Environ Chem Lett (2011) 9:197–202
DOI 10.1007/s10311-009-0263-5

ORIGINAL PAPER

Human bioaccessibility of Cr, Cu, Ni, Pb and Zn in urban soils from the city of Torino, Italy

Julien Sialelli · Christine M. Davidson · Andrew S. Hursthouse · Franco Ajmone-Marsan

Received: 30 June 2009 / Accepted: 13 November 2009 / Published online: 12 December 2009
© Springer-Verlag 2009

Abstract Several physiologically based extraction procedures have been proposed to estimate the fraction of the potentially toxic element content that would be bioaccessible in the human gastro-intestinal tract following accidental ingestion of soil. Many of these procedures are complex, they have been applied to a very limited range of soils, and most work has focussed on arsenic and lead. In the present study, a simplified, two-stage extraction, simulating the human stomach and intestine, was developed and applied to urban soil samples from ten public-access areas in the City of Torino, Italy. The human oral bioaccessibility of chromium, copper, nickel, lead and zinc was estimated. Lead and zinc bioaccessibilities were found to be higher in the stomach, but chromium was more bioaccessible in the intestine. Analyte concentrations were higher in roadside soils than in soils from parks. A higher proportion of the soil metal content was found in bioaccessible forms at roadsides than in parks. Comparison of the current findings with results of earlier work involving sequential extraction of the same soils indicated that the sequential procedure gave a relative, but not an absolute, indication of bioaccessibility. Calculations based on the bioaccessible analyte concentrations suggest that ingestion of only 2–3 g of some of the roadside soil samples from Torino could deliver the tolerable daily oral intake of chromium, nickel and lead to a 20-kg child. The developed procedure is useful for preliminary screening of soils and prediction of whether their bioaccessible metal contents are likely to pose a risk to human health.

Keywords Urban soil · Potentially toxic element · Bioaccessibility

J. Sialelli · C. M. Davidson (✉)
WestCHEM, Department of Pure and Applied Chemistry, University of Strathclyde, 295 Cathedral Street, Glasgow G1 1XL, UK
e-mail: c.m.davidson@strath.ac.uk

A. S. Hursthouse
School of Engineering and Science, University of the West of Scotland, High Street, Paisley PA1 2BE, UK

F. Ajmone-Marsan
DI.VA.P.R.A., Chimica Agraria, Università di Torino, Via Leonardo da Vinci, 44, 10095 Grugliasco, Torino, Italy

Introduction

The urban geochemistry of trace elements is of growing interest and likely to increase in importance since more than half of the world's population now live in urban environments (Wong et al. 2006). Urban soils may be highly variable in composition (Madrid et al. 2006), both spatially and temporally, because they are affected by numerous point and diffuse sources of contaminants, which change as a town or city develops. They may contain multiple contaminants, enriched above local background levels, especially if the city has been heavily industrialised for a prolonged period during its history.

The city of Torino (Turin) lies on an alluvial plane in the Piemonte region of north-western Italy. The city is long-established (there is reputed to have been a settlement in the area since Roman times), large (almost 1 million inhabitants) and has a lengthy association with metallurgical and automobile manufacturing industries (Biasioli et al. 2006). Because of the city's industrial heritage, urban soils in the Torino area are of interest in terms of their potentially toxic element (PTE) content (Biasioli et al. 2006; Hursthouse et al. 2004; Poggio et al. 2009).

Total or pseudototal (i.e. aqua-regia soluble) metal contents alone are insufficient to assess whether a particular contaminated soil poses a significant risk to health. Instead, it is necessary to consider the fraction of the PTE content that may become bioavailable, to a particular organism, by a particular exposure route (for example, uptake via roots in plants, or oral ingestion in humans). This fraction is often estimated by the application of a selective, single or sequential extraction (Bacon and Davidson 2008).

