Bull Environ Contam Toxicol (2010) 84:197–201
DOI 10.1007/s00128-009-9916-0

Lead and Cadmium Accumulation in Medicinal Plants Collected from Environmentally Different Sites

Henok Baye · Ariaya Hymete

Received: 10 May 2009 / Accepted: 18 November 2009 / Published online: 4 December 2009

Abstract Though use of herbal medicine is increasing dramatically worldwide, environmental pollution especially with heavy metals poses serious problem on quality of medicinal plants and their products. In Ethiopia, where more than 85% of the population relies on traditional medicine, data on heavy metals level of plants is unavailable. The purpose of this study was to assess Pb and Cd in plants grown in different parts of Ethiopia. Out of 26 samples analyzed, four for lead (15.4%; 11.56 ± 1.07 to 98.19 ± 1.05 mg/kg) and seventeen for cadmium (65.4%; 0.38 to 1.83 ± 0.06 mg/kg) were found to contain concentrations above WHO limits (10 mg/kg and 0.3 mg/kg respectively).

Keywords Medicinal plants · Heavy metal contamination · AAS · Ethiopian medicinal plants · Pb and Cd accumulation · Herbal medicine

The use of herbal medicines is increasing alarmingly in both developing and developed countries due to their reasonable price and above all due to the assumption that natural products are safe (Rai et al. 2005). In some parts of the world, they are the only options for primary health care to poor peoples like in Ethiopia where more than 85% of the population rely on herbs for primary health care (BBC News 2006). To get the desired therapeutic outcome, the quality of the finished products and plant raw materials must be ensured. Many reports have shown that one of the major quality problems frequently encountered is high heavy metals content of herbal medicines that can be associated to extensive pollution of the environment where medicinal plants used as raw materials grow (Gasser et al. 2009; Barthwal et al. 2008; Naithani and Kakkar 2006; Wong et al. 1993). Pollution of the atmosphere and soil with Pb and Cd from polluted irrigation water, automobile and industrial exhausts, pesticides and fertilizers play important roles in contamination of medicinal plants (Caldas and Machado 2004; Jarup 2003; Zaccaroni et al. 2003). Higher levels of Pb and Cd cause variety of acute and chronic health problems including cancer, brain and kidney damage, heart problems, liver dysfunction, damage to reproductive system, memory impairment, even death and can cross the placental barrier, with potential toxic effects on the fetus (Agoramoorthy et al. 2008; Kadir et al. 2008; Kwon et al. 2003).

Therefore, it is mandatory to assess Pb and Cd concentration in medicinal plants before using them for herbal drugs preparation. Though similar studies have been conducted in different parts of the world (Street et al. 2008; Ang et al. 2003; Rai et al. 2001; Chuang et al. 2000), there is no report on flora originated from Ethiopia. With this notion the work was done to measure the two most common and toxic metals, Pb and Cd, in plants commonly used for preparation of herbal medicines and food items in different parts of Ethiopia with the objective to compare the Pb and Cd accumulation in different plant species and its variation in the same species collected from industrial and rural places.

Materials and Methods

Atomic absorption spectrophotometer grade stock solutions of 1,000 mg/L Pb $(NO_3)_2$ and $CdCl_2$ supplied by BDH,

H. Baye (✉) · A. Hymete
Department of Pharmaceutical Chemistry, School of Pharmacy, Addis Ababa University, P.O. Box 1176, Addis Ababa, Ethiopia
e-mail: henbab12@yahoo.com; hbhenok@phar.aau.edu.et

England were used to construct calibration curves for Pb and Cd respectively. Distilled-deionized water was used for all analytical works. All glasswares were washed with 2% Extran solution, soaked in 3 N HCl for 24 h, and rinsed with distilled-deionized water before use. Perkin-Elmer A-Analyst 600 Graphite Furnace Atomic Absorption Spectrophotometer (GFAAS) with appropriate hollow cathode lamps was used for analysis of samples (with calculated detection limits of 0.0019 for Pb and 0.0022 for Cd).

A total of 26 samples belonging to 8 different plant species were collected in fresh form from four different places in Ethiopia (Addis Ababa, Kombolcha, Shirka and Bonga). The places were chosen on the basis of the possible presence and absence of extensive environmental pollution. Shirka and Bonga were selected to represent industry free (rural and green) areas while the other two were suspected to contain high level of environmental pollution due to high traffic and presence of industries. The studied plants were selected based on their frequent use by the society for the preparation of herbal drugs and as food additives and the most commonly used part of each plant was considered as experimental target.

Freshly collected plant samples were stored separately in polythene bags and transported to the laboratory for processing. The plants investigated, sites of collection, and plant parts analyzed are given in Table 1. The plant samples were washed using distilled water and dried under shade.

