Full Length Research Paper

The fate and bioavailability of some trace elements applied to two vegetable farms in Addis Ababa

Fisseha Itanna[1]*, Jörn Breuer[2] and Mats Olsson[3]

[1]Department of Biology, Science Faculty, Addis Ababa University, P. O. Box 1176, Addis Ababa, Ethiopia.
[2]Federal Agency for Agricultural Chemistry, University of Hohenheim, D-70593 Stuttgart, Germany.
[3]Department of Soil and Environment (SLU), P. O. Box 7001, S-750 07 Uppsala, Sweden.

Metal (Cd, Cr, Cu, Hg, Ni, Pb and Zn) and metalloid (As) status of two urban farms in Ethiopia were determined using the European Community Bureau of Reference (BCR) modified sequential extraction method. Cr, Ni, and As at Kera and Ni at Kolfe increased with depth likely due to leaching. Although total Cr, Ni (at both farms) and Pb (at Kera) exceeded normal limits, the bioavailability of these elements was restricted. Most of the trace elements at Kera (As, Cr, Cu, Ni, and Zn) and Kolfe (As, Cd, Cu, Ni, and Zn) were in residual fractions followed by reducible fractions. Pb at both sites was largely found in reducible form. Oxidizable and acetic acid extractable fractions were generally the least in proportion. Oxidizable Cr, Cu and Pb at both sites were higher than their acetic acid extractable counterparts, while acetic acid extractable Cd (at Kera) and Ni and Zn at both sites, exceeded the respective oxidizable metals. Bioavailability and toxicity of metals are sometimes reduced this way through natural means. However, change in soil pH, oxidation/reduction reactions, or similar mechanisms could cause shift in metal bioavailability. Hence, regular monitoring of the soil conditions at the farms is recommended.

Key words: Bioavailability, leaching, sequential extraction, trace elements.

INTRODUCTION

Addis Ababa, the capital and largest city of Ethiopia has a population of over 3 million inhabitants. With about 3% annual population growth and constantly increasing industrial activities, the city is facing environmental pressure from urbanization and industrialization.

Urban agriculture, particularly vegetable farming has a significant place in the city, where over 160 hectares of land is annually used to grow vegetables. Little or no treatment is given to the municipal and industrial wastes that enter the major rivers in the city, which are used to irrigate the vegetable farms. Therefore, the city's major vegetable farms are continuously contaminated with different pollutants.

Two of the biggest vegetable farms are located at Kera and Kolfe, in the southeastern and on the western side of the city, respectively. The vegetables produced in these farms are sold in the open market to every city dweller

and some are sold directly to hospitals, restaurants and similar other public and private institutions.

Preliminary studies made so far, indicate that there is an increasing trend of metal uptake by the vegetables grown on such urban farms (Itanna, 1998a; Itanna, 2002). Therefore, there is a growing concern of food quality and safety when vegetable products are obtained from contaminated urban soils.

Studies on soil quality should call for better land use planning for sustainable utilization of the scarce soil resources (Karlen et al., 2003). Soils under repeated contamination from different pollutants, eventually lose their resilience and are unable to provide good quality food products. They can thus pose a potentially hazardous situation for human health and the environment. An appraisal of the extent of contamination of such urban farms is thus of paramount importance to suggest alternative remediation measures.

The bioavailability of contaminants is increasingly used as indicator of potential risk. Sequential extraction is widely used to estimate the amounts and proportions of metals in soil and to predict bioavailability and metal

---

*Corresponding author. E–mail: fissehai@yahoo.com

leaching (Jalali and Khanlari, 2008).

Preliminary studies in the past in Addis Ababa addressed only total soil metal composition (Itanna, 1998b; Itanna, 2002). There have been so far very limited studies and knowledge in Ethiopia pertaining to chemical speciation of metals and the risk to humans and the environment (Fitamo et al., 2007). Studies pertaining to heavy metal speciation patterns in the tropics seem to be generally few (Doelsch et al., 2008; Silveira et al., 2006).

Therefore, this study was initiated to determine the proportions of total trace elements and sequentially extracted metal species, their mobility, bioavailability and their projected risk on human life and the environment. Moreover, trace element proportions at the surface shall be compared with those in the subsoil which may explain possibilities of leaching of trace elements through the soil column, among other things.

MATERIALS AND METHODS

Soil sampling and site descriptions

Kera and Kolfe farms are situated in Addis Ababa, the capital of Ethiopia, which is about 2300 m above sea level. The soil at Kera is a Fluvisol, while the one at Kolfe is a Nitisol. Annual rainfall in Addis Ababa is generally between 800 - 1000 mm. The mean annual temperature of Addis Ababa is about 15^oC.

Four (1/2 m depth) pits were dug in each farm, from which surface (0 - 20 cm) and subsurface samples (30 - 50 cm) were collected. These soil samples represent four replications of surface and subsurface soil samples per site. The samples were later air dried and sieved to pass a 2mm mesh sieve for different analyses.

Kera and Kolfe farms are two of the biggest vegetable farms in Addis Ababa that distribute their products to all parts of Addis Ababa city.   Kolfe farm is situated close to the Kolfe Comprehensive High School on the western side of the city, while Kera farm is situated near the former Kera Abattoir, in the southeastern part of the city.

Some chemical and physical analyses

Particle size fractions, pH, organic carbon, carbonate and effective CEC (ECEC) were determined.

The particle size fractions were determined according to ISO 11277 (1995). The samples were treated with hydrochloric acid and hydrogen peroxide to remove carbonates and organic matter, respectively and then dispersed in a solution of 33 g sodium hexametaphosphate and 7 g sodium carbonate in 1 litre of water. The sand fraction (63 - 2000 µm) was separated by passing the suspension through a sieve with 63 µm mesh size and weighed. The clay fraction (< 2 µm) was determined by sedimentation using the pipette method. Silt (2 – 63 µm) was calculated as the difference from the total minus the clay and sand fractions.

pH was measured in a 1:2.5 (sample/solution) suspension of soil in 0.01 M $CaCl_2$-solution using a glass electrode. Exchangeable cations were extracted with an unbuffered 1 M ammoniumchloride solution. The cations were determined by inductively coupled plasma optical emission spectrometry (ICP-OES; Varian Vista Pro) using radial torch viewing and external calibrations. The effective CEC represents the sum of all cations.

