed 96% of Cd at pH 2.3 to 7.3, and 83% of Pb at pH 5.2 to 6.4. Tartarate removed 92 to 97% of Cd at pH 2.1 to 4.5 and 88% of Pb at pH 2.1 to 6.7. For the sandy clay loam, citrate removed 100% of Cd at pH 2.3 to 7.5, 88% of Cu at pH 2.3 to 6.3, 80% of Pb at pH 5.2 to 6.4 and 90 to 96% of Zn at pH 2.3 to 6.3. For the same soil, tartarate removed 95 to 99.9% of Cd at pH 2.1 to 5.5, 88 to 97% of Cu at pH 2.1 to 4.0, 84 to 87% of Pb at pH 2.1 to 6.7 and 90 to 98% of Zn at pH 2.1 to 5.5. Oxalic acid effectively removed Cu and Zn from both soils but not Pb and Cd at pH 1.4 to 7.0. Heavy metals from contaminated soil were solubilized by citrate, tartarate or oxalate in the form of negatively charged water soluble metal-citrate, -tartarate or -oxalate complexes (Farrah and Pickering 1978). The extraction of Pb and Cd was improved by using a mixture of the citric acid and ammonium oxalate in a 1:1 ratio at an optimum pH of 5.8 (Figure 5.lb). The removal of Cd, Cu, Pb and Zn by this mixture was found to be 82, 99.9, 70 and 99.9%, respectively. The other organic acids such as itaconic, fumaric, pyruvic and acetic acids (Figure 5.1 a, 5.1 b and 5.1 c) were not effective

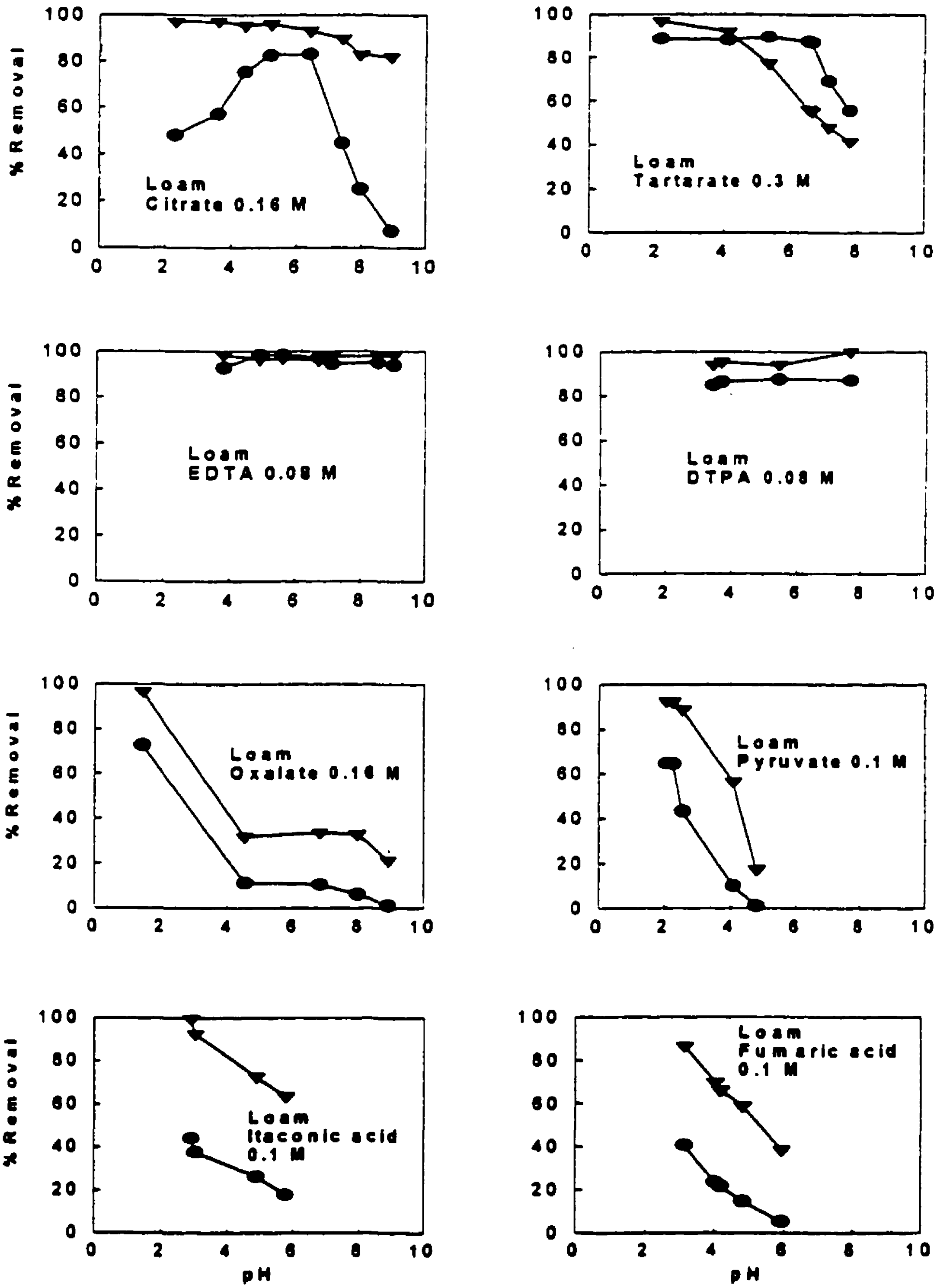

Figure 5.1a Effect of pH on the removal of heavy metals from the soil after 24 h. ▼ Cd and ● Pb.

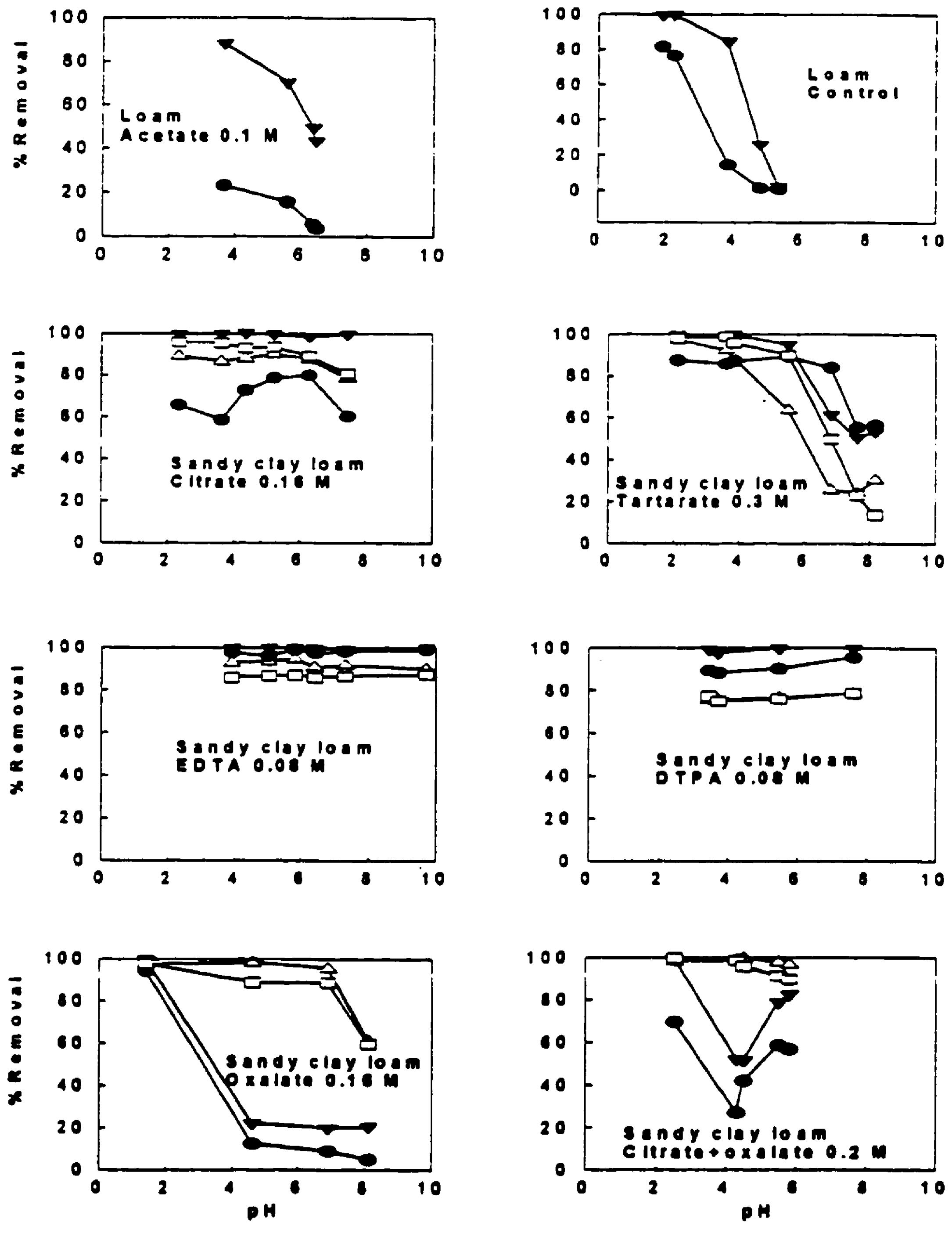

Figure 5.1b Effect of pH on the removal of heavy metals from the soil after 24 h. ▼ Cd, Δ Cu, ● Pb and □ Zn.

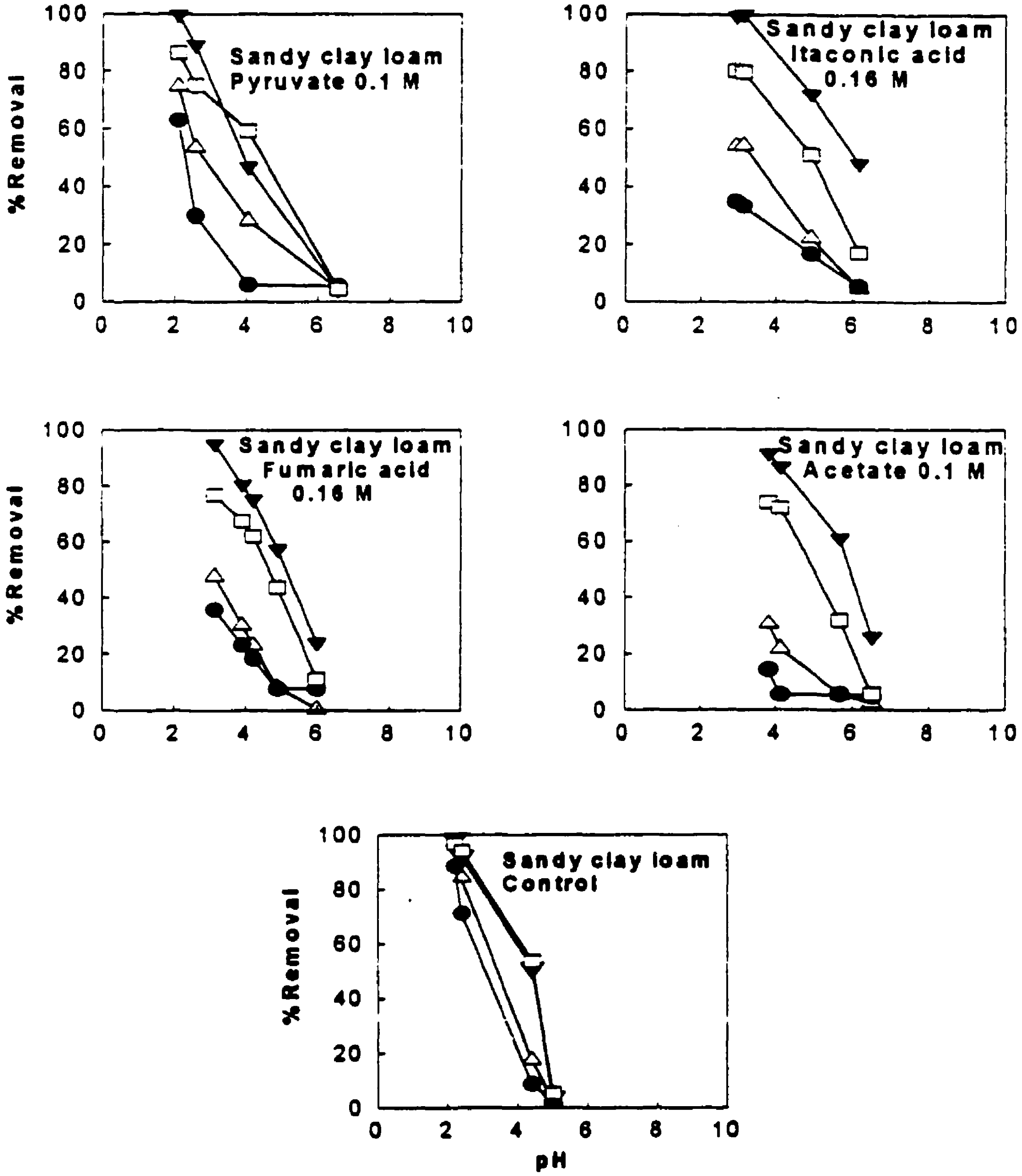

Figure 5.1c Effect of pH on the removal of heavy metals from the soil after 24 h. ▼ Cd, Δ Cu, ● Pb and □ Zn.

in removing a significant quantity of heavy metals at a pH above 2 because of the lower stability constant of their metal complexes as opposed to those of citrate and tartarate.

Zinc met the Quebec soil clean up criteria of level A after two extractions whereas Cu and Pb meet B levels (MEFQ 1994). Probably, two more extractions with these weak organic acids and/or their salts are required to meet the level A of soil clean up criteria for Cd, Cu and Pb.

5.4.2 Concentration effect

Concentration effect of three organic acids such as citrate, tartarate and oxalate as well as EDTA and DTPA on heavy metal extraction from soils has been studied (Figure 5.2). For the maximum removal of Cd and Pb from the loam, the minimum concentration of citrate was found to be 0.04 M for Cd and 0.12 M for Pb, whereas for tartarate, 0.2 M was required for Cd and Pb. The minimum concentration of EDTA and DTPA was 0.02 and 0.06 M, respectively, for maximum removal of these metals. For the sandy clay loam, the minimum concentration of citrate was 0.03 M for Cd and Cu and 0.16 M for Pb and Zn for maximum removal. The minimum concentration of tartarate was 0.25 M for all 4 metals. The sandy clay loam had a lower concentration of metals, thereby holding heavy metals more tightly and requiring more extractants particularly for Pb, as compared to the loam. Similarly, a higher concentration of EDTA and DTPA (0.08 M) was required for maximum removal of the heavy metals from the sandy clay loam as compared to that of the loam.

Clean up of contaminated soil is directly related to the initial metal concentration in the soil as well as the presence of inorganic and organic compounds. For the same treatment imposed, sites contaminated by small amounts of heavy metals are often just as difficult to remediate as grossly contaminated sites because of highest binding energies associated with low sorption densities (Reed et al. 1995). Also because soils have multiple types of surface sites, small quantities of metals would be preferentially adsorbed by the sites with the highest binding energies, making their subsequent release difficult. This phenomenon is illustrated especially for Pb leaching of sandy clay loam.

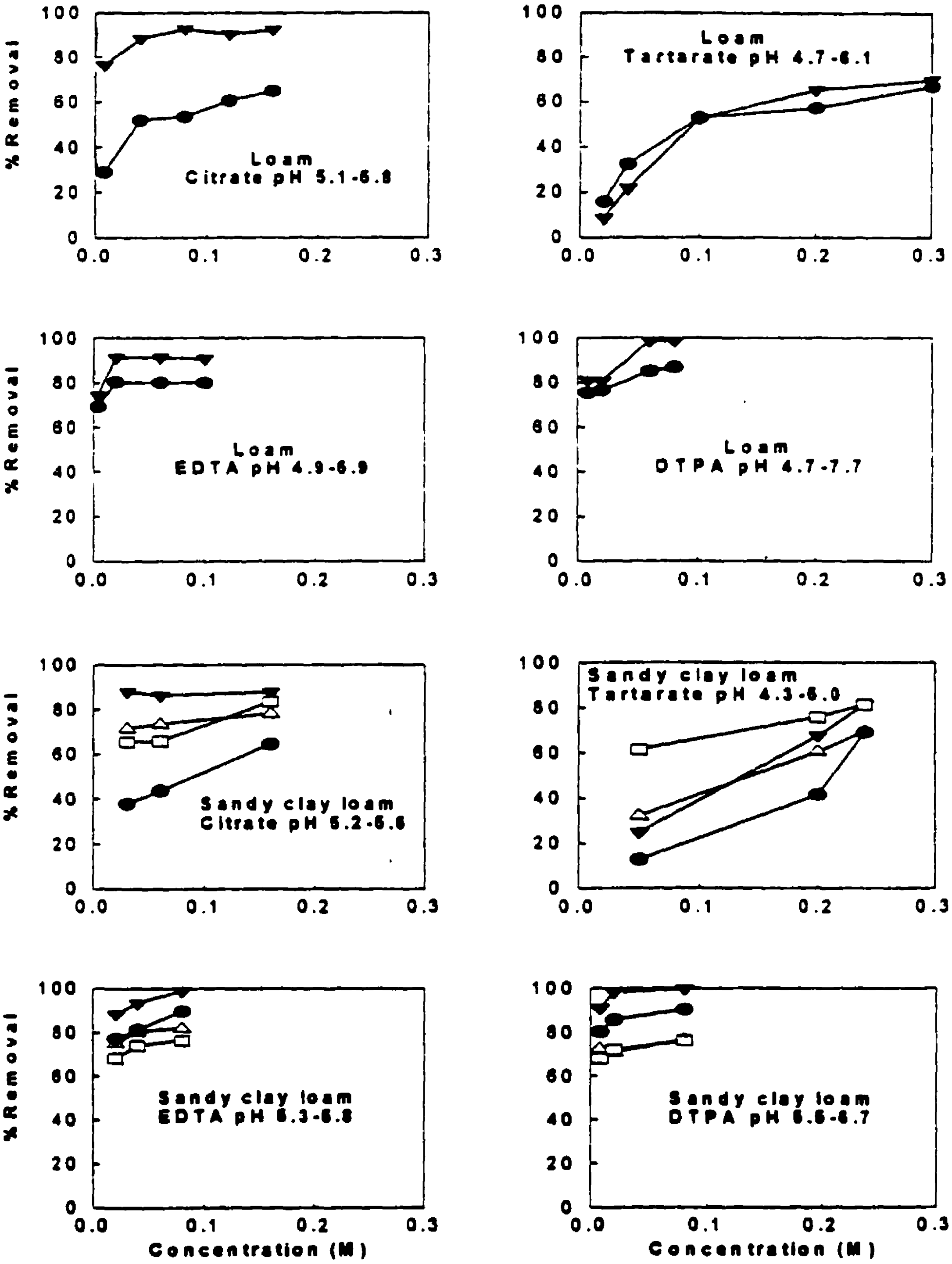

Figure 5.2 Effect of concentration on the removal of heavy metals from the soil after 24 h. ▼ Cd, Δ Cu, ● Pb and □ Zn.