The protocol used to determine levels of the metals in the plant samples was a modification of the method proposed by Rai et al. (2001, 2005). In summery, washed, dried and finely powdered samples (2.00 g each) were digested in Nessler tube containing 15 ml of a mixture of nitric acid and perchloric acid (3:1 ratio) for 2 hours on a water bath at 80°C until formation of brownish gas ceased and volume finally adjusted to 25 mL using distilled-deionized water. Solutions prepared were then diluted with distilled-deionized water until concentrations fall with in the calibration range and stored in labeled acid-washed glass vials. Pb and Cd analysis was carried out immediately on the resultant digests using GFAAS, with the use of prepared standards (run before each batch) to determine sample concentrations. Analyses were carried out in triplicates to ensure precision of results. Metal concentrations were calculated from each replicate absorbance value, which was then used to calculate an average sample metal concentration. All metal concentrations were expressed in mg/kg on a dry-weight basis of the plant sample. All analyses were run in batches, which included known standards, method blanks, and plant samples. The absorbance of a blank sample was determined for background correction. Accepted recoveries ranged from 85% to 105%, and batches with recoveries less than 85% were reanalyzed. Precision and accuracy of analysis were ensured through repeated analysis of samples.

Results and Discussion

Concentrations of Pb and Cd in the plant samples investigated are presented in Table 2. All the 26 analyzed samples were found to contain detectable levels of the two metals. Evaluation of the results obtained was done basically according to three measuring parameters, namely type of studied metals, plant species and site of collection.

Concentrations of Pb in 4 of the samples (15.4%) were observed to exceed the limit of 10 mg/kg specified by WHO (2005) showing obvious signs of environmental contamination.

According to our result, plant samples highly contaminated with lead were *A. sativum* (14.773 ± 0.785 mg/kg), *T. serrulatus* (98.192 ± 1.054 mg/kg), and *H. abyssinica* (13.905 ± 0.166 mg/kg) from Addis Ababa and *T. serrulatus* from Kombolcha (11.562 ± 1.068 mg/kg). Both areas were expected to be potentially polluted due to high industry, vehicle and population density. The results also

Table 1 Plants investigated, places of collection, and parts analyzed

Plant name	Area of collection				Part analyzed
	Addis Ababa	Bonga	Kombolcha	Shirka	
Allium sativum	✔	✔	✔	✔	Bulb
Dodonea angustifolia	✔	✔	✔	✔	Leaves
Hagenia abyssinica	✔	NA[a]	NA[a]	✔	Flowers
Ocimum lamifolium	✔	✔	NA[a]	✔	Leaves
Ocimum utricifolium	NA[a]	✔	✔	NA[a]	Leaves
Ruta chalepensis	✔	✔	✔	✔	Leaves
Thymus serrulatus	✔	✔	✔	✔	Leaves
Zingiber officinale	✔	✔	✔	NA[a]	Rhizome

[a] Place where the plant is not available

Table 2 Summary of results of Pb and Cd concentrations in mg/kg ± SD (n = 3)

Plant	Collection site	Pb(II)	Cd(II)
Ocimum lamifolium	Addis Ababa	0.76 ± 0.01	0.75 ± 0.00
	Bonga	0.39 ± 0.05	0.79 ± 0.06
	Shirka	0.44 ± 0.05	0.75 ± 0.00
Ocimum utricifolium	Bonga	0.36 ± 0.08	0.83 ± 0.04
	Kombolcha	1.12 ± 0.11	0.67 ± 0.06
Allium sativum	Addis Ababa	14.77 ± 0.79	0.13 ± 0.01
	Bonga	1.45 ± 0.11	0.25 ± 0.00
	Kombolcha	0.89 ± 0.02	0.17 ± 0.04
	Shirka	1.04 ± 0.07	0.21 ± 0.01
Thymus serrulatus	Addis Ababa	98.19 ± 1.05	0.46 ± 0.02
	Bonga	1.12 ± 0.02	0.58 ± 0.05
	Kombolcha	11.56 ± 1.07	0.58 ± 0.06
	Shirka	1.05 ± 0.01	0.54 ± 0.05
Zingiber officinale	Addis Ababa	0.34 ± 0.02	0.17 ± 0.06
	Bonga	0.36 ± 0.03	0.25 ± 0.01
	Kombolcha	0.42 ± 0.02	0.25 ± 0.00
Ruta chalapensis	Addis Ababa	1.30 ± 0.06	0.58 ± 0.04
	Bonga	0.41 ± 0.08	0.63 ± 0.01
	Kombolcha	1.42 ± 0.08	0.63 ± 0.02
	Shirka	0.17 ± 0.05	0.71 ± 0.06
Hagenia abyssinica	Addis Ababa	13.91 ± 0.17	0.25 ± 0.00
	Shirka	4.04 ± 0.17	0.29 ± 0.02
Dodonea angustifolia	Addis Ababa	4.81 ± 0.10	1.83 ± 0.06
	Bonga	0.92 ± 0.03	0.46 ± 0.03
	Kombolcha	0.62 ± 0.01	0.42 ± 0.05
	Shirka	1.13 ± 0.04	0.38 ± 0.00

showed that 3 of 4 samples, highly contaminated by Pb, were collected from Addis Ababa indicating relatively higher degree of pollution in the capital.