Carbonates were determined by a manometric method after dissolution with 6 M HCl (Skinner et al. 1959). Organic carbon and total nitrogen were determined by dry combustion combined with IR-spectrometry (elemental analysis) using an elemental analyser (Vario EL, Elementar, Germany). Carbonate carbon was removed by treating the samples with 6 M HCl prior to analysis (ISO 10694, 1995).

Total metal extraction

Total concentrations of metals were determined by aqua regia (AR) extraction according to a modification of ISO standard 11466.2 (Niskavaara, 1995). 15 ml conc HCl was added to soil sample in a boron silicate tube, followed by 5 ml conc. HNO_3. Samples were left overnight. Digestion was carried out at 90^oC in an aluminum heating block for 2 h. Temperature was increased stepwise. The volume was made up to 100 ml and mixed thoroughly. After settling, an aliquot of about 10 ml was decanted into a polystyrene centrifugal tube. Centrifugation was made at 1500 g for 20 min.

As, Cd, and Pb were measured by ICP-MS (Inductively coupled plasma mass spectrometry, ISO 17294-2, 2003, Perkin Elmer Sciex Elan 6000); while Cr, Cu, Ni and Zn were measured by ICP-OES (Inductively coupled plasma- optical emission spectrometry, EN ISO 11885,**1997**; Varian Vista Pro radial). Mercury was measured by cold vapour atomic absorption spectrometry using an atomic absorption spectrometer (Perkin Elmer AAS 1100B) and a flow analysis system (Perkin Elmer FIAS 400).

Sequential extraction

For speciation of metals into various fractions, a three step modification of the standardized sequential extraction procedure of the European Community Bureau of Reference (BCR) method was used (Niskavaara 1995).

Before sub-sampling, the contents of the storage bottles were shaken manually for three minutes to homogenize the soil. Then a subsample was taken using a suitable plastic spatula.

All laboratory-ware was of borosilicate glass or polyfluoroethylene (PTFE). Vessels in contact with samples or reagents were cleaned by soaking in 4 mol L^{-1} HNO_3 (overnight) and rinsed repeatedly with deionized water before use. The extractions were performed using a mechanical end-over-end shaker, at a speed of 30 ± 10 rpm and a room temperature of 22 ± 2^oC. The centrifugation was carried out at 1500 g for 20 minutes. All reagents were of analytical grade or better. Doubly-deionised water was used. For each batch of extraction, a separate 2.5 g sample of the soil was dried in layer of about 1 mm depth in an oven (105 ± 2^oC) until constant weight. From this, a dry matter content of air-dry soil was obtained. Metal concentrations were expressed per kg of dry matter (DM).

The metal concentrations were determined using ICP-OES and ICP-MS, similar to the   determination of total metal contents, depending on the concentrations of metals in the solutions. The extractable contents (mass fractions) were expressed as mg kg^{-1} on a dry matter basis for the trace elements. The three step extraction procedure was performed as follows:

Acetic acid extractable fraction (available) (Step 1): 100 mL of 0.11 mol L^{-1} acetic acid was added to 2.5 g soil in a centrifuge tube, stoppered and extracted by shaking for 16 h (overnight). The extract was separated from the solid residue by centrifugation and the supernatant liquid was decanted into a polypropylene container. The container was stoppered and the extract analyzed immediately, or was conserved by adding 1 mL conc. HNO_3 and stored in a refrigerator at about 4^oC prior to analysis. The residue was washed by adding 50 mL distilled water, shaken for 15 min on the end-over-end shaker and centrifuged for 20 minutes at 1500 g. The

Table 1. Some physical and chemical parameters of contaminated soils in Addis Ababa (means of four replicates are presented with standard error values).

Site	Depth (cm)	Particle size fractions (%)			pH	Ccarb %	Corg %
		Sand	Silt	Clay			
Kera	0-20	31. 31 ±.02	31.2 ± 1.77	37.8 ± 1.26	6.5 ± 0.18	0.5 ± 0.31	2.3 ± 0.3
Kera	30-50	27.1 ± 5.26	30.4 ± 3.16	42.5 ± 2.35	6.5 ± 0.05	b.d.l.*	1.3 ± 0.21
Kolfe	0-20	13.5 ± 1.00	32.1 ± 1.09	54.5 ± 1.25	5.5 ± 0.04	b.d.l.	2.8 ± 0.11
Kolfe	30-50	8.0 ± 0.66	27.5 ± 0.71	64.6 ± 1.26	5.4 ± 0.06	b.d.l.	1.8 ± 0.04

*b.d.l. stands for 'below the detection limit'

supernatant was then decanted and discarded, taking care not to discard any of the solids.

Reducible metal fraction (Step 2): 100 mL of a freshly-prepared 0.5 mol L^{-1} hydroxylammonium chloride (hydroxylamine hydrochloride), was added to the residue from step 1 in the centrifuge tube. The content was re-suspended by manual shaking, stoppered and then extracted by mechanical shaking for 16 h (overnight). The extract was separated from the solid residue by centrifugation and decanted as in Step 1. The extract in a stoppered polypropylene container was then retained, as before, for analysis. The residue was washed by adding 50 mL distilled water and shaken for 15 min on the end-over-end shaker and centrifuged. The supernatant was then decanted and discarded, taking care not to discard any of the solid residues.

Oxidizable metal fraction (Step 3): 25 mL of 300 mg g^{-1} (that is, 8.8 mol L^{-1}) hydrogen peroxide solution was added carefully to the residue in the centrifuge tube, in small aliquots to avoid losses due to violent reaction. The vessel was then covered loosely with its cap and the organic matter digested at room temperature for 1 h with occasional manual shaking. The digestion continued for 1 h at 85 ± 2°C in a water bath. The volume was reduced to less than 3 mL by further heating of the uncovered tube. An aliquot of 25 mL of 300 mg g^{-1} (that is, 8.8 mol L^{-1}) hydrogen peroxide was further added. The covered vessel was heated again to 85 ± 2°C and digested for 1 h. The cover was then removed and the volume of liquid reduced to about 1 mL. It was not left for complete dryness. 100 mL of ammonium acetate (1.0 mol L^{-1}) was added to the cool moist residue and shaken for 16 h (overnight). The extract was then separated from the solid residue by centrifugation and decantation as in step 1. The content was then stoppered and retained before analysis.

The residual metal fraction was calculated as the difference between the total content (AR) and the sum resulting from the three sequential extraction steps.