5.4.3 Kinetic study

For the three organic acids and chelating agents, the kinetic experiments indicate that in most cases equilibrium was attained within 24 h in both soils for maximum heavy metal removal (Figure 5.3a and 5.3b).

A summary of the optimum remediation parameters for the removal of heavy metals from both soils are provided in Table 5.2. The order of desorption efficiency was

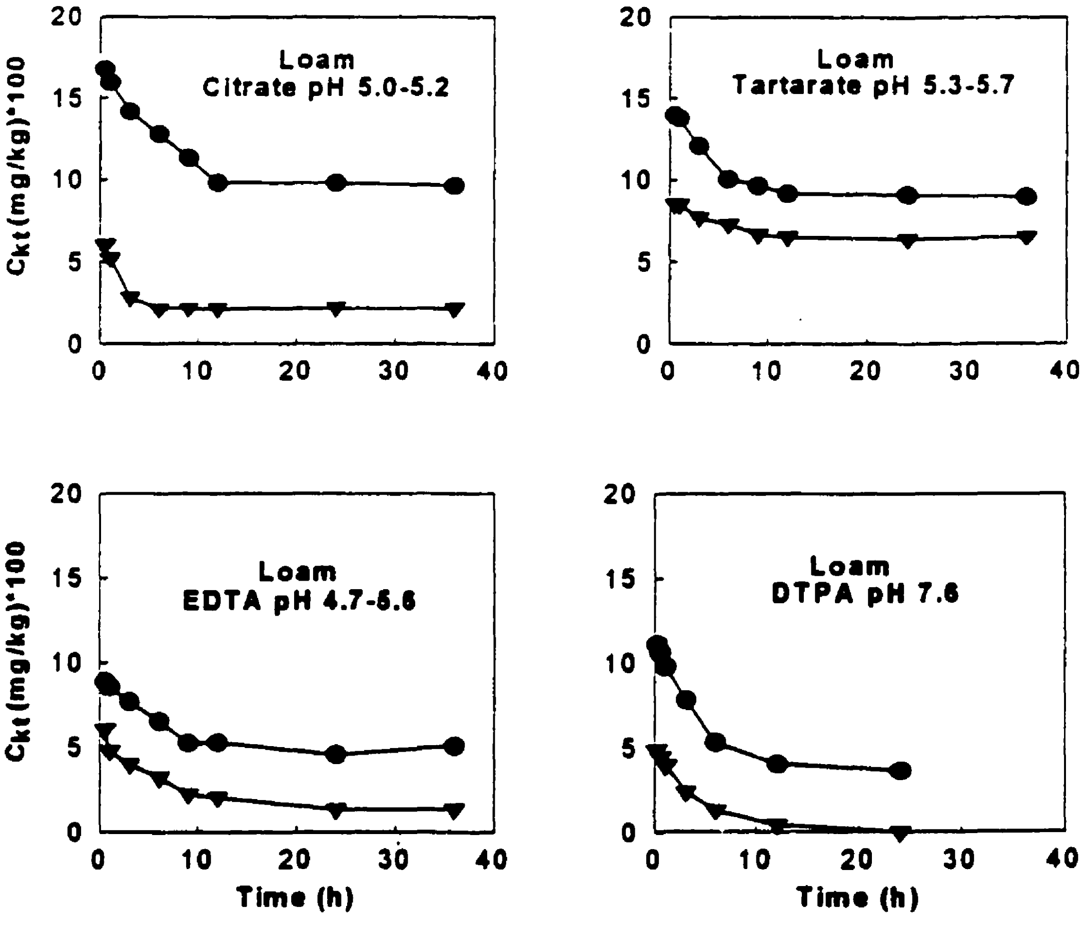

Figure 5.3a Kinetic leaching of heavy metals from the loam. ▼Cd and ● Pb.

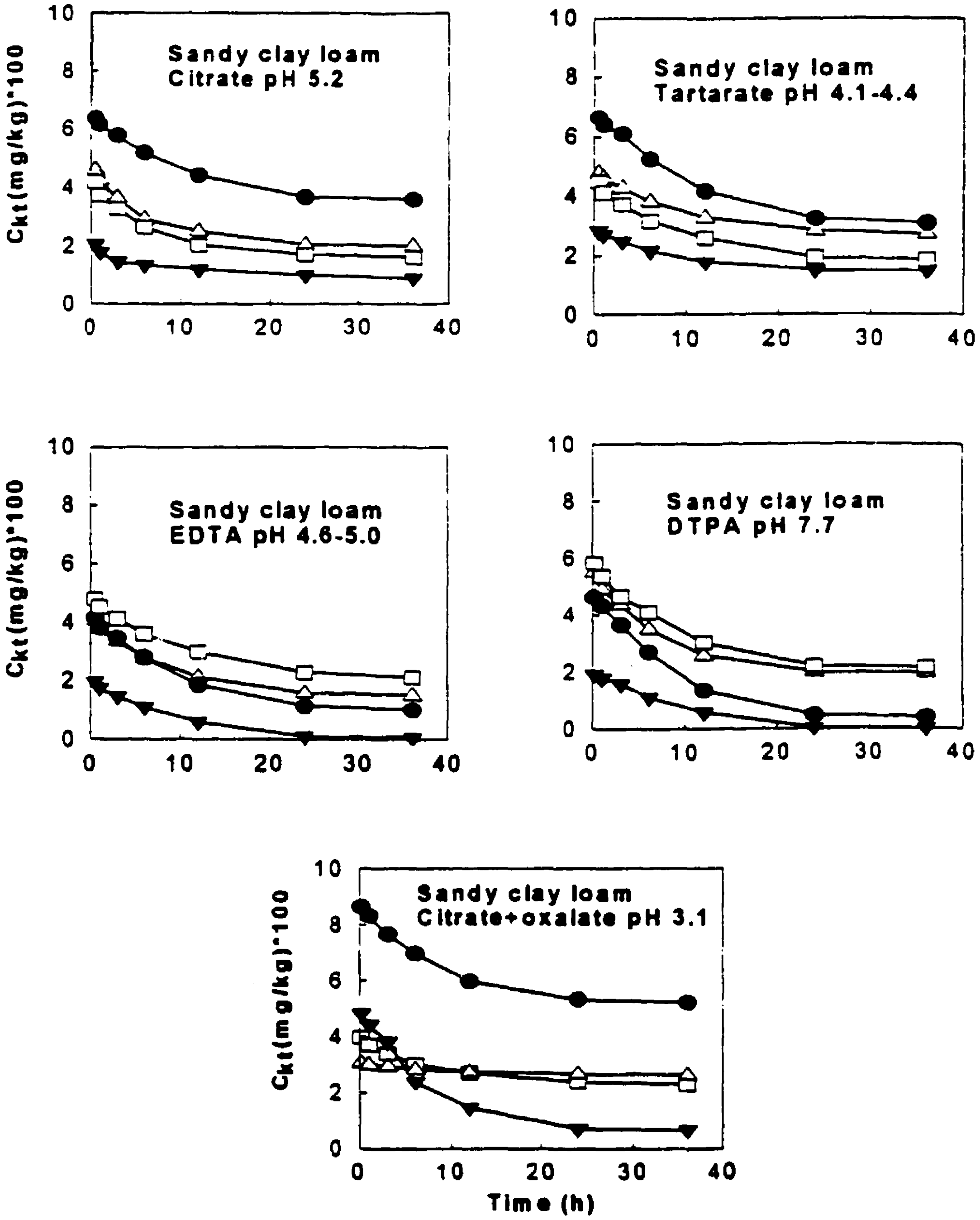

Figure 5.3b Kinetic leaching of heavy metals from the sandy clay loam. ▼ Cd, Δ Cu, ● Pb and □ Zn.

Table 5.2 Optimum conditions for the remediation of soil polluted by heavy metals

Extractant	Leachablity parameter	Loam		Sandy clay loam			
		Cd	Pb	Cd	Cu	Pb	Zn
Citrate	pH	2.3-7.3	5.2-6.4	2.3-7.5	2.3-6.3	5.2-6.4	2.3-6.3
	Con. (M)	0.04-0.16	0.12-0.16	0.03-0.16	0.03-0.16	> 0.16	> 0.16
	Time (h)	6	9	12	24	24	12
	%Removal	96	83	100	88	80	90-96
Tartarate	pH	2.1-4.5	2.1-6.7	2.1-5.5	2.1-4.0	2.1-6.7	2.1-5.5
	Con. (M)	0.20-0.40	0.20-0.40	0.25-0.4	0.25-0.4	0.25-0.4	0.25-0.4
	Time (h)	9	6	12	24	24	24
	%Removal	92-97	88	95-99.9	88-97	84-87	90-98
EDTA	pH	3.8-9.0	3.8-9.0	3.9-9.7	3.9-9.7	3.9-9.7	3.9-9.7
	Con. (M)	0.02-0.08	0.02-0.08	> 0.08	> 0.08	0.08	> 0.08
	Time (h)	24	24	24	24	24	24
	%Removal	97	95	99-100	90-95	96-99	86-87
DTPA	pH	3.5-7.7	3.5-7.7	3.5-7.6	3.5-7.6	3.5-7.6	3.5-7.6
	Con. (M)	0.06-0.08	0.06-0.08	0.08	0.08	> 0.08	0.08
	Time (h)	24	24	24	24	24	24
	%Removal	99.8	97.0	98-99.9	76-79	88-96	75-79
Citrate+ Oxalate	pH	-	-	5.5-5.8	4.30-5.8	5.5-5.8	2.6-5.8
	Con. (M)	-	-	0.20	0.20	0.20	0.20
	Time (h)	-	-	24	24	24	24
	%Removal	-	-	79-82	96-99.9	70	90-99.9

Con.:concentration; std. deviations for pH and concentration effects on remediation are < ± 1 whereas < ± 6 for kinetic study.

Cd ~ Zn > Cu > Pb, which is the reverse of that obtained for soil adsorption (Lo et al. 1992). EDTA and DTPA were effective in removing heavy metals from both soils. These chelating agents have strong binding capacities and their stability constants with metals are higher than those of the organic acids and/or their salts (Anderegg 1977, Perrin 1979). EDTA is so strong in fact that it effectively extracts macronutrients from the soil such as Ca, Mg and Fe species, which are important for plant growth. EDTA extracted 4 to 5 times more nutrients than citrate (Table 5.3). Therefore, these chelating agents have negative side effects compared to salts of weak organic acids. EDTA is stable, has a low biodegradation, and can be adsorbed to a significant extent on the soil particles (Tokunaga 1996, Bourg 1988). EDTA remains longer in the soil and as a result, can further leach out nutrients keeping the soil unfit for plant growth. Washing with the salts (NH_4) of organic acids was found to improve the soil structure. Organic materials remaining after soil washing, must have formed direct links with the soil particles forming stable aggregates (Emerson 1958). Ammonium salts of weak organic acids are preferred because slow release nitrogen fertilizer and leaching problem may be reduced if

Table 5.3 Drop in soil macronutrient level after remediation

Soil	Chemical	Macronutrient content (mg/kg)		
		Ca	Mg	Fe
Loam	Citrate	3.050	1.200	0.350
	EDTA	14.800	5.000	1.420
Sandy clay loam	Citrate	144.001	14.003	20.010
	EDTA	471.004	40.020	68.003

Note: This drop in macronutrient levels corresponds to the amount of Ca, Mg and Fe leached out from soils using either citrate or EDTA.

NH_4-N (ammonium nitrogen) compounds are used instead of NO_3-N (nitrate nitrogen) compounds. But increasing nitrification may lead to soil acidification which reduces leaching loss of nitrate because maximum removal of heavy metals is obtained at pH about 5. Excess amounts of ammonium nitrogen can be collected together with heavy metals in the leachate or drainage system of soil washing processes.

5.4.4 Statistical analysis of the results

The effect of pH on soil remediation is highly significant ($P < 0.05$) for both soils. For the loam and ammonium citrate, higher pH values gave significantly higher removal levels. Similarly, lower pH values with citric acid removed more Cd, whereas for Pb, removal levels were higher only when ammonium citrate was used. Tartaric acid gave significantly higher removal levels at lower pH values (2.1 to 4.1) for Cd, whereas for Pb, good removal levels were obtained with ammonium tartarate at pH 5.3. Coefficients of variance (CV) for heavy metal removal for the loam ranged between 0.322 to 0.517% for citrate and between 0.362 to 2.340% for tartarate. With the sandy clay loam, all pH values gave significantly higher removal levels for Cd by either ammonium citrate or a 1:1 mixture of citric acid and NH_4^+ salt. Removal of Cu and Pb was highest when ammonium citrate was used, whereas Zn removal was significantly higher at both lower and higher pH levels (with the use of ammonium citrate and a mixture of citric acid and NH_4^+ salt). For tartarate, significantly higher levels of Cd and Cu were removed at a lower pH (< 3.8), whereas significantly higher levels of Pb and Zn were removed with ammonium tartarate at a pH of 5.5, and in a 1:1 mixture of tartaric acid and ammonium tartarate (pH 3.6). The CV ranged between 0.298 to 0.762% for citrate and 0.435 to 1.442% for tartarate.

The contrast between reaction time and metal removal from the soil was significant ($P < 0.05$). For the loam, Pb residual was significantly higher than Cd for all reaction times during the extraction with ammonium citrate and tartarate. For the sandy clay loam, Pb residual was highest followed by Cu, Zn and Cd, at all reaction times.

5.5 Conclusions

1. Out of the 7 organic acids tested, only citrate, tartarate and oxalate were able to remove, within 24 h, between 80 to 100% of the heavy metals (Cd, Cu, Pb, Zn) held by the two soils. In comparison, the synthetic chelating agents EDTA and DTPA removed between 75 to 99.9% of the heavy metals along with some macronutrients such as Ca and Mg. Zinc levels met the Quebec soil clean up A criteria after two extractions whereas Cu and Pb levels met criteria B. Probably, two more extractions with these weak organic acids and/or their salts are required to meet the soil level A clean up criteria for Cd, Cu and Pb. EDTA is less prone to biodegradability and it is adsorbed during soil cleaning, but it may reduce the amounts of soil macronutrients available for plants. The organic acids or their salts are also adsorbed during the soil cleaning process, thereby improving the structure of the soil and enhancing the formation of stable granular aggregates.
2. Soils with low concentrations of heavy metals are the hardest to remediate because the heavy metals are preferentially adsorbed by the sites of highest binding energies, which require higher concentration of organic and chelating compounds for remediation.
3. Salts of weak organic acids (citrate and tartarate) were less disturbing for the remediation of soils polluted by heavy metals because they enhanced soil aggregation and leached only small amounts of macronutrients from the soil (80% less compared to EDTA and DTPA).

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CONNECTING STATEMENT

Chapter 6 deals with the rate of leaching of heavy metals using a two-reaction (fast and slow leaching) model. The kinetic data obtained from the previous two chapters (4 and 5) for the clay loam, the loam and the sandy clay loam, were used for determining rate constants for the leaching of heavy metals with citrate, tartarate and EDTA at constant pH, temperature, concentration of extractants and shaking speed.

This paper has been submitted for publication in the *Journal of Environmental Engineering*. 1998. Authors: Wasay, S. A., Barrington, S., Prasher, S. O and Tokunaga, S The contribution of authors are: i) First author carried out entire experimental and computer modeling and write up of manuscript ii) Second author supervised and technical correction of the work iii) Third author suggested first author to use certain computer program for the two-reaction model parameters iv) Fourth author provided his analytical laboratory facilities and manuscript correction.

CHAPTER 6

KINETICS OF HEAVY METAL LEACHING FROM POLLUTED SOILS USING CITRATE, TARTARATE AND EDTA

6.1 Abstract

The rate of leaching of heavy metals from soils with citrate, tartarate and EDTA was determined. A two-reaction (fast and slow) model was used to describe the rate of leaching at a constant pH and temperature. The fast and the slow leaching rates of metal were assumed to be irreversible and reversible, respectively and both were of the first order. The fast leaching rate constant (k_1) is in the order of Cd > Zn > Cu > Pb > Mn > Hg > Cr. This resulted from the release of the exchangeable fraction of metals especially Cd and Zn followed subsequently by metals such as Cu, Pb, Mn, Hg and Cr tightly bound to oxides and the organic fractions. If the slow reacting fraction (α_0) is greater than 0.5, the main component of metals in the soil react slowly and as a consequence, metal is released slowly. If the value of α_0 is found to be less than 0.5, the leaching rate is fast. The value of the quasi-equilibrium constant m_1 was much smaller for Cd, Cu, Pb, Zn and Hg (< 0.6 l/kg), whereas for Mn and Cr, it had a higher value. Values of m_1 for Cr and Mn in clay loam were obtained between 1.1 to 4.5 with citrate and EDTA, while lower values were obtained with tartarate, because of the low pH extraction. This higher values of m_1 indicated difficulty in removing Cr and Mn from the clay loam.

6.2 Introduction

When designing a remediation facility, the leaching rate of heavy metals from soils is an important factor which determines the size of the leaching unit and the total cost of remediation. Kinetics of heavy metals desorption from soils have been investigated to a limited extent (Griffin and Burau 1974, Kuo and Mikkelsen 1980, Jopony and Young 1987, Bowman et al. 1981) while adsorption and desorption kinetics in soil systems have been reviewed (Sparks 1986). Desorption kinetics of plant nutrients, such as nitrogen, phosphorus and potassium, have been studied extensively. A rigorous mathematical treatment of the kinetics of chemical reactions and diffusion in soils has been presented (Sposito 1994). In most research work, desorption of heavy metals from soils was carried out in a chemical environment close to field soil conditions or in a solution that was indicative to plant-uptake. A two-reaction model was developed for the determination of leaching rate of heavy metals from contaminated soils using a strong acid (HCl) solution as an extractant with vigorous mixing (Tuin and Tels 1990).

In the present study, a two-reaction model was used to determine the leaching rate of heavy metals from three naturally contaminated soils using citrate, tartarate and EDTA as extractants. The rate of leaching was determined at a constant temperature, pH, concentration of extractants and shaking speed of 100 rpm.

6.3 Theory of Two-reaction Kinetic Model

Different kinetic models have been used to determine the extraction of various ions from soils (Sparks 1986). A first order rate equation has been used to describe desorption of phosphate and boron (Griffin and Buran 1974, Griffin and Jurinak 1974). It was found that the desorption data could be fitted by two or three different slopes. The different first-order reactions were ascribed to the desorption of ions from two discrete types of binding sites. The fast desorption sites for K^+ was attributed to external surface sites on the inorganic and organic phases of the soil while the slow desorption of K^+ was attributed to the less accessible sites of organic matter and the interlayer sites of the 2:1 clay minerals. A two-rate reaction model is explained by the adsorption of metal on specific and non-specific soil sites and has been useful in studying metal behavior (Selim and Amacher 1997). The less and the more labile fractions of soils minerals are the most important metal reservoirs (Wasay et al. 1998a or Chapter 3) and can explain the actual processes occurring in the real soil-extraction system.