Several groups of researchers have developed extraction procedures that seek to estimate the fraction of the total PTE content that would be bioaccessible, in the human gastrointestinal tract, following (usually) accidental ingestion of contaminated soil (Intawongse and Dean 2006; Oomen et al. 2002). These procedures vary markedly in complexity, from specialist apparatus that simulate human digestion with a high degree of accuracy, to simple, shake-flask methods which, although less accurate, can be performed relatively quickly, thus allowing soils from contaminated sites rapidly to be screened for their bioaccessible PTE contents (Ruby et al. 1999). One of the most popular approaches is the physiologically based extraction test developed by Ruby (Ruby et al. 1996). The procedure involves two stages, a gastric (stomach) and an intestinal phase. It is relatively simple to perform, and results have shown correlations with animal models for arsenic and lead. There remains, however, a strong need for further work to expand the range of analytes, and types of soils, to which bioaccessibility tests have been applied (Juhasz et al. 2009).

This paper describes a preliminary study in which a simplified, two-stage extraction was use to estimate the oral bioaccessibility of chromium, copper, nickel, lead and zinc in ten urban soils, five from roadsides and five from parks or open spaces, in the city of Torino. Analyte concentrations in the soil extracts were determined by inductively coupled plasma atomic emission spectrometry (ICP-AES).

Experimental

Samples

The samples studied were surface soils (0–10 cm depth) collected as part of the EU URBSOIL project (contract EVK-CT-2001-00053) from public-access areas in major European cities. Each sample was obtained by combining five individual specimens collected either at the centre and vertices of a 2-m diameter square (parks and open spaces, PO) or at 1-m intervals along a transect parallel to the road (roadside samples, RD). Samples were returned to the laboratory, air-dried and passed through a 2 mm sieve before being circulated between URBSOIL partner laboratories.

Apparatus

Extractions were performed on triplicate test portions of each soil using a bench-top orbital shaker in a walk-in incubator maintained at a temperature of 37 ± 1°C. Analytes were determined in soil extracts using an Optima 3,000 ICP-AES system (Perkin Elmer, Bucks, UK) and reagent-matched standard solutions. Glass and plastic-ware were soaked in 5% HNO_3 overnight and rinsed with distilled water before use.

Reagents

Tri-sodium citrate (AnalaR), DL malic acid (GPR), sodium hydrogen carbonate (GPR), pancreatin, pepsin and the 1,000 mg L^{-1} Spectrosol stock solutions used to prepare ICP-AES calibrants were from Merck (Poole, UK). DL lactic acid syrup was from VWR (Lutterworth, UK).

Solution A (stomach phase)

2.5 g pepsin, 1 g tri-sodium citrate, 1 g DL malic acid and 840 µl lactic acid syrup were mixed, the mixture was diluted with distilled water, acidified to pH 1.5 with HCl and made up to 2 L in a standard flask.

Solution B (intestinal phase)

Prepared from the stomach phase solution by the addition of 500 mg pancreatin per L of solution and neutralisation to pH 7 by the addition of solid sodium hydrogen carbonate (typically about 13 g per L).

Physiologically based extraction procedure

Phase 1 (stomach phase)

0.50 g soil was weighed into a screw-top polypropylene bottle, and 50 mL of solution A was added. The mixture was shaken for 1 h at 150 rpm and 37°C, allowed to settle under gravity, and a 10-mL aliquot of the supernatant was then removed for analysis by means of a disposable plastic syringe.

Phase 2 (first intestinal phase)

10 mL of fresh solution A was added to restore the original soil:solution ratio. The mixture was then neutralised and pancreatin was added, to bring the composition to that of intestinal solution. The bottle was shaken for 2 h (150 rpm, 37°C) and then a 10-mL aliquot was removed for analysis as described above.

Phase 3 (second intestinal phase)

10 mL of fresh solution B was added to maintain a constant soil:extractant ratio. The bottle was shaken for a further 1.5 h before a final 10-mL aliquot was taken for analysis as described above. All extracts were stored at 5°C prior to analysis and were analysed within 2 weeks of extraction.

Calculations

Results were corrected for the dilution caused by the removal of extract at the end of phases 1 and 2 (10 mL from a total volume of 50 mL) and its replacement with fresh solution i.e.

$$\text{Corrected concentration}_{(\text{phase 2})} = \text{Measured concentration}_{(\text{phase 2})}/0.8 \quad (1)$$

$$\text{Corrected concentration}_{(\text{phase 3})} = \text{Measured concentration}_{(\text{phase 3})}/0.64 \quad (2)$$

Percentage bioaccessibility was calculated as follows:

$$\text{Bioaccessibility}(\%) = (\text{bioaccessible concentration}/\text{pseudototal concentration}) \times 100 \quad (3)$$

The soil intake required to reach the tolerable daily intake for a 20-kg child was calculated from:

$$\text{Mass of soil required} = \text{tolerable daily intake}/\text{bioaccessible PTE concentration} \quad (4)$$

Quality control

No certified reference material is available for these analytes extractable by physiologically based extraction procedures. The accuracy of the extraction cannot therefore be proved. The reproducibility of the procedure was monitored by including specimens of the same soil, obtained from an urban park in Glasgow, in each batch of extractions. Results for this reference soil were always within two standard deviations of mean values, for all three phases.