Ammonium nitrate extraction

The soil samples (particle size < 2 mm) were extracted for 120 min at 22°C with 1 mol/l ammonium nitrate solution using a soil to solution extraction ratio of 1: 2.5 (m/V) in conical test tubes (polypropylene) with screwing caps and an end-over-end shaker (ISO 19730, 2006). Then the closed test tubes were placed in a centrifuge and the solid phase was separated by centrifugation with about 1000 g for 10 min. After that, 10 ml of the supernatant solution was filtered from the sample test tube into a 10 ml syringe equipped with a 0.2 μm disposable in-line membrane filter. After filtration, the extract was stabilized with nitric acid by adding about 1% of its volume and stored for further analysis.

The concentrations of the elements in the extract solution were determined by ICP-OES or ICP-MS depending on the concentration of the elements in the solutions.

Statistical analysis

Minitab 15 statistical software was used to determine the mean and standard error. The means of heavy metals in different soil fractions and the physical and chemical soil parameters are presented with the respective standard error values. The General Linear Model (GLM) was employed to compare means against site, depth and their interaction and run F-test. The P values were used to express statistical significance.

RESULTS AND DISCUSSION

Selected physical and chemical properties of Kera and Kolfe soils

The pH of surface and subsurface soils at Kera and Kolfe is moderately acidic ranging from 5.4 (Kolfe subsoil) to 6.5 (Kera surface and subsoil). Kera farm has significantly higher pH than Kolfe farm (P<0.001), which is about one unit higher both at the surface and at the subsurface (Table 1). There is no significant difference between the pH at the surface and subsurface at both sites (P = 0.407).

pH is one of the most important factors that control chemical speciation, precipitation and sorption (Lee and Saunders 2003). Toxic metals are mobile and soluble in aerated acid sites. A study comparing sorption of metals over a wide range of pH during sulfate reduction indicates that metal ions remained in solution as long as the pH was below 3.5, as the pH increased sorption of metals increased (Lee and Saunders, 2003). Under moderately acidic to close neutral pH conditions at Kolfe and Kera, the solubility and mobility of toxic metals appears to be rather restricted.

Kera soil is clay loam to clay with more or less equal proportions of the particle size fractions sand, silt and clay (Table 1). In contrast, Kolfe soil is a clay clearly dominated by the fraction (54.5 - 64.6%) followed by silt (27.5 - 32.1%) and a low sand content (8-13.5%). Kolfe has significantly higher clay than Kera (P<0.001), while

Table 2. Effective cation exchange capacity (ECEC) of contaminated soils (means of four replicates are presented with standard error values)

Site	Dept (cm)	Exchangeable bases (mmol kg⁻¹)					
		Al^{+++}	Ca^{++}	K^+	Mg^{++}	Na^+	Eff. CEC
Kera	0-20	0.0	218 ± 17.4	17.4 ± 0.59	68.8 ± 3.71	6.3 ± 0.87	310.5 ± 22.1
Kera	30-50	0.0	179 ± 22.8	9.03 ± 1.08	63.3 ± 5.31	5.7 ± 0.28	257.3 ± 28.7
Kolfe	0-20	0.18 ± 0.18	186 ± 26.6	7.83 ± 1.04	35.3 ± 1.93	11.4 ± 3.33	240.5 ± 24.5
Kolfe	30-50	0.0	167.5 ± 18.0	6 ± 0.43	36.5 ± 1.5	11.1 ± 3.58	221.3 ± 15.1

Kera had significantly higher sand than Kolfe (P<0.001). There are also significant differences in clay with depth (P = 0.001). Both at Kera and Kolfe, clay increased with depth while sand and silt fractions showed a decreasing but not significant trend. The high clay percentage at Kolfe could contribute to the sorption and retention of heavy metals. The same could be said about Kera where the finer particles range between 69 - 73% of the total. Heavy metal concentrations in soils often have significant correlations to the finer soil fractions (Zhang et al., 2007); or are strongly absorbed by iron and manganese oxides, which are commonly in close association with clay minerals (Kersten and Smedes, 2002).

Apart from the low content (0.5%) in Kera surface soil, carbonates have not been detected in the two sites (Table 1). Kera and Kolfe have fairly high organic carbon (OC) content ranging between 2.3 and 2.8% on the surface and 1.3 to 1.8 at subsurface. Kolfe soils contain significantly higher OC than Kera (P = 0.037), OC significantly decreasing with depth at both sites (P<0.001).

Ca and Mg are the dominant cations on the exchange sites of both soils contributing to the overall effective cation exchange capacity (CEC) (Table 2). Due to near neutral soil pH, Al practically has no contribution to the CEC with the exception of a slight amount at surface of Kolfe soils. In Kera soils the dominance of exchangeable cations on the exchange sites is in the order of Ca > Mg > K > Na with a slight difference in Kolfe soils where the order is Ca > Mg > Na > K. Exchangeable cations decreased with depth at both sites.

Kera has significantly higher ECEC than Kolfe (P=0.041), but depth differences are not significant (P=0.143). In contrast to its higher clay and organic matter content, Kolfe soil has lower CEC than Kera soil. This may be due to differences in clay mineralogy which was not determined in the present study. Non-cracking high clay soils usually have kaolintic clay mineralogy. Soils or clay fractions with high CEC are known to have high adsorptive capacity for metals (Ali 2006). There are therefore possibilities that some proportions of heavy metals at Kera and Kolfe may have been retained by the mineral phase.

When heavy metals are added to soils, some of them are immobilized or form compounds that have low solubility either by adsorption or precipitation, or both. The degree of heavy metal sorption is affected by environmental factors, soil components and properties as well as the amount of heavy metals added (Jalali and Khanlari, 2008).

Total metal concentrations

Both at Kera and Kolfe farms, the concentrations of total arsenic (As) and cadmium (Cd) are within common ranges in the surface and subsurface soils (Table 3). Total mercury (Hg) is found in trace concentrations, and hence there was no need of running sequential extraction.

Cr and Ni surpass maximum tolerable concentrations both at Kera and Kolfe farms (Table 3), from the standpoint of total metal contents, according to central European guidelines for agricultural soils (Merian, 1991). Total Cr is above maximum tolerable levels at the surface and subsurface of Kera and at the surface of Kolfe farms. The concentration at the Kolfe surface soil is about six times as much as the concentration considered maximum tolerable level in agricultural soils. Cr significantly increases with depth at Kera while at Kolfe it decreases substantially (P = 0.001). There is a significant difference in total chromium also between the two sites (P<0.001).