Two different kinetic patterns were found to explain the removal of heavy metals from soils. The first leaching pattern is very rapid followed by a second much slower leaching. Thus, the leaching of heavy metals from contaminated soils using citrate, tartarate or EDTA, at constant pH, temperature and concentration can be represented by the following two simultaneous reactions (6.1) and (6.2):

$$\equiv S_1\text{-O-M} + \text{O-A} \xrightarrow{k_1} \equiv S_1\text{-O-A} + \underset{\text{(soluble)}}{\text{M-O complex}} \qquad (6.1)$$

$$\equiv S_2\text{-O-M} + \text{O-A} \xrightleftharpoons[k_3]{k_2} \equiv S_2\text{-O-A} + \underset{\text{(soluble)}}{\text{M-O complex}} \qquad (6.2)$$

where $\equiv S_1$-O and $\equiv S_2$-O represent active soil surface binding sites for fast reaction (6.1) and the slow reaction (6.2), respectively. O-A represents organic acids (A represents hydrogen ion of the organic acid which is replaced by a salt (Na^+ or NH_4^+) of an organic acid at higher pH), M is the heavy metal attached to the soil surface site, M-O represents the soluble metal-organic complex and k_1, k_2 and k_3 are reaction rate constants.

Reaction (6.1) is fast compared to the forward slow reaction (6.2) ($k_1 > k_2$) and is irreversible. It represents the exchange of metals bound to surface sites of various fractions of soils (exchangeable, Fe/Mn-oxides, organic matter) which were mobilized by citrate, tartarate or EDTA in the form of complexes. The citrate, tartarate or EDTA concentration is much greater than the metal that re-adsorption of heavy metals to these sites is negligible. Reaction (6.2) is slower and reversible. In contrast to the multiple reactions models (Griffin and Burau 1974, Jardine and Sparks 1984, Selim and Amacher 1997), the two-reaction model involves coupling. Therefore, metal-complexes in solution formed by reaction (6.1) can also be re-adsorbed to the soil according to the backward reaction (6.2). Both forward reactions are pseudo first-order and the backward reaction is also considered to be pseudo first-order for the sake of simplicity. Besides, k_1, k_2 and k_3, a fourth parameter is required to describe the heavy metals reaction according to reactions (6.1) and (6.2). It was assumed that a fraction α_0 reacts slowly (reaction 6.2) while a fraction ($1-\alpha_0$) reacts quickly (reaction 6.1).

The mathematical rate of the two-reactions can be expressed as follows:

$$-R_a = k_1 * C_{ka} \qquad (6.3)$$

$$-R_b = k_2 * C_{kb} - k_3 * C_\ell \qquad (6.4)$$

where R_a and R_b represent rates of leaching (mg/kg/h) according to reaction (6.1) and (6.2), respectively, k_1 and k_2 represent leaching rate constants (h^{-1}), k_3 represent leaching rate constant (kg liquid/kg soil/h), C_{ka} and C_{kb} are heavy metal concentrations (mg/kg) in the soil reacted according to reactions (6.1) and (6.2), respectively and C_ℓ is the heavy metal concentration (mg/l) in the leachate.

If the initial total heavy metal concentration in the soil is C_k0, then α_0 is the fourth parameter:

$$C_{ka}0 = (1-\alpha_0)*C_k0 \tag{6.5}$$

$$C_{kb}0 = \alpha_0*C_k0 \tag{6.6}$$

where α_0 is defined at t = 0 only.

A very slow extraction of metals from the soil can take place even after 24 h (Bowman et al. 1981, Lion et al. 1982). Nickel desorption from Fe/Mn-oxides and organic matter surface takes several days with EDTA and Cd desorption also goes on even after 96 h. Nevertheless, it can be assumed that a quasi-equilibrium state is reached after 36 h of extraction as observed with citrate, tartarate and EDTA solution at constant concentration, pH and temperature (Wasay et al. 1998b and 1998c).

At quasi-equilibrium, ($-R_b = 0$ and C_{ka} eq = 0 and equation (6.4) is reduced to:

$$k_3 = \frac{k_2 * C_{kb}eq}{C_\ell eq} \tag{6.7}$$

where $C_{kb}eq$ represents the total heavy metal concentration in the soil after 36 h of extraction and $C_\ell eq$ represents the heavy metal concentration in the leachate after 36 h of extraction. A linear equilibrium relationship is adopted as follows:

$$C_{kb}eq = m* C_\ell eq + d \tag{6.8}$$

where m represents a quasi-equilibrium constant that depends on the pH value in the extraction process and the term d can be considered as a residual heavy metal concentration that cannot be removed by the citrate, tartarate and EDTA solutions. Equations (6.7) and (6.8) can be combined and re-written as follows:

$$k_3 = k_2 * [m + \frac{d}{C_\ell eq}] = m_1 * k_2 \qquad (6.9)$$

The following mass balance equation for the heavy metals in the system is:

$$M_k0 * C_k0 + M_\ell 0 * C_\ell 0 = M_k(t)\ [C_{ka}(t) + C_{kb}(t)] + M_\ell(t) * C_\ell(t) \qquad (6.10)$$

where M_k0 and $M_\ell 0$ are the mass of soil and extraction liquid at time t = 0, respectively and $M_k(t)$and $M_\ell(t)$ are the mass of soil and extraction liquid at time t, respectively.

In a batch experiment, the change per unit time in $M_k(t)$ and $M_\ell(t)$ can be considered insignificant even if the soil weight is increased slightly from the adsorption of citrate and tartarate, whereas soil weight slightly decreases with EDTA extraction due to loss of nutrients. The soil to extraction solution ratio is considered constant for citrate and tartarate at different intervals of time (1, 12 and 24 h), the ratio can be expressed as follows:

$$\frac{M_k(t)}{M_\ell(t)} = \text{constant} = [\frac{Mk}{M_\ell}]_{mean} \qquad (6.11)$$

The final expression for the total metal concentration in the soil as a function of time is:

$$C_k(t) = C_k 0[\frac{(k_2 - k_1)}{(k_2 A - k_1)}] * \{[\alpha_0 + \frac{k_1(1 - 1/A)}{(k_2 - k_1)}]e^{-k_2 A t} + [1 - \alpha_0]e^{-k_1 t}\}$$

$$+ (1 - 1/A)C_k 0 + (1 - 1/A)\frac{M_\ell}{M_k} * C_\ell 0(1 - e^{-k_2 A t}) \qquad (6.12)$$

where $A = 1 + \frac{M_k}{M_t} * \frac{k_3}{k_2} = 1 + \frac{M_k}{M_t} * m_1$

The three parameters k_1, k_2 and α_0 can be determined by curve fitting the data points [$t(h^{-1})$, $C_k(t)$ (mg/kg)] to equation (6.12) using SigmaPlot 2.01x, Window and the results can be re-conformed by another program Curve Expert 1.3x, Window.

6.4 Materials and Methods

6.4.1 Soil characteristics

Three soils naturally polluted by heavy metals were characterized for their texture, density, saturated hydraulic conductivity, organic matter, pH, heavy metal content and retention form (Wasay et al. 1998a or Chapter 3).

6.4.2 Kinetic procedure

At optimum leaching pH and at a constant temperature of 20 °C (Wasay et al. 1998b and 1998c), the removal of heavy metals from soils was measured at different time intervals. Six series of plastic tubes were filled with a 25 ml solution containing one of the extractant, citrate, tartarate or EDTA. Of the six series, two series each received 1 g of sandy clay loam, two series each received 1 g of loam while the other two series each received 1 g of clay loam. The tubes were shaken at 20 °C for 10 min to 36 h. Then, each soil sample in duplicate was filtered and the supernatant was analyzed for heavy metals by ICP-AES.

6.5 Results and Discussion

The data of the kinetic experiments were fitted to equation (6.12) of the two-reaction model and two different kinetic patterns were found to govern the leaching of heavy metals from soils using citrate, tartarate and EDTA solutions. For the sandy clay loam using citrate, tartarate and EDTA, the leaching of Cd, Cu, Pb and Zn is shown in Figure 6.1a and 6.1b. For the loam and the clay loam using citrate, tartarate and EDTA as extractants, the kinetic patterns were similar to that of the sandy clay loam (Figure 6.1b, 6.1c and 6.1d). The two-reaction model gave a good fit for the experimental data in all cases for reaction time up to 36 h. The values of the model parameters m_1, k_1, k_2 and α_0 are given in Table 6.1. It is clear that Cd leached faster (k_1 is higher) with all three extractants (citrate, tartarate and EDTA) followed by Zn, Cu, Pb, Mn, Hg and Cr at 20 °C. Slower rate of leaching (k_2) followed the same order observed with the faster reaction. This resulted from the release of the exchangeable fraction especially Cd and Zn and then, the release of more tightly bound metals such as Cu, Pb, Hg, Mn and Cr bound to oxides and the organic fractions. A general relationship is difficult to establish between α_0 and the metal fractions in the soil, determined by sequential extraction (Wasay et al. 1998a). However, a rough trend was obtained between α_0 and the metal (Cd, Zn, Cu, Pb, Mn and Cr) fractions in the exchangeable, Fe/Mn oxides and organic matter. Cadmium showed lower value of α_0 and this value increased in the order of Zn < Cu < Pb < Mn < Cr. This means that the leaching of these metals becomes more difficult in this order, because Cd and Zn are mainly retained in the exchangeable fraction of sandy clay loam and loam whereas other metals are mainly bound to oxides and the organic fraction of soils. If the fraction $\alpha_0 \geq 0.5$, the main part of the soil metal react slowly and the quasi-equilibrium concentration is leached slowly.

The fit of the data points by the two-reaction model (Figure 6.1a, b, c, d) showed that a reasonable rate description is possible by the two process model using different rate constants. The model describes the metal concentration curves over a time period of 36 h with a standard error of less than 2.1%. The curves and the model parameters show that the individual metals behave differently in a single treatment. The value of the quasi-equilibrium constant m_1 is much smaller in the case of Cd, Cu, Pb, Zn and Hg (< 0.6 l/kg), whereas Mn and Cr showed higher values of 1.1 to 4.5 for citrate and EDTA, but lower values for tartarate because the extraction was carried out at low pH. This indicates that Cr and Mn are difficult to remove from the clay loam because they are tightly bound as it was observed (Wasay et al. 1998a). A higher m_1 value of Cr and Mn indicate that the removal rate was low in a single extraction with citrate, tartarate and EDTA even after 36 h. The values of m_1 for all soils were very small indicating a high removal efficiency for the metals. The fast initial leaching rates are comparable to the

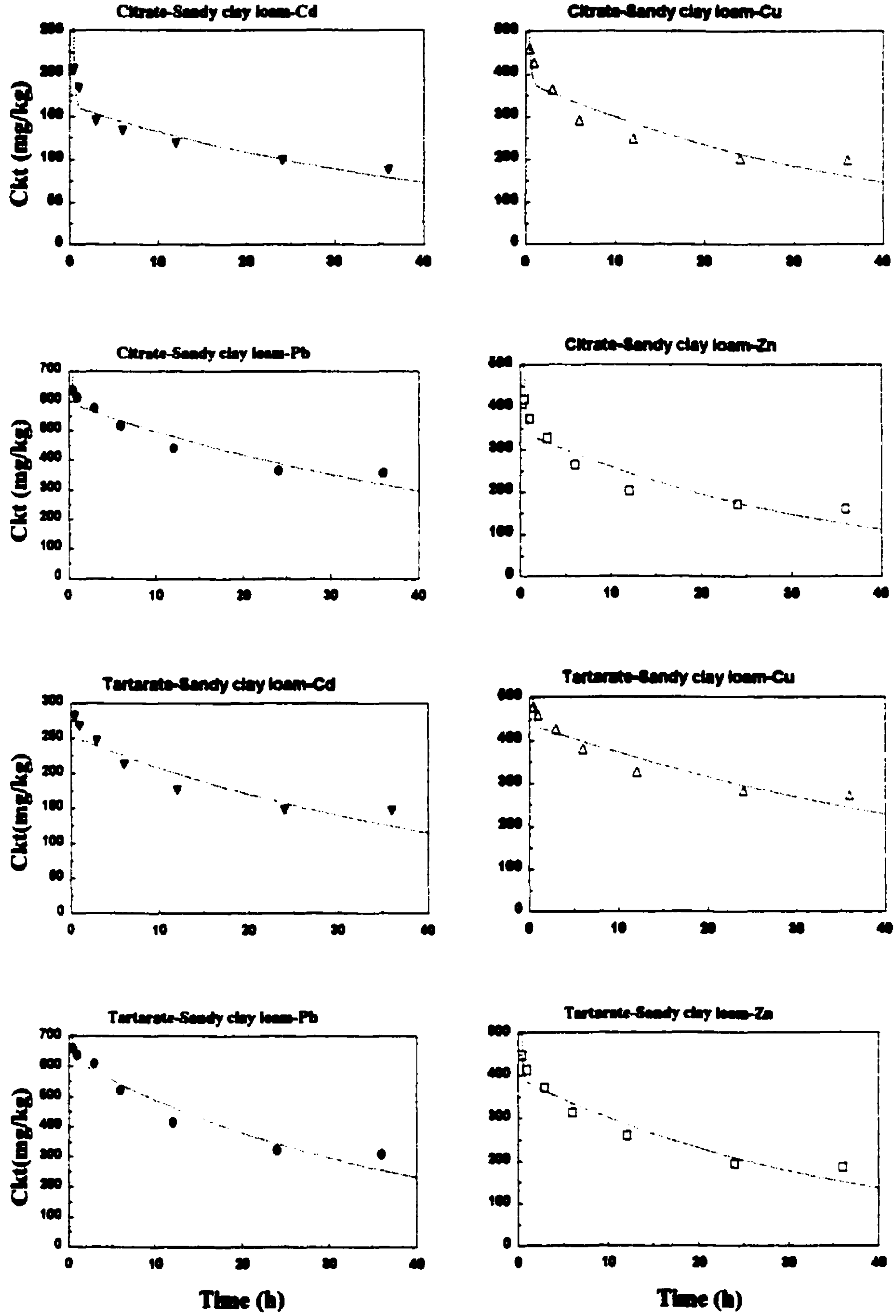

Figure 6.1a Heavy metal $[C_k(t)]$ in the soil as a function of time. Dotted line shows curve fit to experimental data.

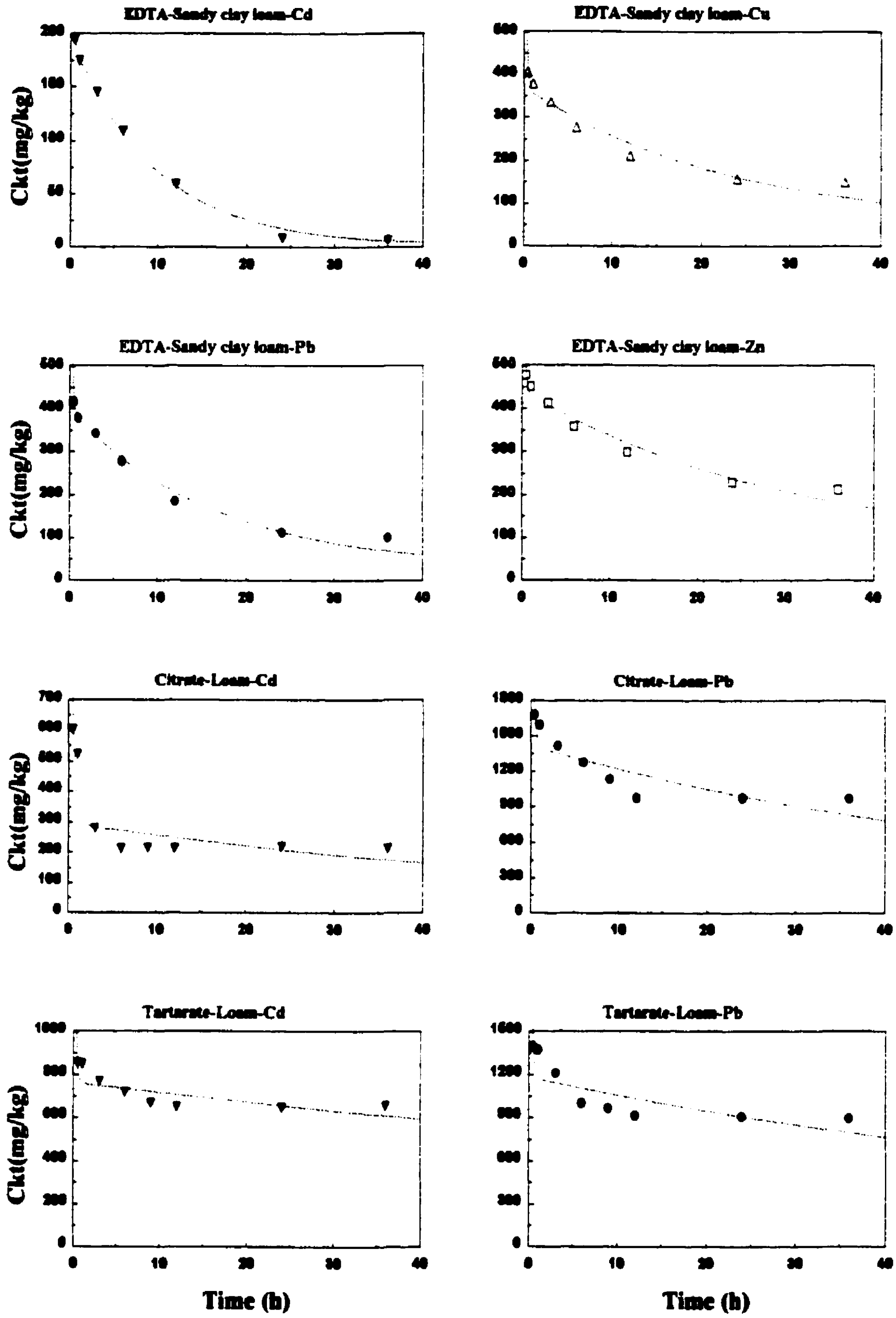

Figure 6.1b Heavy metal $[C_k(t)]$ in the soil as a function of time. Dotted line shows curve fit to experimental data.