Results and discussion

The pseudototal metal concentrations in the urban soil samples are summarised in Table 1 (after Davidson et al. 2006). Levels of contamination are broadly similar to some of those reported in soils from other locations in Torino by previous workers (also shown in Table 1). In a study of eight parks and formal gardens, Hursthouse et al. (2004) found mean concentrations of PTE higher than the present results for park soils, but lower than the present results for roadsides. Overall mean pseudototal analyte concentrations in the current study were greater than those reported by Biasioli et al. (2006) in their comprehensive study of 70 park and roadside soil in Torino. However, samples were from different sites which, together with the intrinsic heterogeneity of urban soils, mean that the levels of (dis)agreement found are unremarkable.

Table 1 Average pseudototal analyte concentrations in urban soils from Torino (mg kg^{-1} dry weight)

	Cr	Cu	Ni	Pb	Zn
PO.118	89.0	36	93.0	48.0	90.0
PO.124	109	59	118	48.0	105
PO.128	102	53	115	58.0	100
PO.140	121	65	142	82.0	138
PO.156	159	73	167	47.0	97.0
Mean (parks)	116	57	127	57	106
RD.112	392	138	312	475	156
RD.132	280	368	239	952	416
RD.151	308	281	155	255	398
RD.161	511	80	492	214	173
RD.167	829	145	878	372	233
Mean (road sides)	464	202	415	454	275
Mean (all sites)	290	130	271	255	191
Hursthouse et al. (2004)	229	111	193	158	242
Biasioli et al. (2006)	191	90	209	149	183

Concentrations of PTE were from 2.6 (for zinc) to 8.0 (for lead) times higher in roadside soils than in those obtained from parks and open spaces suggesting that, as expected, road traffic is a significant source of these contaminants to nearby soil. Similarities exist in the distributions of copper, lead and zinc across the ten sites, with maximum concentrations of all three analytes at site RD.132. Chromium and nickel showed a different distribution from copper, lead and zinc, but similar to each other, with highest concentrations at RD.167.

Concentrations of bioaccessible analytes in the three phases of the physiological extraction are shown in Fig. 1. On average, for samples in which chromium was detectable in all phases, the levels doubled between the stomach phase (phase 1) and the first intestinal phase (phase 2) probably due to the formation of soluble oxo-species stable at higher pH. A further, smaller increase was observed between phase 2 and phase 3, suggesting that equilibrium had not been reached after only 2 h of extraction under intestinal conditions. Concentrations of copper and, to a less extent, nickel also increased from phase 1 to phase 3 for most samples. In contrast, lead and zinc concentrations decreased markedly—sometimes to <50% of gastric solution concentrations—when intestinal conditions were established.