Total Ni is above the maximum tolerable level at both sites and both depths (Table 3). There is no significant difference in total nickel between the two sites (P = 0.151), but there is a significant increase with depth (P = 0.024) at both sites.

Total lead (Pb) is above maximum tolerable level in the surface soil at Kera while it is above common concentrations in the subsurface soil (Table 3). Pb in Kolfe is slightly above common ranges on the surface and within common ranges in the subsurface. Total Pb at Kera is significantly higher than total Pb at Kolfe (P<0.001), and it declines significantly with depth at both sites (P<0.001).

Cu and Zn decline with depth at Kera and Kolfe. Although the decline of Cu with depth is not significant (P = 0.514), there is a significant decline with depth for total Zn (P<0.001). At both sites and both depths they are distributed above common concentration. There is a

Table 3. Total and extracted metal species concentrations (mg kg^{-1}) in some urban farms of Addis Ababa

Site/Depth	Element	Total	Ammon. N. (µg kg^{-1})	acetic a. ext. (avail.)	Reduc.	Oxidiz.	Residual
Kera 0-20	As	6.8±0.28	<25	<0.5	<0.5	<0.5	6.8±0.28
	Cd	0.34±0.03	<2.5	0.10±0.01	0.18±0.02	<0.05	0.07±0.01
	Cr	100.3±11.4	<2.5	<0.5	4.9±0.23	13.1±1.7	82.3±9.6
	Cu	47.0±1.78	57.3±15.2	2.1±0.24	11.8±0.94	4.1±0.25	29.0±0.75
	Hg	0.07±0.009	<0.5	n.a	n.a	n.a	n.a
	Ni	71.8±5.27	56.3±27.0	3.9±0.26	15.8±1.16	2.3±0.1	49.8±4.22
	Pb	101.7±17.2	<2.5	0.3±0.14	90.7±17.1	3.9±0.64	6.8±0.59
	Zn	197.0±11.7	250±148	22.7±2.68	72.4±8.85	3.8±0.68	98.1±0.56
Kera 30-50	As	7.2±0.92	<25	<0.5	<0.5	<0.5	7.2±0.92
	Cd	0.19±0.01	<2.5	0.04±0.01	0.06±0.01	<0.05	0.07±0.01
	Cr	116.3±12.6	<2.5	<0.5	4.3±0.19	11.5±1.1	100.5±11.7
	Cu	41.0±3.1	<2.5	2.1±0.43	11.0±0.8	3.1±0.49	24.9±2.7
	Hg	0.03±0.002	<0.5	n.a	n.a	n.a	n.a
	Ni	83.3±7.3	96.5±17.6	3.0±0.14	14.5±1.9	2.3±0.3	63.4±5.3
	Pb	31.5±1.5	<2.5	<0.1	21.3±2.7	1.6±0.04	9.0±2.1
	Zn	128.8±1.25	9.5±9.5	4.2±0.65	15.4±0.23	1.7±0.3	107.5±1.5
Kolfe 0-20	As	6.0±0.16	<25	<0.5	<0.5	<0.5	6.0±0.16
	Cd	0.13±0.01	3.0±1.1	0.01±0.01	0.02±0.02	0.02±0.02	0.09±0.02
	Cr	596.3±36.4	17.3±2.2	2.1±0.17	47.8±1.5	344.0±17.2	202.4±19.3
	Cu	50.0±0.9	13.5±0.96	1.5±0.08	11.1±1.1	3.7±1.0	33.7±1.3
	Hg	0.04±0.001	<0.5	n.a	n.a	n.a	n.a
	Ni	78.3±1.03	414.8±26.9	2.4±0.15	5.5±0.16	1.7±0.07	68.7±1.1
	Pb	20.5±0.65	<2.5	<0.1	15.4±0.9	0.92±0.05	4.2±0.46
	Zn	148.8±2.98	1937±382	12.7±1.4	27.6±3.6	3.7±1.03	104.8±2.6
Kolfe 30-50	As	4.9±0.12	<25	<0.5	<0.5	<0.5	6.0±0.12
	Cd	0.1±0.003	<2.5	<0.05	<0.05	<0.05	0.09±0.003
	Cr	73.3±5.3	6.3±0.9	0.0±0.0	7.6±0.8	25.0±4.1	95.7±2.5
	Cu	33.0±0.75	13.5±1.3	1.4±0.12	15.5±0.3	4.2±0.8	37.6±0.7
	Hg	0.04±0.003	<0.5	n.a	n.a	n.a	n.a
	Ni	87.8±1.65	667.8±94.0	1.6±0.2	4.03±0.16	1.8±0.19	80.4±1.7
	Pb	16.0±0.41	<2.5	<0.5	9.6±0.3	0.8±0.05	6.6±0.3
	Zn	125.5±0.63	527.3±85.5	4.3±0.3	6.9±0.56	1.9±0.5	117.2±1.2

n.a refers to not analyzed

more or less consistent decline of total metal concentrations with depth in Kolfe soil, except for Ni (Table **3**). This decline with depth can not be observed at the Kera site where Cd, Cu, Hg, Pb and Zn are found in higher concentrations on the surface while As, Cr and Ni increase with depth. This indicates the possibility of leaching of As, Cr, and Ni at Kera while at Kolfe it is only Ni that significantly increases with depth.

Determination of the total concentration levels of metals would not suffice for contaminated soil studies; however, an estimation of their labile fraction is very important (Pueyo, 2008). Total heavy metal contents provide little information on the bioavailability of the heavy metals (Jalali and Khanlari, 2008). The solubility and bioavailability of these metals can be better predicted though through sequential extraction.

Sequential extraction

The acid-extractable fraction contains the metals which are precipitated or co-precipitated with carbonate. The reducible fractions mainly consist of manganese and iron oxyhydroxides, which scavenge metals in the form of coatings on mineral surfaces or as fine discrete particles by one or a combination of the following mechanisms: co-precipitation, adsorption, surface complex formation, ion exchange and penetration of the lattice (Filgueiras et al.,

2002; Martinez-Sanchez et al., 2008). The oxidizable fraction includes trace metals associated with various forms of organic material (Martinez-Sanchez et al. 2008).

Generally, metals in the acid-soluble fractions are considered readily and potentially bioavailable and mobile, while the reducible and oxidizable fractions are relatively stable under normal soil conditions (Martinez-Sanchez et al. 2008). Especially the residual fraction, is considered immobile and tightly bound and may not be expected to be released easily under natural conditions (Jalali and Khanlari 2008).