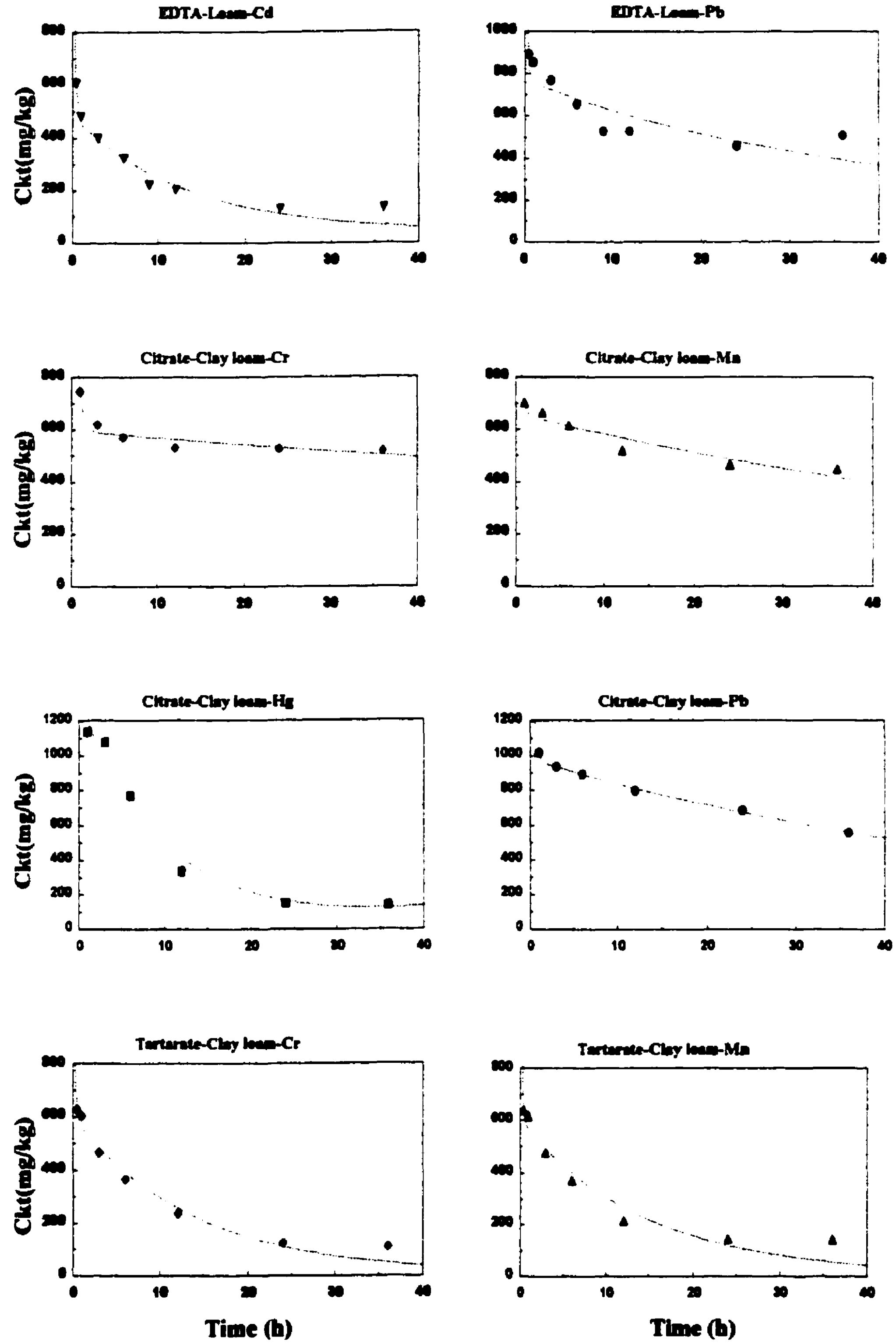

Figure 6.1c Heavy metal $[C_k(t)]$ in the soil as a function of time. Dotted line shows curve fit to experimental data.

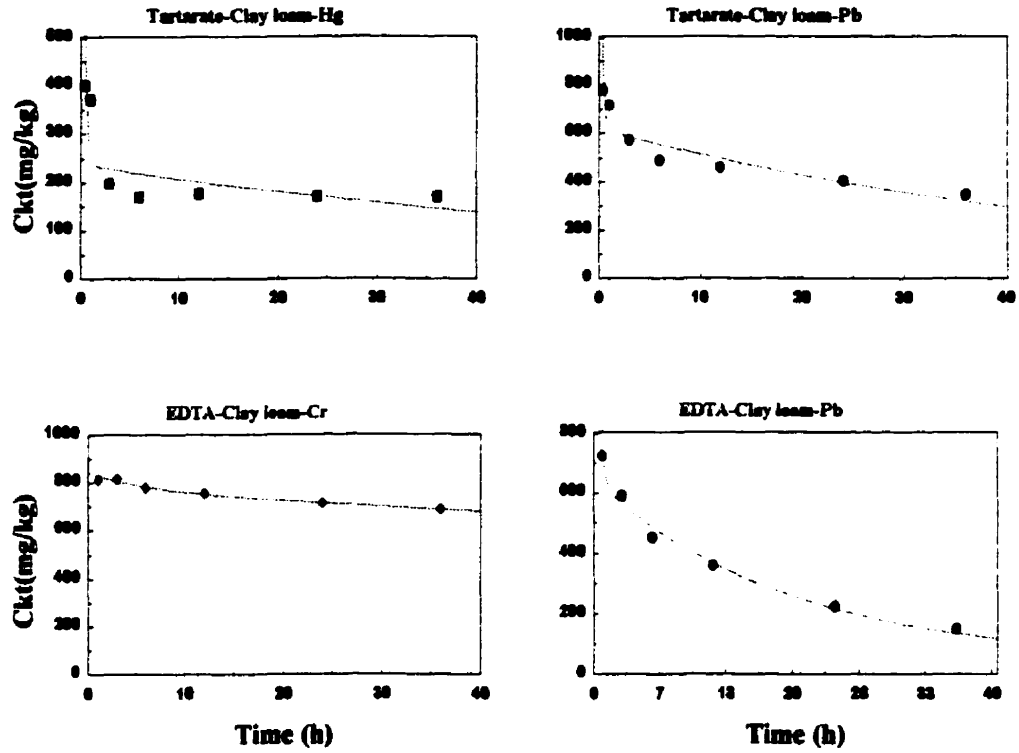

Figure 6.1d Heavy metal [$C_k(t)$] in the soil as a function of time. Dotted line shows curve fit to experimental data.

Table 6.1 Kinetic model parameters for two-leaching reaction rate of metals from soils

Soil	Extractant	Metal	m_1 (l/kg)	k_1 (h^{-1})	k_2 (h^{-1})	α_0 (-)	StdErr (%)	Cor.Coef. (r)
Sandy clay loam	Citrate	Cd	0.141	5.935	0.020	0.201	< 0.555	0.964
		Cu	0.273	3.229	0.023	0.398	< 0.826	0.955
		Pb	0.529	3.989	0.018	0.568	< 1.411	0.969
		Zn	0.183	4.024	0.028	0.328	< 0.912	0.963
	Tartarate	Cd	0.228	5.585	0.020	0.319	< 1.467	0.994
		Cu	0.414	4.462	0.017	0.469	< 1.263	0.964
		Pb	0.424	4.291	0.025	0.605	< 2.135	0.975
		Zn	0.219	4.473	0.027	0.379	< 0.995	0.958
	EDTA	Cd	0.010	8.649	0.104	0.247	< 1.685	0.992
		Cu	0.194	4.788	0.040	0.395	< 1.322	0.979
		Pb	0.107	6.070	0.059	0.393	< 1.666	0.979
		Zn	0.259	5.039	0.030	0.428	< 1.195	0.985
Loam	Citrate	Cd	0.133	3.518	0.015	0.163	< 0.446	0.881
		Pb	0.529	2.570	0.014	0.489	< 0.747	0.918
	Tartarate	Cd	0.564	4.396	0.006	0.416	< 1.138	0.840
		Pb	0.472	3.251	0.010	0.411	< 0.885	0.870
	EDTA	Cd	0.080	4.360	0.071	0.261	< 0.683	0.983
		Pb	0.222	5.232	0.025	0.273	< 1.518	0.885
Clay loam	Citrate	Cr	1.586	0.405	0.001	0.643	< 0.031	0.998
		Mn	1.067	1.154	0.013	0.749	< 0.745	0.982
		Hg	0.126	0.506	0.106	1.137	< 2.059	0.988
		Pb	0.498	2.589	0.016	0.588	< 0.265	0.999
	Tartarate	Cr	0.153	1.671	0.061	0.646	< 0.636	0.992
		Mn	0.200	1.392	0.054	0.599	< 0.655	0.979
		Hg	0.153	3.136	0.012	0.176	< 0.655	0.821
		Pb	0.267	3.177	0.017	0.360	< 0.603	0.960
	EDTA	Cr	4.497	0.151	0.004	0.918	< 0.142	0.988
		Pb	0.098	2.226	0.051	0.388	< 0.342	0.993

magnitude of the exchangeable fraction of metals in soils. The values of the model parameter k_1 obtained in the present study for all metals in polluted soils are in the range of 0.15-6.1 h^{-1} as measured for Cu extraction using EDTA by Jopony and Young (1987). The very slow rate of the 2nd reaction and the fairly large fractions α_0 that reacts according to the slow process indicated that prolonged extraction time will hardly improve the metal removal efficiency from the soils by citrate, tartarate and EDTA solutions. The goodness of the curve fit can be seen with correlation of coefficient, r as shown in Table 6.1.

6.6 Conclusions

The following conclusions can be reached from the present study:

1. The rate of leaching of heavy metals from soils were analyzed using a two-reaction model based on extraction data obtained with citrate, tartarate and EDTA, at a constant pH, concentration of extractants, temperature and shaking speed. The two-reaction model encompasses two processes, the fast irreversible first-order and the slow reversible first-order leaching of metals.
2. Cadmium leached out faster (higher k_1) with all three extractants (citrate, tartarate and EDTA) followed by Zn, Cu, Pb, Hg and Cr at 20 °C. The same order was obtained with the slower leaching rate (k_2). This resulted from the release of the exchangeable fraction of metals especially Cd and Zn and then the release of the more tightly bound metals such as Cu, Pb, Hg and Cr bound to oxide and the organic fraction of the soil.
3. The values of m_1 were very small indicating high removal efficiency of metals from all soils.
4. If the fraction of slowly extractable metals (α_0) is greater than 0.5, most of the metals in soils leach slowly and the quasi-equilibrium concentration is reached slowly. But for $\alpha_0 < 0.5$, the leaching of metal is fast.

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CONNECTING STATEMENT

The last three chapters (4, 5 and 6) dealt with setting up the remediation technique and measuring the kinetic parameters for the clay loam, the loam and the sandy clay loam in batch experiments. The next chapter (chapter 7) deals with the remediation of soils in a column experiment using citrate, tartarate, citrate+oxalate, EDTA and DTPA as flushing solutions and in a large scale level in a tub using citrate as a flushing solution. The optimum conditions established in the previous chapters was used for the maximum leaching of heavy metals from the experimental soils. Each pore volume from the column and tub at different time intervals was collected and analyzed for heavy metals.

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CHAPTER 7

ORGANIC ACIDS FOR THE IN-SITU REMEDIATION OF SOILS POLLUTED BY HEAVY METALS: SOIL FLUSHING IN COLUMNS

7.1 Abstract

Three soils of different texture and polluted by heavy metals were washed in a series of columns at optimum pH with a salt of weak organic acids, namely citrate, tartarate or oxalate+citrate or a chelating agent such as EDTA and DTPA. For the clay loam, Cr, Mn, Hg and Pb were removed by citrate and tartarate at levels of 43 to 45, 37 to 41, 91 to 92 and 75%, respectively. EDTA and DTPA effectively leached only Pb after 20 pore volumes. For the loam, citrate leached out 98 and 89% of Cd and Pb after 20 pore volumes, respectively, while tartarate leached out 91 and 87% of Cd and Pb. EDTA and DTPA removed these metals between 93 to 97% after 20 pore volumes. For the sandy clay loam, Cd, Cu, Pb and Zn were removed between 84 to 91, 73 to 84, 56 to 70 and 72 to 81%, respectively using citrate and tartarate. EDTA and DTPA removed these metals between 93 to 97% after 20 pore volumes. An in-situ soil remediation simulation was also tested using the sandy clay loam in a tub. Cadmium, Cu, Pb and Zn were removed by citrate washing solution at levels of 81, 82, 73 and 90%, respectively after six pore volumes, each retained for 12 h. EDTA and DTPA were effective in removing heavy metals except for Hg because of their strong chelating power. But these strong chelating agents extract important quantity of nutrients from the soil and pollute the soil by being adsorbed on the soil particles. The salts of citrate and tartarate were found to be quite effective in removing heavy metals from the three polluted soils while leaching little macronutrients and improving the structure of soils. Each type of soil reached to C and B level of soil clean up criteria after 10 to 20 pore volumes within 10 to 15 h of flushing time.

7.2 Introduction

Two in-situ remediation techniques are used to remediate soils polluted by heavy metals: i) in-situ immobilization or fixation of heavy metals in the soil where different immobilization materials (such as chelates, ion exchange resins, portland cement/silicate, natural materials-clays, -molecular sieves and -greensand and other additives like hydrated lime, silylated silica gel, insoluble starch xanthate, ferrous sulfate) can be added to the soil to reduce the mobility of heavy metals (Liu and Dezfulian 1995, Czupyrua et al. 1989). This process may lead to future leaching problem of heavy metals into surface and ground waters due to biological activity or acidification of the soil ii) in-situ soil flushing to leach out heavy metals from polluted soil. The soil flushing solution may be an acid or a chelating agent. This process is the most appealing because it generally creates less surface damage, requires a minimal amount of facilities, reduces the potential for human exposure to contaminants, is less expensive and, when effective, reduces or removes the contaminants. However, this remediation process is most suited for permeable soils.

Soil flushing in columns of sandy loam artificially polluted by Pb was investigated using HCl, EDTA and $CaCl_2$ as flushing solutions (Reed et al. 1996). Lead removal efficiency was 85%, 100% and 78%, with HCl, EDTA and $CaCl_2$, respectively. Several flushing solutions such as acid, EDTA, DTPA and chlorine were applied at various concentrations to an artificially Zn contaminated soil contained in a column to determine Zn remediation efficiency (Davis and Singh 1995). The effects of ionic strength, flow rate, retention forms and levels of Zn contamination were also studied on remediation. Complete Zn removal was obtained with EDTA and DTPA at the lowest and highest flow rates. Removal efficiency was strongly dependent on the retention form of Zn in the soil. The chemicals used for washing either destroyed the soil properties or were ineffective in removing heavy metals.

Various nonhumified (weak organic acids) and humified (humic and fulvic acids) organic acids have been identified in soils. Nonhumified acids such as citric, tartaric, oxalic, formic and fumaric acids are present in the soil due to the decay of plants, animals and microbial tissues (Tan 1986). These natural weak organic acids have chelating characteristics which can mobilize heavy metals. These natural chelators are biodegradable and form humified substances which can solubilize heavy metals. These natural organic acids were found to be abundant in tree roots exudates to enhance the availability of metals to plants. Some organic acids were used for leaching out Zn from mine tailings in a column study (Bruckhard et al. 1995). The highest Zn concentration was occurred in the leachate with the highest citric acid concentration (10 mM). The amount of Zn leached out from the column was similar to that leached out by formic and succinic acid solutions. Oxalic acid leached slightly more Zn than formic and succinic acids.

The objective of the present study was to test salts of weak organic acids to leach heavy metals from soils held in laboratory columns. The leaching efficiency of these salts was compared to that of chelating agents (EDTA and DTPA) and the leaching effect of agents on the soil properties was evaluated.

7.3 Materials and Methods

7.3.1 Experimental soil sample

Each soil sample was obtained from a site polluted for 3 years by heavy metals (Table 7.1). The soil samples were air dried and sieved to remove the particles greater than 2 mm because about 90% of the contaminants are present on particles smaller than 2 mm. The soil samples were homogenized and stored in plastic bags for further study. Soils were characterized physically and chemically and their forms of heavy metal retention was determined using sequential extraction (Wasay et al. 1998a).

7.3.2 Materials

Soil flushing solutions were prepared and their pH was corrected for maximum leaching according to an earlier study (Wasay et al. 1998b, 1998c). A flushing solution was prepared using ammonium citrate (0.2 M) and its pH was corrected to 5.0 using NaOH (0.06 M) in a ratio of 4:1 (ammonium citrate:NaOH). Another flushing solution was prepared by mixing tartarate (2:1 ratio of tartaric acid and ammonium tartarate, 0.3 M each) and water in a ratio of 3 : 2 to obtain a desired pH of 3.6. An admixture of

Table 7.1 Physical and chemical characterization of contaminated soils.

Soil characteristic	Clay loam	Loam	Sandy clay loam
Particle size			
% sand	29.0±0.6	50.0±0.5	53.5±0.9
%Silt	36.0±0.4	30.0±1.2	24.0±1.6
% clay	35.0±1.0	21.0±0.7	23.0±0.7
Bulk density (g/cm^3)	0.8±0.0	0.9±0.0	0.9±0.0
Particle density (g/cm^3)	2.0±0.0	2.0±0.0	2.1±0.1
Total porosity (%)	45.4±0.1	44.1±0.2	48.0±0.4
k_s(cm/s)	1.1±0.1x10^{-4}	1.3±0.1x10^{-4}	4.0±0.2x10^{-4}
EC (mS/cm)-water	9.2±0.2	8.0±0.2	2.3±0.2
pH-water	4.2±0.0	5.1±0.1	6.3±0.1
-$CaCl_2$	4.3±0.1	5.2±0.1	6.0±0.0
CEC (cmol(+)/kg)	2.0±0.2	4.7±0.0	12.2±0.2
Organic matter (%)	9.9±0.1	5.4±0.1	7.1±0.1
Heavy metals (mg/kg)			
Cr (Total)	840	-	-
Mn	868	-	-
Hg	1280	-	-
Pb	1664	2797	1036
Cd	-	1821	794
Cu	-	-	926
Zn	-	-	1029

ammonium citrate (0.2 M) and ammonium oxalate (0.2 M) in a 1:1 ratio was prepared and corrected to a pH of 5.8. Soil flushing solutions of ethylenediaminetetraacetic acid (EDTA) and diethylenetriaminepentaacetic acid (DTPA) (0.1 M) were dissolved in

NaOH solution (0.1 M) in a ratio of 4:1 to obtain desired pH of at least 5.5.

7.3.3 Pore volume

The total pore volume of the soil columns was determined using an air dried soil sample of 40 g. This sample was weighed in a moisture can and a known amount of water was added until saturation was obtained. This point is reached when the soil paste glistens as it reflects light, flows slightly when the container is tapped and the paste slides freely and cleanly off the soil spatula. Free water should not collect on the soil surface nor should the paste stiffen markedly or lose its glistening appearance on standing. If the paste does stiffen or loses it glisten on standing for an hour or more, it is remixed with additional quantity of water. The total porosity (f) was determined by the following formula:

$$f = \frac{V}{V + V_s} \tag{7.1}$$

$$V_s = \frac{M}{\rho_s} \tag{7.2}$$

where V (ml) is the volume of water added, Vs (ml) is the volume of the soil particles, M (g) is the mass of the dry soil and ρ_s (g/ml) is the density of soil particles. The pore volume of the soil was measured by the following formula:

$$V_p = \frac{f}{100} * \pi . r^2 h \tag{7.3}$$

where V_p (ml) is the pore volume of the soil column, r (cm) is the radius of the column, h (cm) is the height of the soil column and f is the total porosity of the soil.