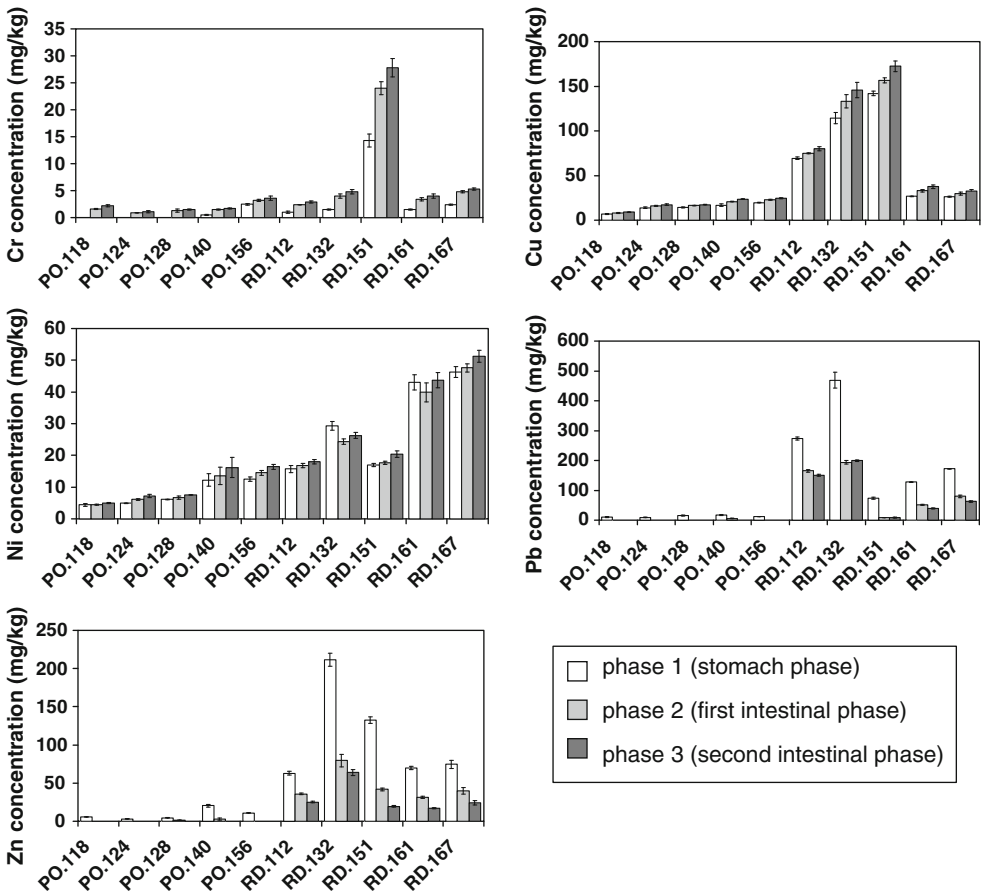

Fig. 1 Bioaccessible analyte concentrations (mg kg^{-1}dry weight). *Error bars* represent one standard deviation ($n = 3$); where no bar is present this indices a result < LOD

This effect, which has been observed previously for lead (Ruby et al. 1996) is likely to be due to pH dependent precipitation and absorption. In general, the park soils contained lower concentrations of bioaccessible analytes than the roadside soils.

Only the phase yielding the highest estimate of bioaccessible concentration for each analyte (i.e. phase 1 for lead and zinc, but phase 3 for chromium, copper and nickel) is considered in the following discussion, since this gives the most conservative ('worst-case') estimate of risk.

The fraction of the pseudototal content found in bioaccessible forms (calculated using Eq. 3 in the experimental section) differed between elements. The mean (intestinal) percentage bioaccessibilities for chromium, copper and nickel were 2.2, 23, and 8.5%, respectively, whilst the mean (stomach) value were 36% for lead and 24% for zinc. The lower proportion of chromium and nickel in bioaccessible forms, relative to copper, lead or zinc, is probably due to a high geogenic contribution of refractory chromium- and nickel-containing minerals from serpentinites in the area (Biasioli et al. 2006). The finding is also in agreement with previous work (Madrid et al. 2008) that applied a single step bioaccessibility extraction test (involving a 1-h extraction in 0.4 mol L^{-1} glycine at pH 1.5) to different particle size fractions of urban soils from Sevilla and Torino. The proportions of analytes in bioaccessible forms were greater, and appeared to be more variable, in the roadsides soils than in soils from parks (Table 2).

A good correlation was observed between pseudototal concentrations and bioaccessible concentrations for most analytes. The R^2 values were 0.87 for copper and 0.85 for nickel (phase 3 vs. pseudototal) and 0.98 for lead and 0.91 for zinc (phase 1 vs. pseudototal). The exception was

Table 2 Percentage bioaccessibilities of analytes in urban soils from different locations

Soil origin	Cr[a]	Cu[a]	Ni[a]	Pb[b]	Zn[b]
Parks	1.7 ± 0.6	31 ± 4	7.8 ± 2.6	23 ± 3	8.0 ± 4.9
Roadsides	2.6 ± 3.6	46 ± 16	8.9 ± 3.2	48 ± 12	39 ± 8

Values presented as mean ± one standard deviation ($n = 5$) calculated from Eq. 3 in the experimental section

[a] Second intestinal phase

[b] Stomach phase

chromium where the most significant correlation, with an R^2 value of 0.61, was found between pseudototal levels and phase 3 concentrations only when the result for site RD.151, which is considerably richer in chromium than other sites, was excluded.