Comparison by metal species

Kera Farm

All arsenic in surface and subsurface soils at Kera is found in residual fraction (Figures 1 and 2). About 30% of Cd in the surface and 26% in the subsoil at Kera is in the acid extractable fraction, while the rest (>70%) is in reducible and residual fractions. All of Cr is found in forms not readily available for plant uptake, both at the surface and subsurface soils of Kera farm. In both cases, the residual Cr constitutes more than 80% of the total followed by small proportions of oxidizable and reducible fractions, respectively (Figures 1 and 2).

Only about 4.5% (surface) and 5.1% (subsurface) Cu is in the acetic acid extractable fraction while the rest is in the stable and less mobile fractions, in the order of residual > reducible > oxidizable fractions. Similarly, about 5.4% of Ni in the surface soil and 3.6% in the subsoil is in the acetic acid extractable fraction while the rest is in residual > reducible > and oxidizable fractions. Most of the lead (Pb) is in the reducible fraction (89.2% in the surface and 67.6% in the subsurface). Lead is not detectable in the acetic acid extractable fraction. The residual and oxidizable Pb comprises of low concentrations of the total, respectively. Zn is also largely found in the less bioavailable residual, reducible and oxidizable forms, at both surface and subsurface soils in Kera farm (Figures 1 and 2). The available Zn is higher in the surface (11.5%) soil than subsurface (3.3%).

Kolfe Farm

Arsenic in surface and subsurface at Kolfe is also exclusively found in residual forms (Figure 3 and 4). About 61.5% of the surface and 100% of the subsurface Cd is in the residual form. Acetic acid extractable Cd is below 10% of total metal content.

Most of the Cr is also found in the less available oxidizable and residual fractions (Figures 3 and 4). The oxidizable Cr fractions are greater than the residual fractions on the surface while the opposite is true with the subsurface. Ordinarily, with a very high surface and sub-

surface total Cr concentrations one would expect Cr toxicity. However, Cr is bound to mineral and organic fractions that do not easily release it. This appears to be a natural mechanism by which the metal is kept from bioavailability.

Over 70% of Cu is also found in residual and oxidizable fractions thus making it less bioavailable like those metals mentioned above (Figures 3 and 4). About 90% of the nickel is also in the residual form. Similar to Cr, despite high total Ni concentrations, Ni is found mostly in stable fractions and hence is not considered bioavailable. Greater than 70% of the Pb in the surface and more than 50% of the Pb in the subsurface is in the reducible fraction. Bioavailability of Zn is also restricted since about 70% of the Zn on the surface and 90% in the subsurface is in the residual fraction (Figures 3 and 4).

Kera versus Kolfe

Although largely there are similar trends in the distribution of metal species at the two sites, there are some exceptional cases which deserve mentioning. Cd is found in more or less similar proportion in acid soluble, residual and reducible fractions at surface and subsurface, while the residual form is dominant at Kolfe. Above 80% of Cr is in the residual fraction at Kera, while the oxidizable fraction is the highest at the surface of Kolfe, and all in all, the oxidizable fractions both at the surface and subsurface at Kolfe are proportionately higher than their counterparts at Kera. Pb and Zn fractions have similar trends at both sites, but still reducible Pb and Zn are higher at Kera than Kolfe, comparatively.

Comparison by phase

The residual fraction is the dominant form (6.7-100% of total metal percentage) in which most of the metals (As, Cr, Cu, Ni, and Zn) are found in the surface soils of Kera followed by the reducible fraction in which Pb and Cd comprise highest proportions of total metal (89 and 53%, respectively) (Table 3). The acetic acid extractable fractions range between 0.3 - 29.4% and the oxidizable fractions range between 0 - 13% of the total metal contents. The organically bound and the plant available metals hence comprise the lowest proportion of the total metal in surface soils at Kera.

Comparison of the acid extractable soluble metals shows that the potential mobility and bioavailability of metals is in a decreasing order of Cd > Zn > Ni > Cu, at the surface, while at the subsurface the order would be Cd > Cu > Ni > Zn.