7.3.4 Method

Three of soil columns, 2 cm in internal diameter and 4 cm in height were packed with 5.0 to 8.5 g of one of the 3 contaminated soils (Figure 7.1). Three columns each holding one of the 3 different soils, were connected to a 1.0 L reservoir holding a solution of either one of the salts of organic acids (NH_4 and Na), one of the two chelating agents (EDTA and DTPA) at an optimum pH or a control solution of deionized water. Each flushing solution was allowed to pass through the soil column and each volume equivalent to that of the soil pore was collected with time. The flow rate of the flushing solution through the soil column was maintained between 4 to 10 ml/h. The pore volume was collected periodically for heavy metal analysis. All experiments were carried out in triplicate.

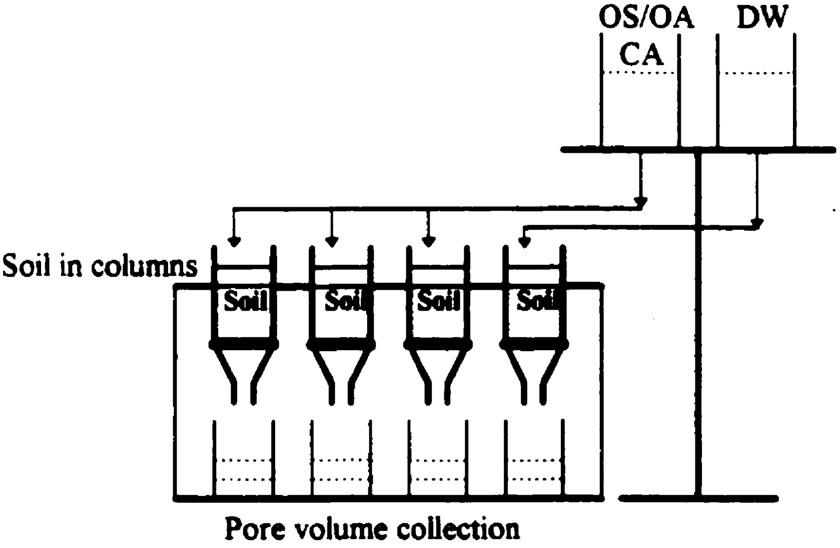

Figure 7.1 Schematic diagram for the soil flushing in a column experiment. Soil flushing liquid: organic salt (OS), organic acid (OA), chelating agent (CA). Control: deionized water (DW).

7.3.5 Large scale experiment

A large scale experiment was conducted with the sandy clay loam in a tub to simulate in-situ conditions. The tub contained 7.7 kg of the soil contaminated with Cd, Cu, Pb and Zn. The surface of the soil was irrigated with a 0.16 M ammonium citrate solution (washing solution) at a pH of 5.0. The washing solution was applied to the soil surface and allowed to soak in the soil for 12 h before being drained. This procedure was repeated 6 times. Each pore volume was collected by gravity through 4 perforated wells after a 12 h retention time (Figure 7.2). The volume of leachate collected was measured in terms of the soil's pore volume and sampled to determine its heavy metal content After flushing the soil in the tub, residual metals were determined by acid digation.

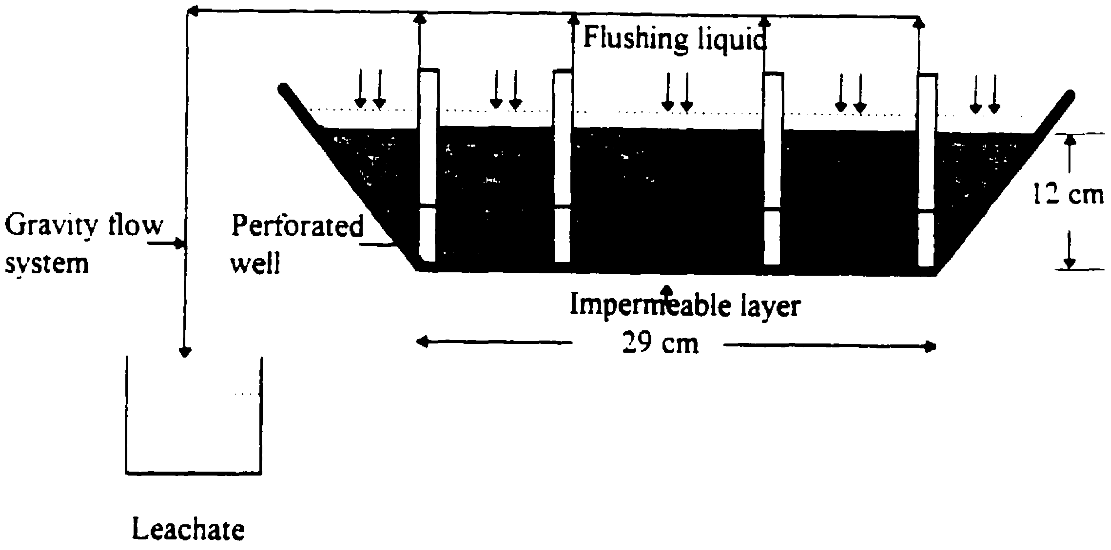

Figure 7.2 Schematic diagram for soil flushing in a tub experiment.

7.3.6 Statistical analysis

The SAS repeated measure analysis of variance procedure was used for the leaching rate of heavy metals from the column (SAS 1990).

7.4 Results and Discussions

7.4.1 Soil flushing experiment

The amount of heavy metals removed with time for each soil pore volume for the clay loam, is illustrated in Figure 7.3a. Each pore volume (leachate) for clay loam was equivalent to 3.8 ml per 5.0 g of soil and it took 0.5 to 0.7 h to leach one pore volume. For the clay loam, Cr, Mn, Hg and Pb were removed at levels of 11, 31, 20 and 95%, respectively, using EDTA and DTPA after 20 pore volumes. With ammonium citrate(0.16 M), the leaching of Cr, Mn, Hg and Pb reached levels of 43, 37, 92 and 75%, respectively, also after 20 pore volumes. The percentage removal of Cr, Mn, Hg

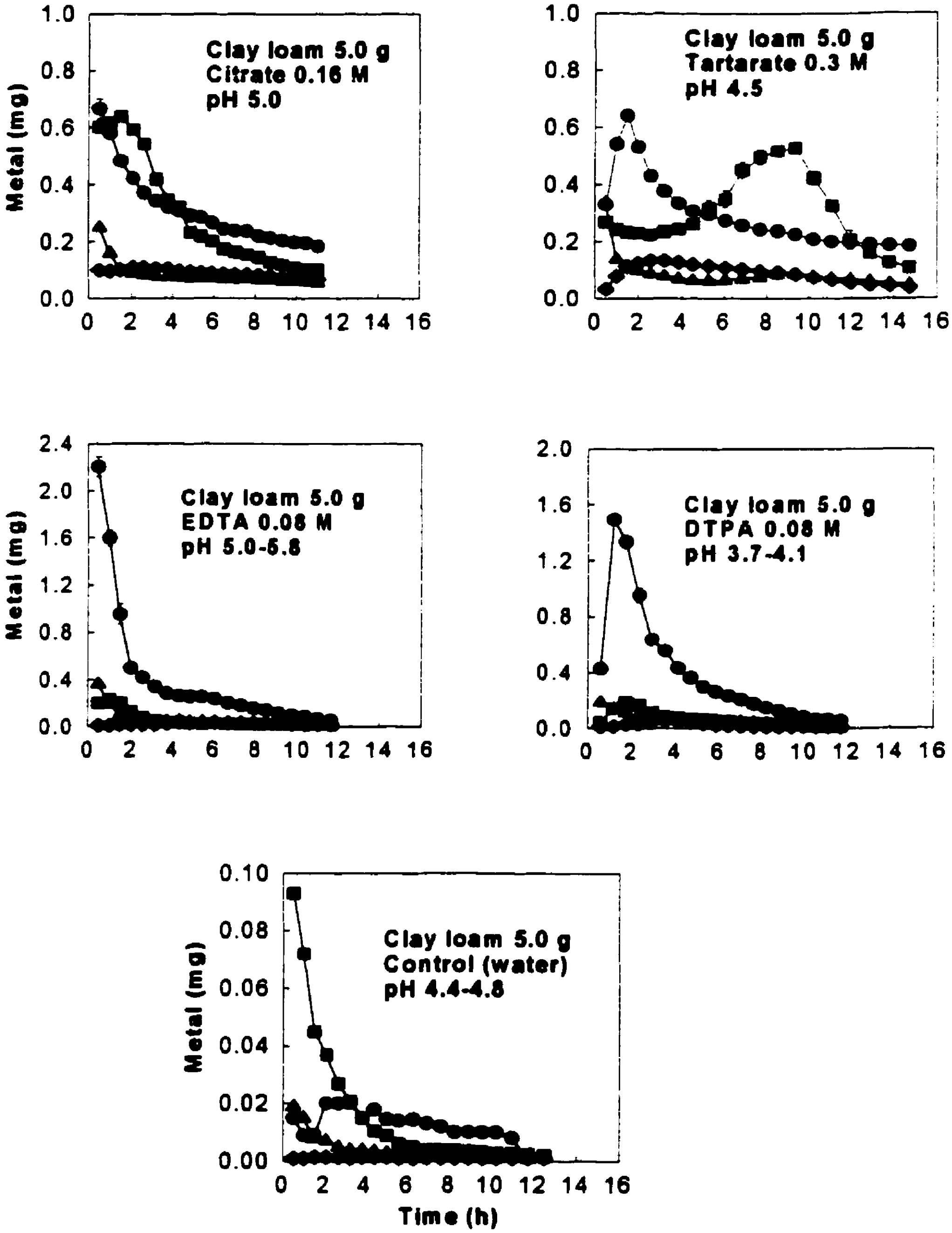

Figure 7.3a Soil flushing in a column for an in-situ remediation. ◆ Cd, ■ Hg, ▲ Mn and ● Pb.

and Pb was found to be 45, 41, 91 and 75% using ammonium tartarate (0.3 M), respectively. The heavy metals leached out by the water in the control treatment was found to be 0.54, 5.78, 1.97 and 2.83% for Cr, Hg, Mn and Pb, respectively, after 20 pore volumes. The optimum conditions such as pH, concentration of salts of organic acids and chelating agents and amount of clay loam washed are shown in Figure 7.3a. Because the heavy metals were tightly bound on the clay loam, it was difficult to remove particularly Cr(III) and Mn by both the weak organic salts, such as citrate and tartarate, and the strong chelating agents, such as EDTA and DTPA.

For the loam, levels of heavy metals removed with time for each soil pore volume are illustrated in Figure 7.3b. Each pore volume was equivalent to 3.6 ml per 5 g of soil and it took 0.5 to 0.63 h to leach one pore volume. Ammonium citrate at optimum pH of 5.0, removed 98 and 89% of Cd and Pb, respectively after 20 pore volumes which required a total time of 9.6 h. At 7 pore volumes and 3.5 h, removal efficiency was found to be 83 and 62% for Cd and Pb, respectively. This fraction of heavy metals was mainly released from the easily exchangeable form of metals. After 7 pore volumes, heavy metals were released more slowly because they were moderately to tightly bound to oxides and organic matter. For this soil, Pb was mainly bound to oxides and the organic fraction of the soil, whereas Cd was mainly present in exchangeable form (Wasay et al. 1998a). Therefore, citrate easily leached Cd as compared to Pb. With tartarate, Cd and Pb were leached at levels of 91 and 87%, respectively in 20 pore volumes and after 12 h, at an optimum pH of 3.6 to 4.0. At 10 pore volumes, the removal of Cd and Pb was found to be 82 and 60%, respectively and was also much slower thereafter. EDTA and DTPA leached out 93 to 97% of Cd and Pb after 20 pore volumes, at optimum pH of 5.5 to 7.5. At 10 pore volumes and 5 to 4.3 h, the leaching of Cd and Pb was 86 and 81% for EDTA and DTPA, respectively and was much slower thereafter. For the control treatment (with water), the leaching of Cd and Pb reached levels of 13.2 and 0.95%, respectively after 20 pore volumes.

For the sandy clay loam, levels of heavy metal removed with time with each soil pore volume are shown in Figure 7.3c. Each soil pore volume was equivalent to 5.2 ml per 8.5 g of soil and it took 0.72 to 1.0 h to leach out one pore volume. Leaching of Cd,

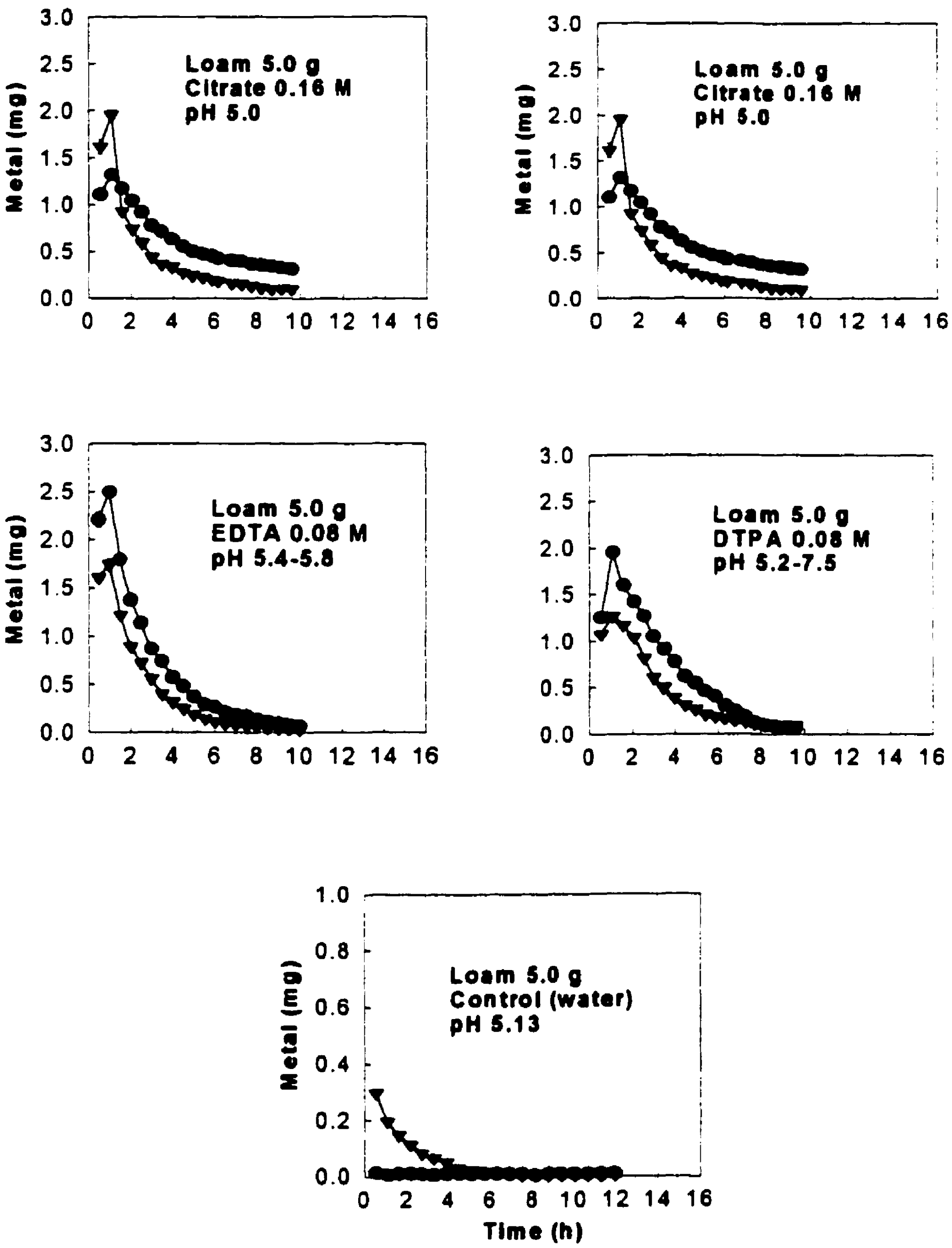

Figure 7.3b Soil flushing in a column for an in-situ remediation. ▼ Cd and ● Pb.

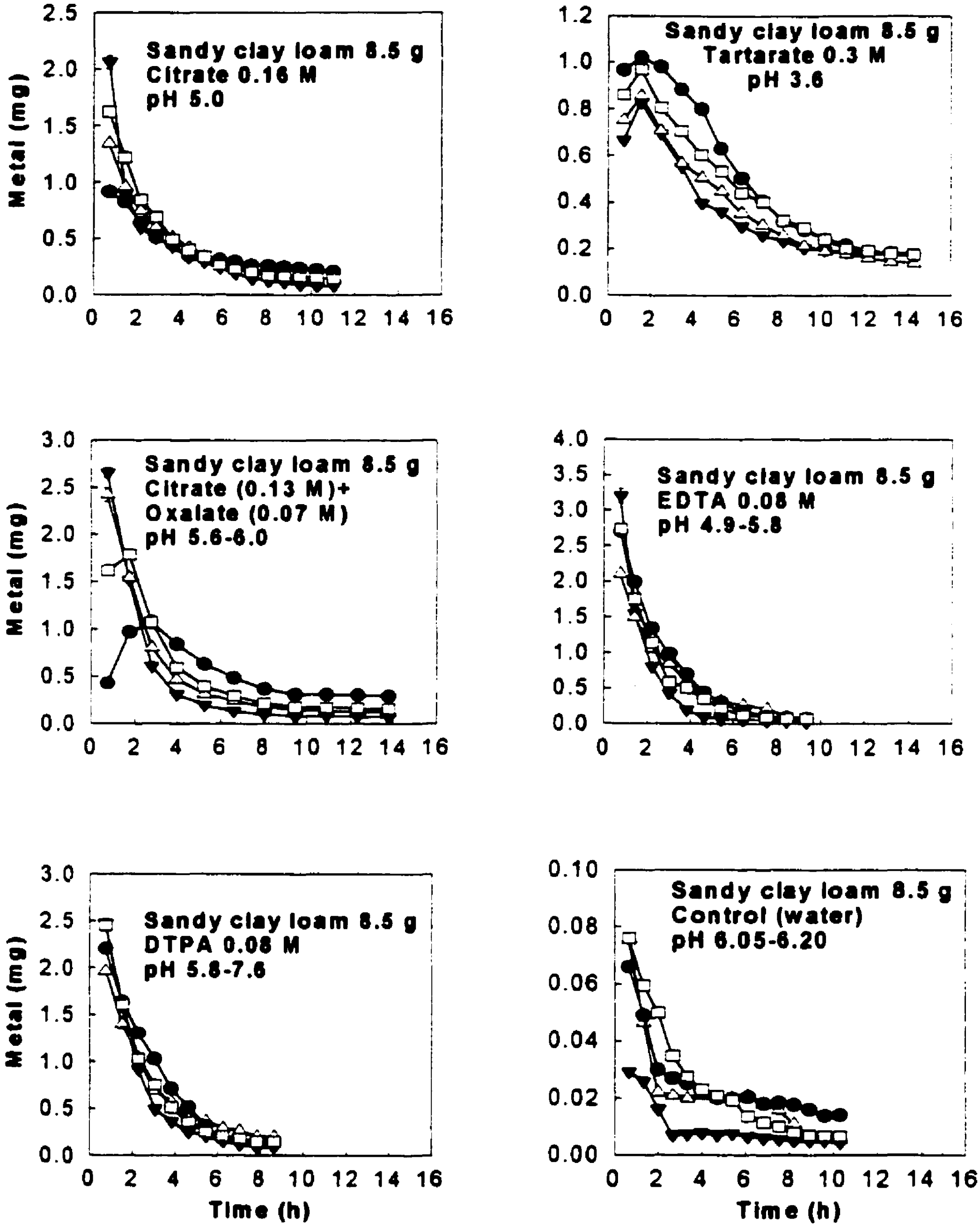

Figure 7.3c Soil flushing in a column for an in-situ remediation. ▼ Cd, ● Pb, Δ Cu and □ Zn.