Previous studies on these soil samples used the revised sequential extraction scheme developed under the auspices of the Commission of the European Communities, Community Bureau of Reference to fractionate the PTE content (Davidson et al. 2006). The highest bioaccessible concentrations of analytes in the current work were all found in the soil samples indicated by the previous study to contain the highest proportions of species extractable in the first three steps of the sequential extraction. This reinforces the importance of chemical speciation in influencing bioaccessibility and suggests that the sequential extraction may give an indication of relative levels of bioaccessible PTE in soils. However, for the soils studied, the sequential extraction released larger amounts of analytes than the gastro-intestinal simulation procedure: the sum of the first three steps (exchangeable + reducible + oxidisable) was approximately twice the result of the physiologically based extraction for copper, lead and zinc, but up to eight times the result of the physiologically based extraction for chromium and nickel. Hence, as emphasised in a recent review (Bacon and Davidson 2008), the sequential extraction procedure does not reliably estimate the absolute bioaccessibility but only the relative risk from different metals and soils.

A key aim of bioaccessibility testing is to obtain an estimate of the likelihood of adverse health effects, especially to young children who may come into contact with contaminated soil through play, are prone to hand-to-mouth transfer and may absorb PTE such as lead more efficiently than adults (Ruby et al. 1999). A crude estimation of the relative risk posed by the soils studied can be obtained by calculating the amount of a particular soil that would require to be ingested to reach a toxicologically significant level e.g. the tolerable daily intake, for a hypothetical child. Estimates of toxicological thresholds vary but reports by the UK Department of Environment, Food and Rural Affairs suggest that reasonable values for tolerable daily intake of chromium, nickel and zinc are 3, 5 and 4 μg kg^{-1} body weight per day, respectively (DEFRA 2002), corresponding to 60, 100 and 80 μg day^{-1} for a 20-kg (approximately 6-year-old) child. Tolerable daily intakes of 3 mg day^{-1} Cu and 12 mg day^{-1} Zn have also been reported for children aged 4–8 years (Selinus 2005).

Results of calculations based on these values (see Eq. 4 in the experimental section) are shown in Table 3. These suggest that copper and zinc are not a significant risk at these sites, since more than 17 g of soil would require to be ingested, per day, to reach the tolerable daily intake values for these elements for a 20-kg child. Chromium and nickel contamination are of slightly greater concern since ingestion of ~2 g of soil per day at specific sites (RD.151 for Cr, and RD.161 and RD.167 for Ni) would deliver the tolerable daily intake. The high levels of lead in the roadside soils, together with high bioaccessibility, mean that ingestion of <1 g per day of these soils would provide the tolerable daily intake. Indeed, only 200–300 mg would be required at sites RD.112. and RD.132.

Table 3 Average mass of soil required to be ingested to reach the tolerable daily intake for a 20-kg child (g)

	Cr	Cu	Ni	Pb	Zn
PO.118	27.3	337	20.0	7.2	2,069
PO.124	54.5	174	13.9	7.7	3,750
PO.128	40.0	173	13.3	4.8	2,609
PO.140	35.3	127	6.2	4.1	591
PO.156	16.7	122	6.1	5.8	1,091
RD.112	20.7	38	5.6	0.3	191
RD.132	12.5	21	3.8	0.2	57
RD.151	2.2	17	4.9	1.0	91
RD.161	15.0	80	2.3	0.6	173
RD.167	11.3	93	1.9	0.4	161

Conclusion

This preliminary study has demonstrated the potential usefulness of a simple, two stage physiologically based extraction to estimate human health risks from oral ingestion of urban soils contaminated with PTE. Chromium bioaccessibility was found to be greater in the intestinal phase, whereas lead and zinc were extracted more efficiently under simulated stomach conditions. The bioaccessible PTE concentrations at roadside sites were typically higher, and represented a larger proportion of pseudototal concentrations, than soils from parks and open spaces. Relationships were found between PTE extractable by the physiologically based extraction and pseudototal concentrations and between results of the physiologically based extraction and PTE extractable by the revised sequential extraction procedure developed under the auspices of the Commission of the European Communities, Community Bureau of Reference. A simplistic risk assessment suggested that ingestion of only a few grams of selected roadside soils from Torino could deliver the tolerable daily intake for chromium, lead and nickel, for a 20-kg child. However, this is almost certainly an overestimate of true risk from soil ingestion since published tolerable daily intake values are often conservative (for example, DEFRA chromium values are derived from USEPA data for the

more toxic Cr^{VI} rather than for total chromium) and it is highly doubtful whether the same individual would be in a position to ingest contaminated soil, from within 1 m of a busy road, on a regular daily basis.