The highest acetic acid extractable fraction (close to 30% out of total metal percentage), is that of Cd but the overall Cd concentration is within common ranges of agricultural soils (Figure 1). Apparently what seemed to
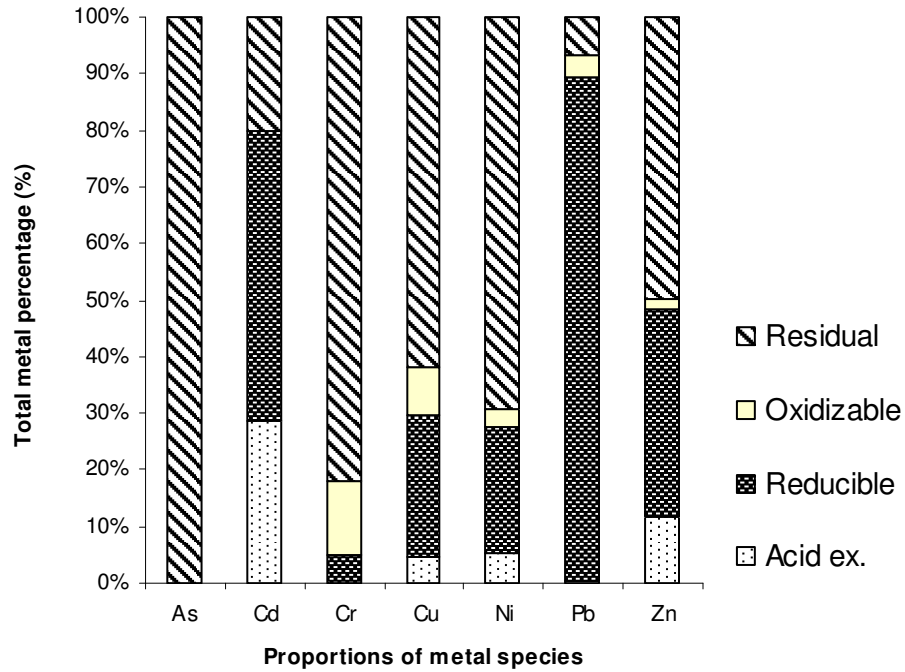
Figure 1. Proportions (mean of 4 replicates) of metals species at surface soils (0 - 20 cm) of Kera farm.
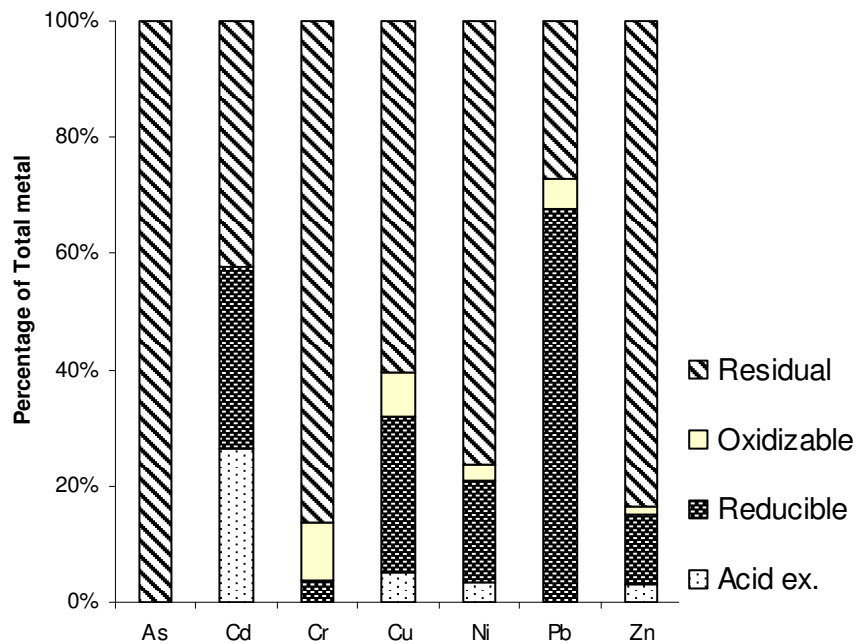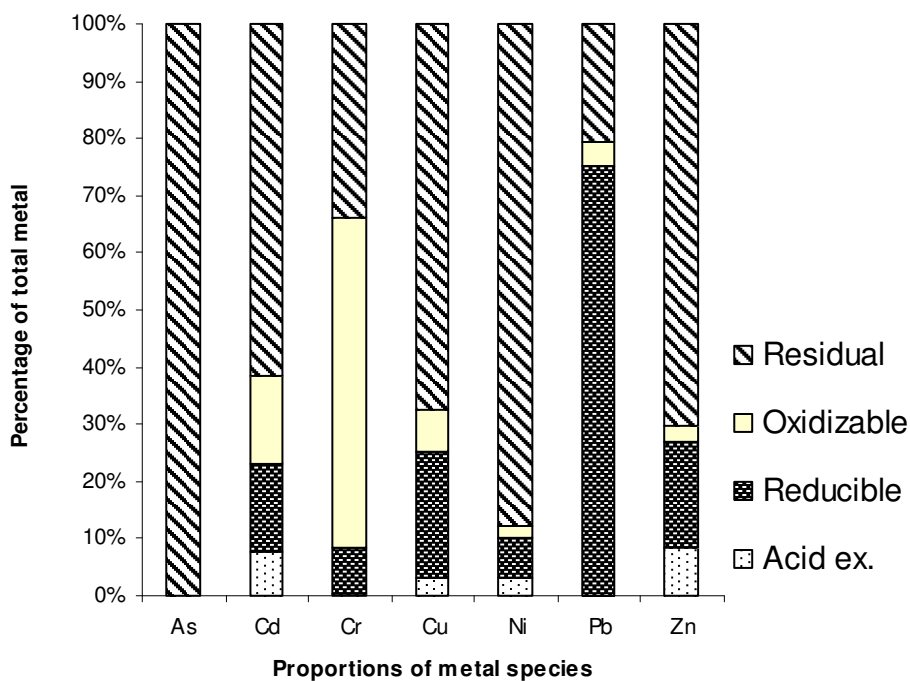

Figure 2. Proportions (mean of 4 replicates) of metals species at surface soils (30 - 50 cm) of Kera farm.

be toxic range for Cr and Ni from the total concentrations, is also found to be largely (> 70%) in the residual and stable form for both elements (Figures 1 and 2). The bioavailability and mobility of the metals studied in Kera surface soil is hence very restricted because of the stable phases in which these metals are found in.

In the surface soil, at Kolfe, similar to Kera, most of the metals (As, Ni, Zn, and Ni) are bound in the residual

Figure 3. Proportions (mean of 4 replicates) of metals species at surface soils (0 - 20 cm) of Kolfe farm.
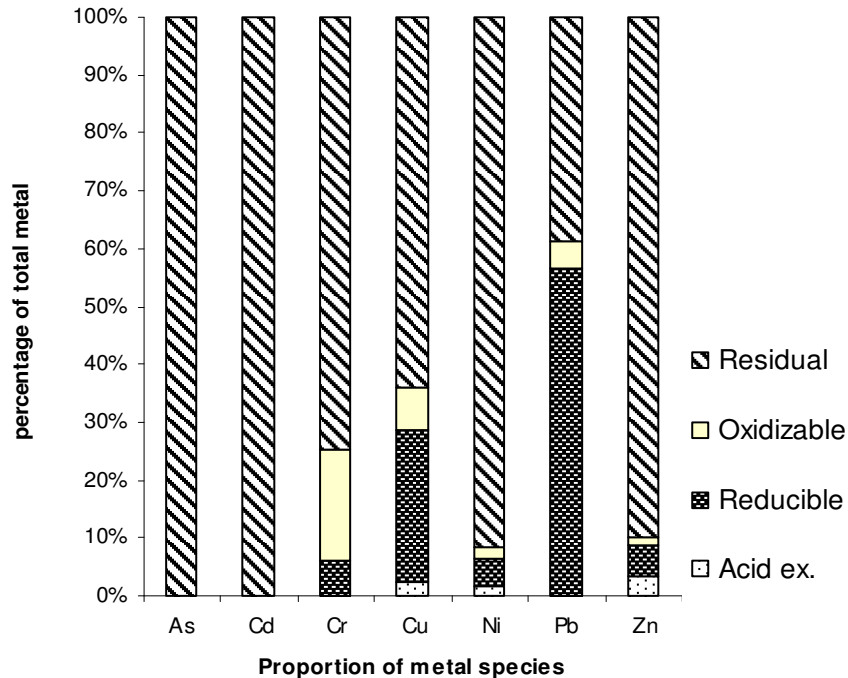

Figure 4. Proportions (mean of 4 replicates) of metals species at surface soils (30 - 50 cm) of Kolfe farm.

fraction (Figure 3). All of arsenic (100%) and greater parts of Ni (88%), Zn (70%), Cu (67.4) and Cd (61.5%) are embedded in the residual fraction. Cr is largely (57.7%) found in the oxidizable form, while most of Pb (75.1%) is in the reducible form. The reducible form follows the residual form in proportion, especially for Cd

(15.4%), Cu (22.2%), and Zn (18.6%). The oxidizable fractions are higher in Cd, Cr, Cu, and Pb than the exchangeable forms, while it is the least in Ni and Zn. Except for Ni and Zn, the exchangeable fraction is the lowest in proportion.