Cu, Pb and Zn by ammonium citrate was found to reach levels of 91, 84, 70 and 81%, respectively after 15 pore volumes and 11 h. At 10 pore volumes and 7.3 h, the release of Cd, Cu, Pb and Zn reached levels of 84, 73, 56 and 72%, respectively. Tartarate released Cd, Cu, Pb and Zn at levels of 83, 73, 89 and 79%, respectively, after 15 pore volumes and 14.7 h. At 10 pore volumes and 9.6 h, the leaching of Cd, Cu, Pb and Zn was found to be 69, 63, 77 and 67%, respectively. For EDTA and DTPA, the leaching of Cd, Cu, Pb and Zn ranged between 96 to 98, 90 to 94, 92 to 99.6 and 85 to 87%, respectively after 11 pore volumes and a leaching time of 8.6 to 9.7 h.

Both EDTA and DTPA were found to be significantly effective in removing these metals because of their strong chelating properties as compared to natural chelators such as citrate and tartarate. Therefore, EDTA and DTPA also extracted all other nutrients from soils, thereby decreasing the chemical value of the soil. Furthermore, the chelating agent can be adsorbed on the soil surface (Bourg 1988). Because EDTA resists biodegradation and is adsorbed on the soil particles, soils remediated by such chelating agents are unfit for agricultural use. The salts of organic acids, such as ammonium citrate, effectively removed heavy metals from the soil while being adsorbed on the surface of the soil particles. But these natural chelators are biodegradable and produce humified substances such as humic and fulvic acids, which can improve the structure of the soil by cementing the soil particles into water stable aggregates.

Three soils reached category C and B levels of soil clean up criteria after 10 to 20 pore volumes within 10 to 15 h of flushing time using citrate, tartarate, EDTA and DTPA. Soils require more flushing to reach A level of soil clean up criteria before disposal on agricultural land. For all 3 contaminated soils, the loss of macronutrients, such as Ca, Mg, Fe, by EDTA and DTPA was found to be 4 to 5 times higher than that of the citrate. EDTA and DTPA form strong chelates with soil macronutrients whose stability constant is very high as compared to citrate or tartarate (Bell 1977). Therefore, EDTA and DTPA have a negative impact on the soil pollution and soil nutrient content.

7.4.2 Large scale experiment

The release of heavy metals from the sandy clay loam using ammonium citrate (0.16 M) was plotted as a function of retention time (Figure 7.4). The leaching of Cd, Cu, Pb and Zn was found to be 81, 82, 73 and 90 % of the total 6.114, 7.130, 7.977 and 7.923 g of contaminants in the soil, respectively after six pore volumes, each retained for 12 h. The soil still retained 0.306, 1.070, 1.835, and 0.713 g of Cd, Cu, Pb and Zn, respectively after 6 pore volumes. By extrapolating to 10 pore volumes, Cd and Zn are expected to meet the clean up criteria (level A) whereas Cu and Pb could have reached the clean up criteria for agricultural soils (level B) which requires more flushing (MEFQ 1994). The release of Pb was the slowest among all other heavy metals, whereas Zn and Cd were the fastest.

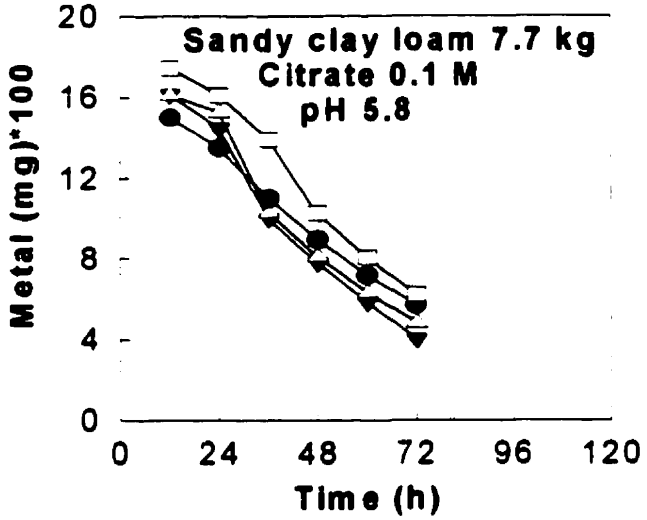

Figure 7.4 Soil flushing in a tub for an in-situ remediation. ▼ Cd, ● Pb, Δ Cu and □ Zn.

7.4.3 Statistical analysis

The flushing solutions had a highly significant effect on the rate of leaching of the heavy metals ($P < 0.0001$). For the clay loam, EDTA and DTPA leached out Pb faster and decreased Pb to a minimum amount in 8 pore volumes followed by citrate and tartarate.

Manganese was removed faster by EDTA followed by tartarate, citrate and DTPA. Chromium was leached out faster by citrate and tartarate as opposed to EDTA and DTPA. Mercury was flushed out faster from the clay loam by citrate followed by tartarate, EDTA and DTPA. For the control using water, the leaching of all metals was found to be minimum as compared to all other treatments. The coefficient of variance (CV) varied between 1.8 to 7.0% for all metals, at different time intervals.

For the loam and for the first 2 pore volumes, Cd leaching was faster by citrate followed by EDTA and tartarate or DTPA and thereafter tartarate removed Cd faster as compared to other chemicals. Leaching by water (control) was found to be the least effective. For Pb leaching, EDTA and DTPA were the most effective, followed by citrate and tartarate. The coefficient of variance (CV) was found to range between 1.26 and 4.53% for both Cd and Pb.

For the sandy clay loam, Cd leaching was faster for the first pore volume, with EDTA followed by citrate+oxalate, DTPA, citrate and tartarate. After four pore volumes, the leaching of Cd was better with tartarate followed by citrate, DTPA, EDTA and citrate+oxalate. Deionized water was the least effective in leaching Cd. Citrate+oxalate leached out Cu faster than EDTA followed by DTPA, citrate and tartarate for the first 2 pore volumes. EDTA and DTPA had a better efficiency for Cu until 5 pore volumes, and thereafter citrate and tartarate achieved the best efficiencies. For Pb and Zn, EDTA and DTPA demonstrated the highest rate whereas tartarate, citrate and citrate+oxalate demonstrated a slow rate for both metals. The coefficient of variance (CV) ranged between 1.28 to 5.77% for all metals at each pore volume.

7.5 Conclusions

The following conclusions are:

1. For the clay loam, the removal of Cr, Mn, Hg and Pb by citrate and tartarate ranged between 43 to 45, 37 to 41, 91 to 92 and 75%, respectively. EDTA and DTPA could only remove Pb effectively whereas for Cr, Mn and Hg, they were found to be less effective after 20 pore volumes.

2. For the loam, ammonium citrate at optimum pH leached out 98 and 89% of Cd and Pb, respectively after 20 pore volumes. Ammonium tartarate leached out 91 and 87% of Cd and Pb, respectively after 20 pore volumes. EDTA and DTPA also removed Cd and Pb between 93 and 97%, at 20 pore volumes.
3. For the sandy clay loam, the leaching of Cd, Cu, Pb and Zn ranged between 83 to 91, 73 to 84, 56 to 70 and 72 to 81% with citrate and tartarate, respectively. The removal of these metals by EDTA and DTPA ranged between 85 to 98% after 15 pore volumes.
4. The removal of Cd, Cu, Pb and Zn in the large scale experiment (tub) reached 81, 82, 73 and 90%, respectively after six pore volumes, each retained for 12 h.
5. Three soils were reached to C and B level of soil after 10 to 20 pore volumes within 10 to 15 h of soil flushing using citrate, tartarate, EDTA and DTPA. Soils require more flushing to reach A level of soil clean up criteria before disposal on agricultural land.
6. EDTA and DTPA were effective in removing all heavy metals except for Hg because of their strong chelating properties. But they also extract all other important nutrients from the soil and they pollute the soil by being adsorbed and non biodegradable on the surface of the soil particles.
7. Organic salts such as ammonium citrate and tartarate, are also quite effective in removing heavy metals from polluted soils. Even if the salts of these organic acids are also adsorbed on the surface of the soil particles, they improve the soil structure and help in forming water stable aggregates.

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CONNECTING STATEMENT

The next chapter deals with the bioremediation of soils polluted by heavy metals using the fungus, *Aspergillus niger*. After successfully studying the effectiveness of weak organic acids such as citrate to remediate soils in both batch and column experiments (chapter 4, 5, 6), a bioremediation process was developed using *Aspergillus niger* to generate citrate in-situ to further minimize the cost of remediation. To generate citrate, *Aspergillus niger* was grown on the surface of the contaminated soil fed sucrose as a carbon source and rest at a constant temperature under aerobic condition. The leachate in the form of pore volume was collected every day from soil column while *Aspergillus niger* was continuously supplied citrate.

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CHAPTER 8

BIOREMEDIATION OF SOILS POLLUTED BY HEAVY METALS USING *ASPERGILLUS NIGER*

8.1 Abstract

A bioremediation process was developed using the fungus *Aspergillus niger* to produce weak organic acids for the leaching of heavy metals from contaminated soils. The fungus was cultivated on the surface of three contaminated soils (a clay loam, a loam and a sandy clay loam) for 15 days at 30 °C and at a pH < 4 to favor the production of citric acid rather than oxalic acid which hinders Pb leaching. For the clay loam, Cr, Mn, Pb and Hg were leached to levels of 37, 41, 85 and 91%, respectively. For the loam, the leaching of Cd and Pb was found to reach levels of 99.7 and 83%, respectively. For the sandy clay loam, Cd, Cu, Pb and Zn were leached to levels of 99, 94, 58, 99%, respectively. The three contaminated soils could have been remediated to an A category after 20 to 25 days of leaching using this technique.

Key words: Soil pollution, heavy metals, bioremediation, leaching, *Aspergillus niger*, column experiment.

8.2 Introduction

Soil pollution by heavy metals is a growing concern in Canada, USA, Japan, Europe and various developing countries. The metal mining and smelting industries are the main sources of heavy metal pollution (Asami 1988). Soil cleaning is expensive because heavy metals are very tightly bound to soil particles. The estimated clean up cost of contaminated soils ranges between US $ 8 to 12 per head in industrialized countries (Ellis and Mellor 1995). It has been estimated that the clean up cost by current technology (ex-situ or excavation and reburial) for one hectare of soil over a depth of one meter can range between US $ 0.6 to 3 million depending on the pollution load (Moffat 1995).

Earlier studies showed that strong acids (HC1, HNO_3 and H_2SO_4), strong synthetic chelating agents (EDTA and DTPA) and microorganisms are effective in leaching heavy metals from polluted soils and sewage sludges facing major disadvantages (Tuin and Tels 1990, Benschoten et al. 1994, Cline and Reed 1995, Davis and Singh 1995, Strasser et al. 1994 and Wong and Henry 1985). Both strong acids and chelating agents can disturb the properties of the soil by extracting important macronutrients. From 8 to 11% by weight of the total soil material was dissolved in 0.1 M HCl after 30 min and about 13-14% by weight after 24 h of extraction (Tuin and Tels 1990). Therefore, strong acids tend to destroy soils because their higher tendency to lose protons as compared to weak organic acids. Four to 5 times more macronutrients were lost with EDTA as compared to citrate extraction (Wasay et al. 1998a and 1998b). Furthermore, EDTA is generally adsorbed on the surface of soil minerals and makes the soil unfit for further use because it is relatively non biodegradable (Tokunaga 1996).

The microbial oxidation or reduction of metals has been used to remediate soils or sludges contaminated by heavy metals. Microorganisms such as *Thiobacillus ferrooxidans* and sulfate reducing bacteria are capable of leaching heavy metals from contaminated soils and sewage sludge. *T. ferrooxidans* oxidize most metal sulfides to soluble metal sulfates except for those of Pb and Hg. The bioleaching process is suitable when metal mining waste or heavy metals are present as sulfide and conditions are anaerobic. Therefore, it is difficult to remove all metals from soils by such microbial process. Currently, bioleaching processes are used to leach metals from mining ores (Means and Hinchee 1994). However, the sulfuric acid produced by the microbial process also leaches the nutrients along with the heavy metals by lowering the soil pH. A secure disposal facility is required afterwards to protect groundwater against acidification. Biological activity used for the mobilization of heavy metals is analogous to soil washing or chemical leaching because microorganism produce strong acid such as sulfuric acid (Means and Hinchee 1994). Therefore, environmentally friendly technologies are required for bioremediation.

Citric acid and/or its salt, among other weak organic acids, is quite effective in remediating soils polluted by heavy metals (Wasay et al. 1998a and 1998b). The importance of using weak organic acids and/or their salts are: i) less destructive to soil properties as compared to strong acids (HCl, HNO_3, H_2SO_4) and strong chelating agents (EDTA, DTPA) ii) improve the soil properties by enhancing the formation of stable soil aggregates.

Several microorganisms such as yeast and fungi accumulate considerable amount of organic acids such as citric, oxalic, malic, tartaric, fumaric and pyruvic acids (Rehm and Reed 1983). Some organisms have conversion rate of organic acids reaching up to 90% of the carbon supplied (Steinbock 1991). Citric acid is a common metabolite in most living organisms such as *Aspergillus niger* and the factors influencing its accumulation are pH, time and concentration and type of sugar.

The objective of the present study was to test the capability of the fungus *Aspergillus niger* in producing weak organic acids for heavy metal washing from contaminated soils.

8.3 Materials and Methods

8.3.1 Soil characterization

The experimental soils (a clay loam, a loam and a sandy clay loam) were characterized physically for their particle size, bulk and particle densities, porosity and saturated hydraulic conductivity, k_s (Carter 1993, Klute 1986) and chemically for their pH, electrical conductivity, cation exchange capacity (CEC), organic matter and heavy metal content.

The soil pH was measured by soaking in deionized water at a ratio of 1:2. The electrical conductivity (EC: mS/cm) of the soil supernatant in deionized water was measured using an electrical conductivity meter. The CEC was calculated by exchanging heavy metals and other cations present in the contaminated soils with a $BaCl_2$ solution (Carter 1993). All metals were determined by ICP-AES (ICPS-1000 II, Shimadzu Corp., Kyoto, Japan), except for Na and K which were determined by flame emission spectrophotometry (Perkin Elmer).

8.3.2 Medium for fungus

The growth medium for the *Aspergillus niger* was prepared by dissolving the following constituents in sterilized deionized water: $(NH_4)_2SO_4$ 1.0%, K_2HPO_4 0.3%, $MgSO_4.7H_2O$ 0.05%, KCl 0.05%, $FeSO_4.7H_2O$ 0.001%, Sucrose 5.0%, Agar 2.0%, pH 6.0.

8.3.3 Microbes culture preparation

Pure dry spores of *Aspergillus niger* (IFO, Japan) mixed with a few ml of sterile deionized water and a few drops of this culture was transferred to a slanted tube containing agar and autoclaved growth medium. The *Aspergillus niger* was grown in the slanted tube for 5 days at 30 °C.

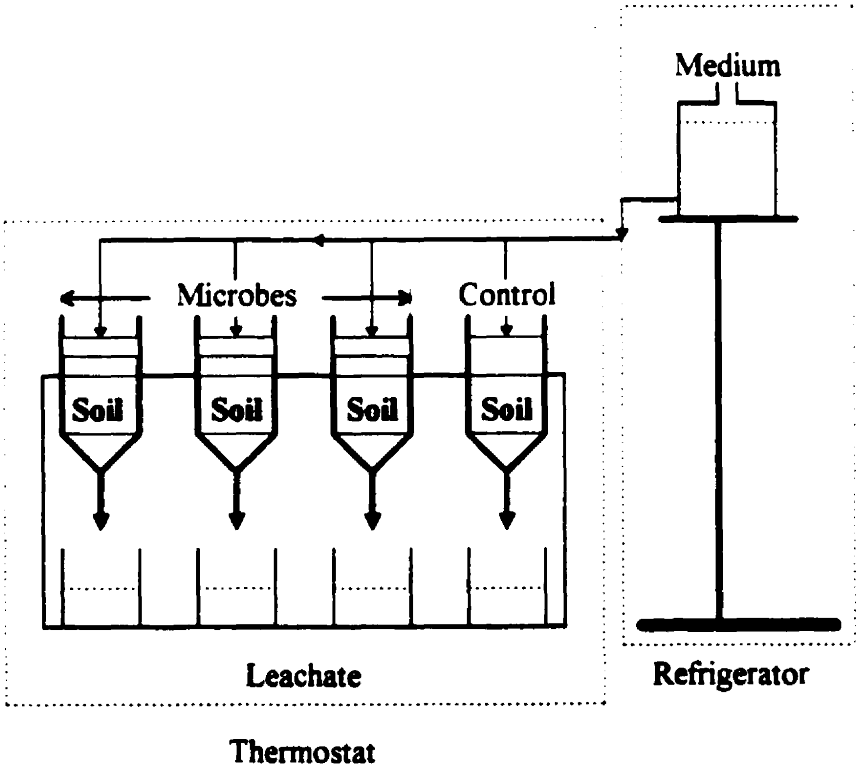

Figure 8.1 Schematic diagram for bioremediation of soil. Cultivation of *Aspergillus niger* on the surface of contaminated soil column.