References

Bacon JR, Davidson CM (2008) Is there a future for sequential extraction? Analyst 133:25–46

Biasioli M, Barberis R, Ajmone-Marsan F (2006) The influence of a large city on some soil properties and metals content. Sci Total Environ 356:154–164

Davidson CM, Urquhart GJ, Ajmone-Marsan F, Biasioli M, Duarte AC, Diaz-Barrientos E, Grcman H, Hossack I, Hursthouse AS, Madrid L, Rodriguez S, Zupan M (2006) Fractionation of potentially toxic elements in urban soils from five European cities by means of a harmonised sequential extraction. Anal Chim Acta 565:63–72

DEFRA (Department for the Environment, Food and Rural Affairs) and the Environment Agency (2002) Contaminants in soils: collation of toxicological data and intake values for humans. R&D Publications TOX 4 Chromium; TOX 6 Lead; and TOX 8 Nickel

Hursthouse A, Tognarelli D, Tucker P, Ajmone-Marsan F, Martini C, Madrid L, Madrid F, Diaz-Barrientos E (2004) Metal content of surface soils in parks and allotments from three European cities: initial pilot study results. Land Contam Reclam 12:189–196

Intawongse M, Dean JR (2006) In vitro testing for assessing the oral bioaccessibility of trace elements in soils and food samples. Trends Analyt Chem 25:876–886

Juhasz AL, Weber J, Smith E, Naidu R, Marschner B, Rees M, Rofe A, Kuchel T, Sansom L (2009) Evaluation of SBRC-Gastric and SBRC-Intestinal methods for the prediction of in vivo relative lead bioavailability in contaminated soils. Environ Sci Technol 43:4503–4509

Madrid L, Diaz-Barrientos E, Ruiz-Cortés E, Reinoso R, Biasioli M, Davidson CM, Duarte AC, Grčman H, Hossack I, Hursthouse AS, Kralj T, Ljung K, Otabbong E, Rodrigues S, Urquhart GJ, Ajmone-Marsan F (2006) Variability in concentrations of potentially toxic elements in urban parks from six European cities. J Environ Monit 8:1158–1165

Madrid F, Biasioli M, Ajmone-Marsan F (2008) Availability and bioaccessibility of metals in fine particles of some urban soils. Arch Environ Contam Toxicol 55:21–32

Oomen AG, Hack A, Minekus M, Zeijdner E, Cornelis C, Schoeters G, Verstraete W, Van de Wiele T, Wragg J, Rompelberg CJM, Sips AJAM, Van Wijnen JH (2002) Comparison of five in vitro digestion models to study the bioaccessibility of soil contaminants. Environ Sci Technol 36:3326–3334

Poggio L, Vrscaj B, Schulin R, Hepperle E, Ajmone-Marsan F (2009) Metal pollution and human bioaccessibility of topsoils in Grugliasco (Italy). Environ Pollut 157:680–689

Ruby MV, Davis A, Schoof R, Eberle S, Sellstone CM (1996) Estimation of lead and arsenic bioavailability using a physiologically based extraction test. Environ Sci Technol 30:422–430

Ruby MV, Schoof R, Brattin W, Goldade M, Post G, Harnois M, Mosby DE, Casteel SW, Berti W, Carpenter M, Edwards D, Cragin D, Chappell W (1999) Advances in evaluating the oral bioaccessibility if inorganics in soil for use in human health risk assessment. Environ Sci Technol 33:3697–3705

Selinus O (ed) (2005) Essentials of Medical Geology. Elsevier, Amsterdam

Wong CSC, Li X, Thornton I (2006) Urban environmental geochemistry of trace metals. Environ Pollut 142:1–16