Metals are largely bound to the residual fractions at the subsurface in Kolfe, more than at the surface (Figure 4). Except for Pb, more than 50% of the metal proportion is in the residual form for As (100%), Cd (100%), Ni (91.6%), Zn (89.9), Cr (74.6%), and Cu (64.1%). There appears to be also little leaching of metals to the subsurface. The reducible fraction is next higher fraction for Cu (26.4%), Pb (56.5%), Zn (5.3%) and Ni (4.6%). The exchangeable fraction is the least, except for Zn in which it exceeds the oxidizable fraction. There appears to be limited leaching of acetic acid extractable fraction to the subsurface, and except for the residual fraction, the metals in the other fractions are lower at the subsurface than the surface.

It is however true that metals could change from one form into another depending on some factors. For instance, if the soil pH for some reason declines as a result of acid amendments or leaching of bases occurs due to rainfall, etc., the Cr and Ni in the residual form could be transformed to a readily available form, thereby becoming more bioavailable and mobile in the soil system. Similarly, the high proportion of Mn or Fe oxide bound Pb could also become more soluble and bioavailable under reducing soil conditions.

Metals may be transformed from one fraction to another or exist in these different forms simultaneously (Martinez-Sanchez et al. 2008). Metal fractions associated with organic matter or as sulfides (oxidizable fraction) could become bioavailable in an aerobic environment, due to the potential oxidation of sulfides and degradation of the organic matter (Oygard et al., 2008).

There are some natural mechanisms of metal retention that are important remediation measures to control bioavailability of metals. Natural attenuation processes, such as increase in soil pH or metal diffusion to internal sorption sites in the aluminosilicate phases by migration, adsorption, surface precipitation or redox processes, involve an increase in trace element fixation in the soil with time and a decrease in mobility. These natural attenuation processes, which lead to an eventual reduction of the risk in metal-impacted sites, may be implemented as an alternative remediation strategy to other more active methods (Powell et al., 2004; Pueyo 2008). There is more or less similar trend in the surface soil of Kera Farm with a slight increase of residual fractions and general decline of the acid extractable fractions.

The reducible form may be considered stable as long as the pH is neutral. In an aerobic environment, metals contained within this fraction have a low mobility, and are not considered bio-available. Their status could change however, in the event of changes in the redox conditions.

Reducing conditions would dissolve Fe(III)-oxyhdroxides, due to the reduction of Fe(III) to Fe(II), with the subsequent solution of the metals bound in or onto those Fe(III)-oxyhdroxides (Oygard et al. 2008).

Ammonium nitrate extracted metals

The ammonium extracted trace elements represent the exchangeable and plant available forms. The ammonium nitrate trace elements are found to be generally low both at Kolfe and Kera. However, an exceptional observation made here is that the Ni and Zn contents of Kolfe are significantly higher (P<0.001) than those for Kera, although there is no significant difference between total Zn and Ni at both sites (P = 0.151). There is a significant difference in pH (P<0.001) of Kera and Kolfe farms, whereby the lower pH level at Kolfe may have contributed for relatively higher solubility and availability of these metals.

Ammonium nitrate (AN) extractable metals are very low in both soil horizons at the Kera site (Table 3). This fits well to the low results for the acetic acid extractable fraction. There are limiting values for AN-extractable cadmium (0.04 mg/kg) and lead (0.1 mg/kg) on arable land in the German soil protection regulation. AN-extractable contents were below quantification limit (0.0025 mg/kg) for both elements in the Kera soil and therefore far below those limiting values. In spite of the relatively high total lead concentration in the surface soil the mobility of this metal in the soil seems to be limited by strong bonding to the reducible soil fraction.

As for the Kolfe soil the AN-extractable metal contents are very low and in the case of Cd and Pb far below the German limiting values for arable soils (Table 3).

Conclusions and Recommendations

Total Cr, Ni (Kera and Kolfe) and Pb at Kera, surpass normal concentrations in agricultural soils, although total concentrations can not be used to predict bioavailability. Total Cr at surface of Kolfe is particularly very high, about six times as much as the maximum tolerable concentration. Sequential extraction has, however, revealed that all of Cr, and Pb and the greater part of Ni are found in forms not readily bioavailable.

Most metals, with the exception of lead, are largely found in the residual fraction. The acid soluble forms are considered the readily bioavailable forms, and these happen to be the least at both sites. Cd in the acetic acid extractable fraction is proportionately the highest bioavailable form detected, where it forms 25-30% of the total metal at Kera surface and subsurface, followed by acetic acid extractable Zn, constituting about 11.5% of the total. Acetic acid extractable Zn in Kolfe surface soil is also about 8.5% of total Zn. Although proportion of

soluble Cd to total metal concentration, is relatively high in Kera soils, the total Cd in the soil is within common ranges. All the rest of the acetic acid extractable metals are below 5.5% of the total.

From the above, it is evident that the bioavailability of all of the metals studied at the moment is very low and hence little or no great risk could be anticipated on soil and crop quality under current circumstances. This may be due to moderate to neutral acidic soil pH, high fine fractions at both sites, and high organic matter content of the soils. Natural attenuation processes, such as increase in soil pH or metal diffusion to internal sorption sites in the aluminosilicate phases could result in metal fixation in soils and reduced mobility. Such natural attenuation processes could be adopted as alternative metal remediation strategies.

However, it is known that whenever changes in pH, oxidation reduction reactions, organic substances etc. occur within the soil system, there is a possibility of shift of metals from non-bioavailable forms to bioavailable forms. For instance, a drop in soil pH or similar causes could increase the relatively very high levels of total Cr, Ni and Pb in these soils into bioavailable forms, which could then have tremendous negative impact on soil and crop quality. Therefore, it is very important to monitor the soil conditions regularly in order to avoid the solubility of such metals and their entrance into food chain.