8.3.4 Remediation procedure

Three experimental soil columns and a control column for each soil were set up for metal flushing study (Figure 8.1). Either 3.5, 5.0 or 5.0 g of clay loam, loam or sandy clay loam was used to fill each column, respectively. The bottom and the top of the column was sealed with glass wools to prevent the migration of fine particles. The soil columns were leached with 2 pore volumes of deionized water to remove the air trapped in the soil pores. The top surface of the soil was covered by a few cm of coarse straw. The *Aspergillus niger* culture prepared in the slanted tube was mixed with 10 ml of medium and transferred to the soil columns and kept at a constant temperature controlled by an air conditioning system. Each column was directly connected to a reservoir containing the medium. The *Aspergillus niger* produced mainly citric acid and some oxalic acid which was slowly percolated through each soil column. The leachate in terms of pore volume was collected every day from each column for heavy metals analysis. The citrate production was also tested using p-dimethyl-aminobenzaldehyde in acetic acid to develop pink color with citric acid indicating its production in the system (Rohr et al. 1979). After flushing soil column, the residual heavy metals in the soil was also determined by acid digestion as discussed in Chapter 3.

8.3.5 Statistical analysis

The SAS repeated measures analysis of variance procedure was used to compared the rate of leaching among different heavy metals at a optimum pH condition (SAS 1990).

8.4 Results and Discussions

8.4.1 pH effect on the organic acid production of *Aspergillus niger*

The organic acids produced by *Aspergillus niger* decreased the pH of the leachate to 2. Thus, NH_4OH was added to the growth solution in sufficient quantities to maintain the pH of the leachate between 3.8 to 4.0 (Figure 8.2a, 8.2b and 8.2c). A pH of 5.0 would have lead to the production of oxalic and gluconic acids which may have immobilized Pb rather than enhance leaching. Also, about 20 to 30% more leaching of metals occurred at a pH about 4 as opposed to a pH 2. The optimum pH for maximum heavy metal leaching ranges between 5 and 7 but only 5 to 10% more Pb extraction is achieved at the optimum pH range as compared to a pH 4 (Wasay et al. 1998a and 1998b). Also the pH range of 3.8 to 4.0 optimizes the production of citric acid by *Aspergillus niger*, rather than oxalic and gluconic acids.

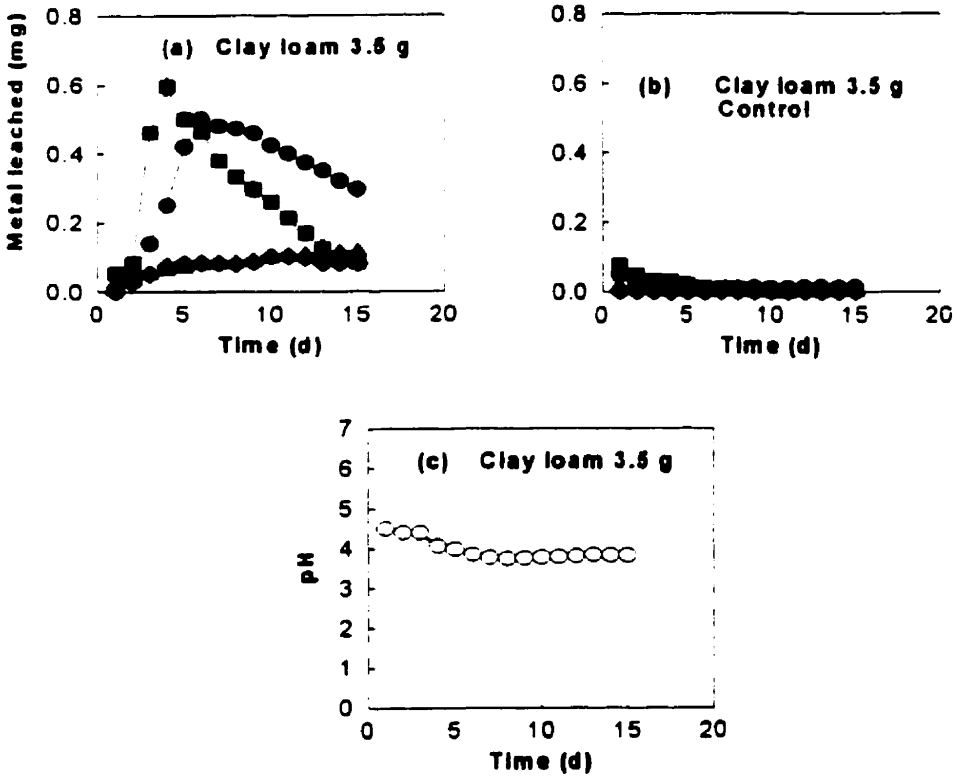

Figure 8.2 Leaching of heavy metals from clay loam with time. (a) by *Aspergillus niger*, (b) control without *Aspergillus niger* and (c) pH change in experiment (a). ◆ Cr, ■ Hg, ▲ Mn and ● Pb.

8.4.2 *Aspergillus niger* and heavy metal mobilization

The three experimental soils are described in Table 8.1. According to the Quebec guidelines (MEFQ 1994), these soils cannot be disposed on agricultural land because their high heavy metal content. The growth of *Aspergillus niger* for citric acid production *is* quite sensitive to heavy metals (Steinbock 1991). *Aspergillus niger* can be grown under either submerge or surface cultivation. Submerge cultivation is useful only when their is no contamination of heavy metals in the soil otherwise surface cultivation is preferred.

Table 8.1 Physical and chemical characterization of contaminated soils.

Soil characteristic	Clay loam	Loam	Sandy clay loam
Particle size			
% sand	29.0±0.6	50.0±0.5	53.5±0.9
%Silt	36.0±0.4	30.0±1.2	24.0±1.6
% clay	35.0±1.0	21.0±0.7	23.0±0.7
Bulk density (g/cm^3)	0.8±0.0	0.9±0.0	0.9±0.0
Particle density (g/cm^3)	2.0±0.0	2.0±0.0	2.1±0.1
Total porosity (%)	45.4±0.1	44.1±0.2	48.0±0.4
k_s(cm/s)	1.1±0.1x10^{-4}	1.3±0.1x10^{-4}	4.0±0.2x10^{-4}
EC (mS/cm)-water	9.2±0.2	8.0±0.2	2.3±0.2
pH-water	4.2±0.0	5.1±0.1	6.3±0.1
-$CaCl_2$	4.3±0.1	5.2±0.1	6.0±0.0
CEC (cmol(+)/kg)	2.0±0.2	4.7±0.0	12.2±0.2
Organic matter (%)	9.9±0.1	5.4±0.1	7.1±0.1
Heavy metals (mg/kg)			
Cr (Total)	840	-	-
Mn	868	-	-
Hg	1280	-	-
Pb	1664	2797	1036
Cd	-	1821	794
Cu	-	-	926
Zn	-	-	1029

About 90% of the sugar fed to *Aspergillus niger* can be converted to citrate (Steinbock, 1991). Therefore, a 5% sucrose solution was presumed sufficient to produce enough citrate (about 0.2 M) for the leaching of heavy metals from these contaminated soils. The optimum concentration of citrate ranges from 0.03 to 0.16 M for maximum heavy metal leaching (Wasay et al 1998a and 1998b). The effect of time and heavy metal mobilization for the three soils is shown in Figure 8.2, 8.3, 8.4. Cadmium, Zn, Cu, Pb and Hg showed similar leaching behavior while Cr and Mn showed relatively slow leaching behavior.

For the clay loam, Cr, Mn, Pb and Hg were leached to 37, 41, 85 and 91%, respectively after 15 days at 30 °C (Figure 8.2a). The leaching of Pb and Hg was found to peak at day 4 with the maximum yield of citric acid by *Aspergillus niger* at about pH 4 (Figure 8.2c). After 15 days of leaching, soil Pb levels reached the B soil criteria of the Quebec guidelines (MEFQ 1994). Lead is the least mobile of all heavy metals, especially in soils under reducing condition. Lead has a particularly high affinity for Mn oxides and Pb(II) oxidized to Pb (IV) is quite insoluble (McBride 1994). The level of Hg met only the C soil criteria of the Quebec guidelines. By extrapolating the results, at least 20 to 21 days of leaching are required to reach the A soil criteria level for both Pb and Hg. Chromium and Mn leached very slowly and thus, an even longer period would be required to reach the permissible A soil criteria level. Chromium is usually present in soils in its trivalent state (Bartlett and Kimble 1976). The organic matter of polluted soils can reduce Cr(VI) to Cr(III) (Grove and Ellis 1980, James et al. 1983). Chromium (III) forms chromium hydroxides and ultimately, the very insoluble chromium oxide, Cr_2O_3. The possible formation of polymeric chromium hydroxy complexes reduces its solubility even more (Wilkinson 1987). This may be the reason why Cr leached slowly with citrate. When Cr is present in soil as Cr(III), an oxidizing agent is the preferred extractant. The Cr(VI) is known to be much more mobile in soils, although it can be reduced back to Cr(III) by organic matter. The slow leaching of Mn may result from the formation of Mn-Fe oxides. In this experiment, Mn was probably released slowly from the soil by spontaneous dissolution or cation exchange, especially under acidic or reducing conditions. The control experiment leached only 0.7, 2.5, 3.9 and 6.7% of Cr, Mn, Pb and Hg, respectively from the clay loam (Figure 8.2b).

For the loam, Cd and Pb were leached to 99.7 and 83%, respectively after 15 days at 30 °C, whereas for the control treatment, Cd and Pb were leached to only 13 and 2%, respectively (Figure 8.3a and 8.3b). The Cd leaching peaked at day 4 with the production of citric acid by *Aspergillus niger*. At the beginning, Pb removal was very slow as oxalic acid was produced. When the pH dropped below 5 (Figure 8.3c), citric acid production started and Pb leaching reached a peak at day 7. To improve Cd levels from the B to the A soil criteria, an additional 5 days of leaching was required. Lead

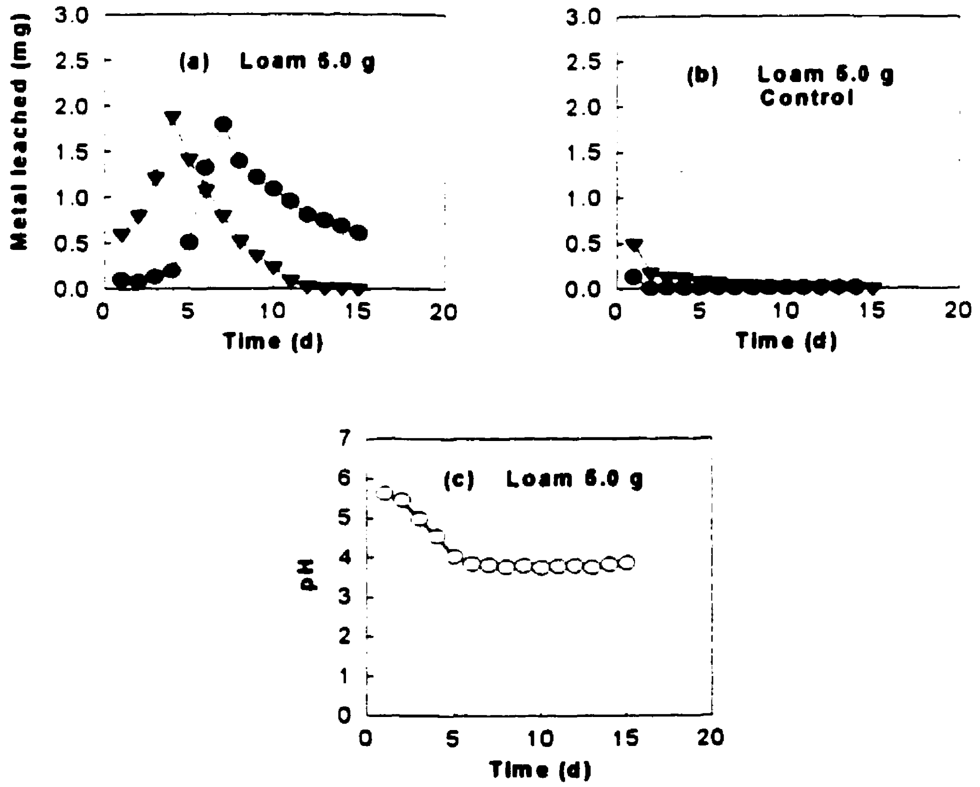

Figure 8.3 Leaching of heavy metals from loam with time. (a) by *Aspergillus niger*, (b) control without *Aspergillus niger* and (c) pH change in experiment (a). ▼ Cd and ● Pb.

levels met the B soil criteria after 15 days and the A soil criteria was expected after an additional 8 to 10 days.

For the sandy clay loam, Cd, Cu, Pb and Zn were leached to 99, 94, 58 and 99%, respectively after 15 days at 30 °C whereas for the control experiment, only 3 to 5% for all these metals were leached (Figure 8.4a and 8.4b). The leaching of Cd, Cu and Zn peaked at day 4 with the citric acid production by ***Aspergillus niger*** and then slowly decreased to meet the A soil criteria at day 15. The leaching of Pb was slow at the beginning as oxalic acid was produced. When the pH dropped below 5 (Figure 8.4c), Pb

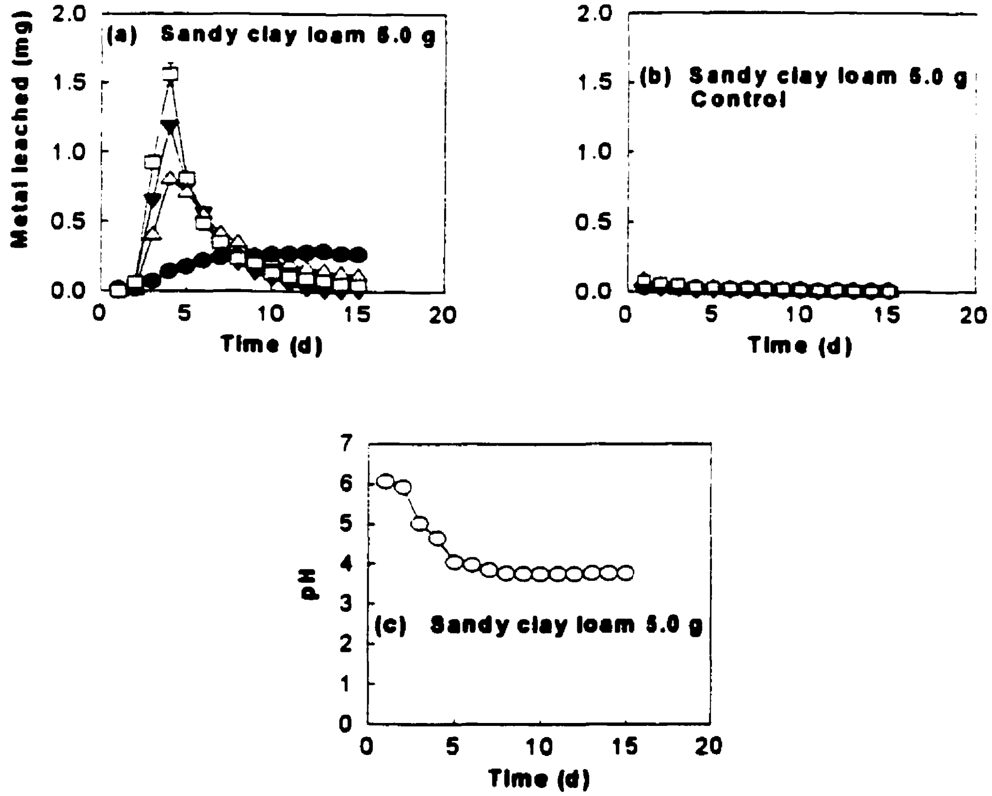

Figure 8.4 Leaching of heavy metals from sandy clay loam with time. (a) by ***Aspergillus niger***, (b) control without ***Aspergillus niger*** and (c) pH change in experiment (a). ▼ Cd, ● Pb, Δ Cu and □ Zn.

leaching was enhanced. Cadmium level was still high after 15 days, and an additional 7 to 8 days of leaching would have been required to meet the A criteria for soils by applying kinetics for preduction. Copper and Zn both met the A soil criteria after 15 days. Lead levels met the B soil criteria and was expected to meet the A soil criteria after an additional 10 days of leaching by applying kinetics for preduction.

The extent of soil contamination is directly related to the initial metal concentration in the soil as well as the presence of inorganic and organic compounds. If the same treatment levels are imposed, sites contaminated by small amount of heavy metals are open just as difficult to remediate as those grossly contaminated because the binding energies associated with low sorption densities are large. Assuming that soils have multiple types of surface sites, small quantities of metals would be preferentially adsorbed by the sites with the highest binding energies, making their subsequent release difficult (Reed et al. 1995). This phenomenon is illustrated by the slow release of Pb from sandy clay loam.

8.4.3 Statistical analysis

Citrate produced by *Aspergillus niger* had a highly significant effect on the rate of leaching of heavy metals from all three experimental soils ($P < 0.005$).

With the clay loam, Hg leaching by *Aspergillus niger* was faster for the first 5 days followed by Pb, Cr and Mn. From day 5 to 13, the leaching of Pb was predominant followed by Hg, Mn or Cr. After 13 days, Mn leached slightly faster than Hg but still more slowly than Pb. With the loam, Cd leached out faster than Pb for the first 5 days after which Pb leaching dominated. With the sandy clay loam, Zn, Cd and Cu leaching was predominant for the first 7 days, after which Pb was leached more slowly as compared to the other three metals.

8.5 Conclusions

The following conclusions were reached:

1. A bioremediation process using *Aspergillus niger* was proven to be effective for the leaching of heavy metals from three soils naturally contaminated by heavy metals.

2. All levels of heavy metals can meet the A soil criteria after 20 to 25 days of leaching time, at optimum pH of 3.8 to 4.0, and at 30 °C.

3. Most metals (Cd, Hg, Cu, Zn) reached peaked at day 4 with continuous supply of citrate by *Aspergillus niger*. The maximum leaching rate for Pb was reached after 5 to 7 days when oxalate supply by *Aspergillu niger* was reduced as pH dropped below 4. Chromium and Mn were leached very slowly by citrate and took more time to meet the B soil criteria.

4. For the clay loam and after 15 days, Cr, Mn, Pb and Hg were leached to 37, 41, 85 and 91%, respectively. For the loam and also after 15 days, Cd and Pb were leached to 99.7 and 83%, respectively. For the sandy clay loam and after 15 days, Cd, Cu, Pb and Zn were leached to 99, 94, 58 and 99%, respectively.

5. This bioremediation process was found to be environmentally friendly as compared to the leaching process using the bacteria T. *ferroaxidans*, strong acids or chelating agents.

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CONNECTING STATEMENT

This final experimental chapter deals with the treatment aspects of leachate generated in the previous chapters (soil washing) using granular activated carbon, ferric chloride solution and granular activated alumina. Granular activated carbon was found to be quite effective in removing metal chelates from the leachate. The treatment of the leachate was carried out at different pH levels to optimize the condition for maximum removal of metal chelates. The kinetic and adsorption capacity of granular activated carbon for metal chelates in the leachate were also studied at optimum pH and constant temperature.

This paper has been accepted for publication in the *Journal of Water, Air and Soil Pollution*. 1998. Authors: Wasay, S. A., Barrington, S. and Tokunaga, S. The contribution of authors are: i) First author carried out entire experimental research work and manuscript write up ii) Second author supervised research work and manuscript correction iii) Third author provided his analytical laboratory facilities and manuscript correction.

CHAPTER 9

TREATMENT OF LEACHATE FROM SOIL WASHING PROCESS

9.1 Abstract

Granular activated carbon (GAC), granular activated alumina (GAA) and a ferric chloride solution (FCS) were tested for the treatment of leachate contaminated by heavy metals. The leachate was collected following the remediation of soils using weak organic acids and/or their salts, EDTA and DTPA. Only GAC was found to effectively remove heavy metals (Cd, Cu, Cr, Hg, Mn, Pb and Zn as chelates) from the leachate. At optimum pH ranging from 5.4 to 6.9, 97% of Hg was removed while at optimum pH ranging from 6.9 to 8.0, 78 to 96% of Cd, Cu, Mn, Pb and Zn were removed. Some 77% of the Cr was removed at optimum pH of 5.4. The brown color of the leachate turned colorless after GAC treatment.

9.2 Introduction

Heavy metals are effectively washed from polluted soils using weak organic acids and/or their salts (citrate, tartarate and oxalate) and chelating agents EDTA and DTPA (Wasay et al 1998b). The recovered leachate contains heavy metals in the form of metal chelates forming negatively charged complexes which are hard to treat by lime precipitation.

For wastewater treatment, various techniques have been used to remove heavy metals. Heavy metals are generally precipitated as hydroxides by adding lime or caustic soda and raising the pH until reaching minimum solubility and maximum precipitation. Heavy metals may also be precipitated as sulfides and in some cases as carbonates or by ion exchange processes (Eckenfelder 1989). Granular activated carbon (GAC) removes, by adsorption, organic materials, such as dichlorophenol. Adsorption is a common process in water and wastewater treatment and in the transport process of chemical species in aquatic systems. Aqueous solutions of Hg are often treated by various combinations of activated carbon, EDTA, tartaric acid, citric acid and calcium. In the presence of these chelating agents, activated carbon is even more effective (Thiem et al. 1976).

The objective of the present study was to test the efficiency of various adsorption media in treating leachate containing heavy metal chelates generated from the remediation of soils using weak organic acids and/or their salts, EDTA or DTPA.

9.3 Materials and Methods

9.3.1 Leachate samples

Leachate samples were collected from the three soils extracted with weak organic acids and/or their salts, EDTA or DTPA(Wasay *et al.*, 1998a and 1998b). The leachate samples were stored at 4 °C in refrigerator. The leachate samples were analyzed for heavy metals content using ICP-AES (ICPS 1000 II, Shimadzu Corp., Kyoto, Japan).

9.3.2 Adsorbents

Granular activated carbon (GL-H, Wako, Japan) was pretreated by soaking with dilute HCl (0.1 M) at 40-50 °C for 25 h, washed several times with distilled water and then dried in a vacuum desicator for a week before it was used as an adsorbent for metal chelates. A granular activated alumina (GAA) and a 5% ferric chloride solution (FCS) were also used for the treatment of metal chelates in the leachate but they required no pretreatment.

9.3.3 Procedure

A 22 ml leachate sample was taken in four sets of tubes and each set of tubes contains four plastic tubes in triplicate. After adjusting the pH of each tube between 5.4 and 8.0 using a few drops of NaOH solution (1 M), the total volume in each tube was adjusted to 25 ml. A 2 g of GAC was added to each tube of the first set while each tube of the second and third sets received granular activated alumina (2.0 g) and ferric chloride (2 ml), respectively. Similarly the fourth set of tubes received no adsorbent and was used as a control. The tubes were shaken for 12 h at 20 °C. The mixture was centrifuged at 10,000 rpm for 20 min. (in refrigerated centrifuged machine) and the final pH of the supernatant was measured in each tube and analyzed for heavy metals by ICP-AES. The final pH was plotted as a function of % removal of heavy metals in Figure 9.1.

A set of six tubes was prepared in triplicate with leachate sample for the measurement of adsorption kinetics. A 2 g of GAC was added to 18 tubes containing 25 ml of leachate at optimum pH. After 15, 60, 120, 240, 360 and 780 minutes of shaking at 100 rpm and 20 °C, 3 tubes were taken out and filtered to analyze heavy metals content in the supernatant by ICP-AES.

Another set of five tubes was prepared in triplicate with leachate sample (25 ml at optimum pH) for the effect of quantity of GAC for adsorption capacity. A 0.25, 0.325, 0.500, 1.013 and 1.5 g of GAC was added to each tube containing 25 ml leachate in triplicate. The set of five tubes in triplicate was shaken for 12 h at 100 rpm and 20 °C in a shaker. Then the tubes were filtered and filtrates were analyzed for heavy metals by ICP-AES.

9.4 Results and Discussion

9.4.1 Characterization of leachate

The leachate from the soil washing process using weak organic acids and/or their salts, EDTA or DTPA, was found to contain 36.4, 15.0, 7.4, 2.5, 32.5, 76.3 and 13.8 mg/l of Cd, Cr, Cu, Hg, Mn, Pb and Zn, respectively. The color of the leachate was dark brown and its pH was 4.4.

9.4.2 pH effect on the removal of metal chelates

The removal of heavy metals from the leachate by adsorption on GAC was highly pH dependent because pH affects the surface charge of the adsorbent and the degree of ionization and speciation of adsorbent (Figure 9.1). By increasing the pH from 5.4 to 6.9, Hg removal reached 97% but fell to 89% as the pH was further increased to 8.0. The removal of Cd, Cu, Mn, Pb and Zn increased as pH increased from 5.4 to 8.0. The removal efficiency increased from 55 to 88% for Cd, 50 to 86% for Cu, 59 to 78% for Mn, 69 to 95% for Pb and 55 to 84% for Zn as the pH was increased from 5.4 to 8.0. As pH increased from 5.4 to 8.0, the removal of Cr was decreased from 77 to 18%. The adsorption of metal at the solid surface may be governed by the presence of metal complexes rather than free metal ions (Wasay et al. 1995, Bourg 1988, Thiem et al 1976). Other adsorbents (GAA and FCS) were also used for the removal of these heavy

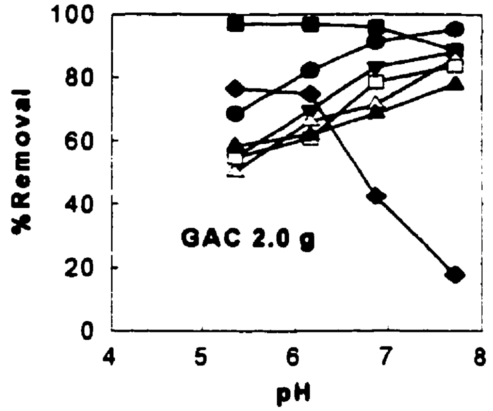

Figure 9.1 pH effect on the removal of metal chelates by GAC Cd ▼, Cr ◆, Cu Δ, Hg ■, Mn ▲, Pb ● and Zn □.

metal chelates. Removal by GAA at pH 8.0 was 27% for Cd, 9% for Cu, 31% for Cr, 58% for Hg, 33% for Mn, 36% for Pb and 34% for Zn. By adding ferric chloride (2.0 ml) to the leachate, heavy metal removal reached levels of 11% for Cd, 40% for Cu, 14% for Cr, 82% for Hg, 16% for Mn, 18% for Pb and 14% for Zn at optimum pH of 6.8.

For the control experiment without adsorbent, hydrolysis (precipitate) of metal chelates was found to range between 5 and 8% in the pH range of 5.4 to 8.0. Even at a pH of 9.5, removal rate was found to be 16, 9, 10, 52, 15, 15 and 12% for Cd, Cu, Cr, Hg, Mn, Pb and Zn, respectively.

The adsorption of heavy metals on GAC resulted from the development of positive charges on the surface of the carbon and the presence of negative charged on the metal chelates of the leachate. The functional oxidized groups present on the surface of the carbon particles play a major role in removing metal chelates from the leachate (Wasay et al. 1995). The mechanism involved at the granular activated carbon water interface is:

$$C_xO + H_2O \longrightarrow C_x^{2+} + 2OH^- \qquad (9.1)$$

$$C_xO_2 + H_2O \longrightarrow C_xO^{2+} + 2OH^- \qquad (9.2)$$

The adsorption reaction may take place in the following manner:

$$C_x^{2+} + MY^{n-} \longrightarrow C_x\text{-}MY^{(2-n)} \qquad (9.3)$$

$$C_xO^{2-} + MY^{n-} \longrightarrow C_xO\text{-}MY^{(2-n)} \qquad (9.4)$$

where MY represents the metal chelates in the leachate.

9.4.3 Adsorption rate constant

The rate constant for the adsorption of metal chelates on the GAC was determined using Lagergren's equation (1898):

$$\log(q_e - q) = (\log q_e) - \frac{k_{ad} * t}{2303} \qquad (9.5)$$

where q_e and q (both in mg/l) are the amount of metal chelates adsorbed at equilibrium and at time t (min), respectively, and k_{ad} (min^{-1}) is the rate constant for adsorption of metal chelates. The value of k_{ad} was measured at 20 °C from the slope of the linear plot of log(q_e-q) vs time, for the total metal content of the leachate and was found to be 0.01 min^1 (Figure 9.2).

9.4.4 Adsorption isotherm

The analysis of the equilibrium data for the adsorption of metal chelates on GAC has been carried out in the light of the rearranged Langmuir adsorption model:

$$\frac{C_e}{q_e} = \frac{1}{Q^0 b} + \frac{C_e}{Q^0} \qquad (9.6)$$

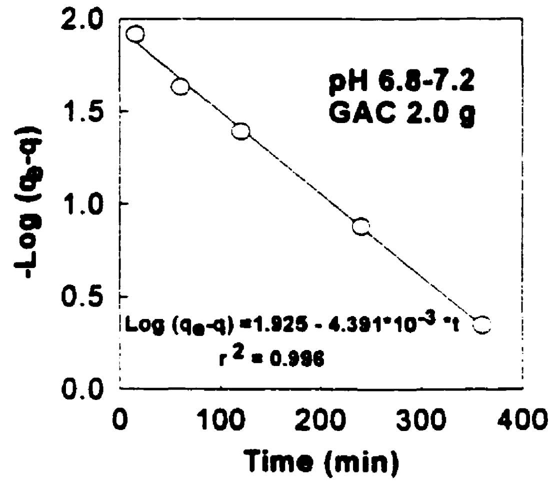

Figure 9. 2 Lagergren plot for the adsorption rate of metal chelates.

where C_e (mg/l) is the equilibrium concentration of metal chelates and Q^0 (mg/g) and b (l/mg) are the Langmuir constants related to the capacity and energy of adsorption, respectively. In Figure 9.3, the linear relationship between C_e/q_e and C_e, at 20 °C, suggests the applicability of the above model for the present system. It also suggests the formation of a monolayer of metal chelates at the outer surface of the GAC (Gupta et al. 1990). This was also experimentally confirmed by extending the time of adsorption from 780 to 1440 min, wherein the amount of adsorption remained the same and no multilayer curve was obtained. The adsorption capacity (Q^0) and b at 20 °C were found to be 19 mg/g and 0.315 l/mg, respectively.

After treatment with GAC, the brown color of the leachate had disappeared leaving a colorless liquid.

9.5 Conclusions and Summary

The following conclusions may be drawn from the present investigation:

1. GAC can effectively adsorb metal chelates present in the leachate following soil remediation with weak organic acids and/or their salts, EDTA or DTPA, whereas GAA and FCS were ineffective.

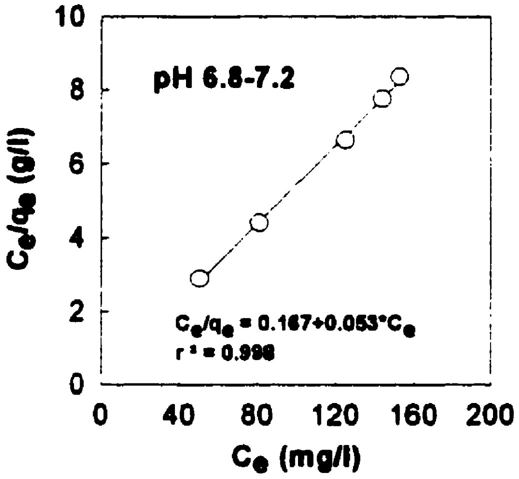

Figure 9.3 Langmuir plot for the adsorption capacity of metal chelates.

2. With GAC, optimum pH for maximum Hg removal was found to range from 5.4 to 6.9 whereas for Cd, Cu, Mn, Pb and Zn, it was found to range from 6.9 to 7.7. Maximum removal of Cr occurred at a pH of 5.4 to 6.2. Metal chelates removal efficiency ranged from 77 to 97%, at these optimum pHs.

3. The present data fits the Langmuir model and therefore confirms the formation of a monolayer of metal chelates at the outer surface of the GAC.

4. The values of k_{ad}, Q^0 and b were found to be 0.01 min^{-1}, 19 mg/g and 0.315 l/g, respectively.

5. The leachate solution was also treated to a colorless liquid during its treatment for heavy metals removal.

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CHAPTER 10

SUMMARY AND CONCLUSIONS

For the leaching of heavy metals from three naturally polluted soils, a study was undertaken using weak organic acids and/or their salts. The performance of these acids was compared against that of synthetic chelating agents, EDTA and DTPA. Based on the findings of this study, the following final conclusions were drawn:

1. To evaluate the possible remediation process, three soils of different texture, polluted by heavy metals were tested for forms of metal retention. The soils were initially characterized for particle size, bulk and particle densities, porosity, saturated hydraulic conductivity, electrical conductivity, pH, sulfur, organic matter and presence of heavy metals. The heavy metals in the soil were bound tightly to Mn/Fe oxides and the organic fractions except for Cd and Zn which were mainly present in an exchangeable form.
2. Citrate and tartarate showed high removal capabilities within 12 h of retention time. Their efficiency was dependent on pH for the clay loam and especially for the leaching of Hg and Pb whereas EDTA and DTPA were ineffective to remove Hg within the same pH range studied. Other metals such as Mn and Cr were difficult to remove because they were tightly bound to the soil fractions, but citrate and tartarate removed them more effectively than EDTA and DTPA. Four extractions with citrate and tartarate would allow the polluted clay loam to meet the Quebec category A for its disposal on agricultural land. Weak organic acids and/or their salts were found to offer an environmentally friendly remediation technique because an improved soil structure was observed after treatment. Furthermore, citrate desorbed significantly less macronutrients (Ca and Mg), compared to EDTA.
3. For loam and sandy clay loam, citrate and tartarate, and oxalate together with citrate, were able to remove Cd, Cu, Pb, Zn effectively within 12 h of retention time. Desorption or leaching efficiency of heavy metal was found to be pH dependent. In comparison, the synthetic chelating agents EDTA and DTPA also removed these heavy metals effectively. With these organic salts and EDTA, Zn levels met the Quebec soil clean up

criteria (A level) after two extractions whereas Cu and Pb levels reached to B. Probably, two more extractions with these weak organic salts are required to meet the Quebec level A soil clean up criteria for Cd, Cu and Pb. EDTA is less prone to biodegradability and is adsorbed during soil cleaning process leaving the soil polluted after cleaning. It also removed higher amounts of soil macronutrients as compared to citrate. The organic acids or their salts are adsorbed during the soil cleaning process. However, due to their low molecular weight, biodegradability and nontoxic in nature, they are environmentally friendly for soil remediation as well as improved the soil structure and enhanced the formation of stable granular aggregates.

4. The rates of leaching of heavy metals from the three soils were analyzed using a two-reaction model based on the extraction data obtained with citrate, tartarate and EDTA solutions at a constant pH, concentration of extractants, temperature and shaking speed. For the high leaching or desorption rate (k_1), Cd leached faster with all three extractants (citrate, tartarate and EDTA) followed by Zn, Cu, Pb, Mn, Hg and Cr at 20 °C. The slow leaching rate (k_2), the same order of leaching was followed by these metals. The k_1 rate results from the release of the exchangeable fraction of metals especially for Cd and Zn and then the k_2 rate results from the release of metals such as Cu, Pb, Hg, Mn and Cr more tightly bound by oxides and the organic fractions of soils. The values of m_1 (quasi-equilibrium constant) for all soils were very small indicating high removal efficiency of metals from all soils. A rough trend was obtained between α_0 and the metal leaching depending on the retention form of metals in the soil. The value of $\alpha_0 < 0.5$ indicated the fast leaching of metal, whereas α_0 is greater than 0.5 for Pb, Cr and Mn indicated difficult to leach out form the soil.

5. The three experimental soils were flushed in columns using citrate, tartarate, oxalate+citrate, EDTA and DTPA as flushing solutions. The clay loam and loam were remediated to C and B level of the soil clean up criteria after 20 pore volumes within 10 to 15 h of flushing time. Similarly for sandy clay loam, heavy metals in soil reached to C and B level of the soil clean up criteria after 10 to 15 pore volumes within 9 to 15 h of flushing time. Therefore, more flushing is required to meet the A level of soil clean up criteria.

6. A bioremediation process using fungus, *Aspergillus niger* was proven effective for the leaching of heavy metals from the three naturally contaminated soils. With *A. niger* all levels of heavy metals can meet the A criteria for soils after 20 to 25 days of leaching time, at optimum pH of 3.8 to 4.0 and 30 °C. Chromium and Mn were very slowly leached out by citrate and took more time to meet the B criteria for soils. This bioremediation process was found to be environmentally friendly and cost effective. The roughly estimated cost of present bioremediation is at Can $ 2 to 3 of substrate per ton of soil as opposed to strong acid (HCl) for the present standard techniques (Can $ 70 depending on the pollution load).

7. Granular activated carbon was found to be the most effective adsorbent for metal chelates present in the leachate following soil remediation with weak organic acids and/or their salts, EDTA or DTPA. The metals were removed between 77 to 97% from the leachate by GAC. The brown leachate solution was also changed to a colorless liquid after treatment indicating that the wastewater containing metal chelates is cleaned enough to discharge into water body.

10.1 Recommendation for Future Research

This research is limited to three Japanese soils namely clay loam, loam and sandy clay loam because laboratory facility was used in Japan. The laboratory research was successfully conducted only at a small scale level in batch and column experiments. Various experimental conditions were optimized for maximum removal of heavy metals from soils. Bioleaching experiment using *Aspergillus niger* had also been studied successfully to remediate these soils. The encouraging results of this study suggest that the research in future should be pursued as follows:

1. A large scale level experiment should be conducted for a site contaminated by heavy metals in Quebec using natural materials such as citrate and tartarate in a laboratory column as well as in field.
2. Since bioremediation is cost effective and environmentally friendly, further studies on bioleaching of soil polluted by heavy metals using *Aspergillus niger* should be conducted

on a large scale level in a laboratory column and in field by growing the fungus on the surface of the soil.

CHAPTER 11

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