Total Ni and Cr (significantly) and As (not significantly) increase with depth at Kera, while at Kolfe it is only total Ni that increases significantly with depth. Leaching could be the most likely cause for this. Since Kera has coarser texture on surface compared to the clay rich surface of Kolfe, leaching looks to be a likely cause for most metal movement downwards. However, since most of these metals were found in stable forms, it is appropriate to make thorough studies in the future to definitely conclude whether leaching was the only cause.

The soil at Kera has coarser texture, higher CEC, and higher pH than Kolfe soil, parameters that can contribute to metal leaching, ion retention and mobility. Although both soils have similar trends in metal proportions to a larger extent, some anomalous conditions include such as Cd prevailing more in the acid soluble fractions at Kera while the residual form is dominant at Kolfe. Above 80% of Cr at Kera is in the residual form, while especially at surface of Kera, oxidizable Cr constitutes the highest proportion.

In many developing countries like Ethiopia, there is a general lack of awareness of the risk associated with excess build up of toxic metals. There are no clear policies put to enforce environmental laws related to waste management and the like. Studies of this nature are almost absent. Therefore, publicizing such research results and supporting policy measures in developing countries are important to mitigate environmental risks such as the ones that are brought from heavy metal contamination.

REFERENCES

Ali G (2006). Adsorption characteristics of heavy metals in soil zones developed on spilite. Environ. Geol. 51: 333-340.

Doelsch E, Moussard G, Macary H S (2008). Fractionation of tropical soilborne heavy metals-Comparison of two sequential extraction procedures. Geod. 143: 168-179.

EN ISO 11885 (1997). Determination of 33 elements by inductively coupled plasma atomic emission spectroscopy. International Organization for Standardization, Geneva, Switzerland.

Filgueiras AV, Lavilla I, Bendicho C (2002). Chemical sequential extraction for Metal partitioning in environmental solid samples. J Environ. Mon 4: 823-857.

Fitamo D, Itanna F, Olsson M (2007). Total contents and sequential extraction of heavy metals in soils irrigated with wastewater, Akaki, Ethiopia. 2007. Environ. Man 39: 178-193.

ISO 10694 (1995). Soil quality: Determination of organic and total carbon after dry combustion (elementary analysis). International Organization for Standardization, Geneva, Switzerland.

ISO 11277 (1995). Soil quality: Determination of particles size distribution in mineral soil material; Method by sieving and sedimentation following removal of soluble salts, organic matter and carbonates. International Organization for Standardization, Geneva, Switzerland.

ISO 17294-2 (2003). Water quality – Application of inductively coupled plasma mass spectrometry (ICP-MS) – Part 2: Determination of 62 elements. International Organization for Standardization, Geneva, Switzerland.

ISO 19730 (2006). Soil quality – extraction of trace elements from soil using ammonium nitrate solution. International Organization for Standardization, Geneva Switzerland.

Itanna F (1998a). Metal concentrations of some vegetables irrigated with industrial Liquid waste at Akaki, Ethiopia. Eth. J. Sci. (SINET) 21: 133-144.

Itanna F (1998b). Comparative study of soil pollution with toxic substances on farmlands close to old and new industrial sites in Ethiopia. Bull of the Chem. Soc. Eth. 12: 105-112.

Itanna F (2002). Metals in leafy vegetables grown in Addis Ababa and toxicological implications. Eth. J Health Dev. 16, 295-302.

Jalali M, Khanlari ZV (2008). Effect of aging process on the fractionation of heavy metals in some calcareous soils of Iran. Geod. 143: 26-40.

Karlen DL, Andrews SS, Weinhold BJ, Doran JW (2003). Soil quality: Humankind's foundation for survival. Research editorial. J Soil Water Cons. 58: 171-178.

Kersten M, Smedes F (2002). Normalization procedures for sediments contaminants in spatial and temporal trend monitoring. J Environ. Monit. 4: 109–115.

Lee MK, Saunders JA (2003). Effect of pH on metal precipitation and sorption: Field bioremediation and geochemical modeling approaches. Vadose Z J 2: 177-185.

Martinez-Sanchez MJ, Navarro, MC, Perez-Sirvent C, Marimon J, Vidal J, Garcia-Lorenzo ML, Bech J (2008). Assessment of the mobility of metals in a mining-impacted coastal area (Spain, Western Mediterranean). J Geochem. Explor. 96: 171-182.

Merian E (Ed) (1991). Metals and Their Compounds in the Environment:

Occurrence, Analysis and Biological Relevance. VCH, Weinheim, pp. 687-711.

Niskavaara H (1995). A comprehensive scheme of analysis for soils, sediments, humus and plant samples using inductively coupled plasma atomic emission spectrometry. Geological Survey of Finland, Current Research 1993–1994, Geological Survey of Finland Special Paper 20, pp. 167–176.

Oygard JK, Gjengedal E, Mobbs HJ (2008). Trace element exposure in the environment from MSW landfill leachate sediments measured by a sequential extraction technique. J Hazard Mat. 153: 751-758.

Powell KR, Serkiz SM, Pishko AL, Barr CS, Brewer K, Lee CM, Schank A (2004). From sequential extraction to transport modeling: monitored natural attenuation as remediation approach for inorganic contaminants. Preprints of extended abstracts presented at the ACS National Meeting, American Chemical Society, Div. of Environ. Chem. 44: 654-657.

Pueyo M, Mateu J, Rigol A, Vidal M, Lopez-Sanchez JF, Rauret G (2008). Use of the modified BCR three-step sequential extraction procedure for the study of trace element dynamics in contaminated soils. Environ Poll 152, 330-341.

Silveira ML, Alleoni LRF, O'Connor GA, Chang AC (2006). Heavy metal sequential extraction methods – a modification for tropical soils. Chemo 64: 1929-1938.

Skinner SIM, Halstead RL, Brydon JE (1959). Quantitative manometric determination of calcite and dolomite in soils and limestones. Canad. J. Soil Sci. 39: 197-204.

Zhang W, Yu L, Lu M, Hutchinson SM, Feng H (2007). Magnetic approach to normalizing heavy metal concentrations for particle size effects in intertidal sediments in the Yangtze Estuary, China. Environ. Poll. 147: 238-244.