Comparison of the data for Pb content in the same species collected from different areas revealed that, samples from Addis Ababa contained maximum amount of Pb within each species except for *Z. officinale* and *R. chalepensis*. This shows that the environmental factors seem to exhibit more influence on the specific accumulation of Pb than genetic factor (Gross et al. 1987).

Among the Addis Ababa samples, lowest Pb content was observed for *Z. officinale* (0.342 ± 0.015 mg/kg). On the other hand, among the *Z. officinale* and *R. chalepensis* samples, highest lead content was observed in the samples collected from Kombolcha. Samples taken from Shirka and Bonga were found to contain Pb in concentrations significantly lower than the WHO limit. The maximum Pb level observed for samples taken from shirka was 4.042 ± 0.170 mg/kg in *H. abyssinica*. This may be a good indicator for the contribution of vehicle exhaust to Pb contamination as the samples were taken from the sides of a road. *A. sativum* was found to contain maximum amount of the metal among the samples collected from Bonga (1.451 ± 0.105 mg/kg) for which the reason was not clear. Generally, all samples taken from Addis Ababa except *Z. officinale* were found to contain the highest amount of Pb and *R. chalepensis* samples from Kombolcha were shown to contain significant amount of Pb though it is less than that of samples for Addis Ababa.

With regard to Cd content, 17 of the 26 samples (65.4%) contained the metal in concentrations above the limit recommended by WHO (2005) for medicinal plants. Among the samples analyzed, the highest concentration of Cd (1.833 ± 0.057 mg/kg) was observed in *D. angustifolia* of Addis Ababa and the least in *A. sativum* (0.125 ± 0.007 mg/kg) obtained from the same area. The results in Table 2 clearly indicate that all samples of *O. lamifolium, O. utricifolium, T. serrulatus, R. chelapensis* and *D. angustifolia* contained the metal beyond the WHO limit but samples of *A. sativum, Z. officinale* and *H. abyssinica* contained Cd in concentrations well below the toxic limits irrespective of the area of collection. This could be taken as an indicator for selective accumulation of heavy metals in certain plant species regardless of the area where they grow. This signifies that on the level of species, the accumulation of Cd seems to be dominated by genetic factors rather than by environmental factors (Gross et al. 1987). In addition, most samples collected from rural areas such as Shirka and Bonga were found to contain appreciable amount of Cd; most of them being in toxic levels. This may be associated with extensive use of Cd based fertilizers, pesticides and herbicides as the areas are performing extensive agriculture.

Generally, the concentrations of Pb and Cd found in this work are inline with those reported from different parts of the glob. In Poland, lead content up to 110.2 mg/kg and cadmium level up to 2.9 mg/kg was reported by Królak (2003) and Bloniarz et al. (2001) reported that 70% of herbal preparations for Cd and 42% for Pb were found to contain the metals beyond the acceptable limits. In Malaysia, up to 20.72 mg/kg lead was reported in herbal preparations by Ang et al. (2003). In India, many research outputs including Agoramoorthy et al. (2008) and Barthwal et al. (2008) reported how threatening the problem is in the country. Moreover, similar results were reported from different parts of Africa like Street et al. (2008) from South Africa and Abou-Arab and Abou Donia (2000) from Egypt.

Data were statistically treated by correlation matrix and ANOVA. The results of the treatments are summarized as follows.

Correlation between Cd content and samples of the same plant species collected from different areas was strong (r-values > 0.900) where as the correlation was relatively loose (r-values < 0.720) for lead with in species. But this loose correlation was significantly improved when results of *A. sativum*, *T. serrulatus*, and *H. abyssinica* are ignored. Possible reason for this being the maximum lead content was registered in these plant species that can make the correlations different. Metal contents of different species of *Ocimum* are highly correlated both within the same species and between the two species irrespective of the area of collection. Correlation between metal ions in the green areas (Bonga and Shirka) and those considered as 'polluted' areas (Addis Ababa and Kombolcha) was also examined. In this combination lead was poorly correlated with Cd. Correlation between Pb and Cd concentration within the same area of collection was generally found to be poor though it was positive in all cases.

One way and two-way ANOVA treatments were made at 95% confidence interval. There is a significant difference between metal contents when all the areas of collection and plant species are considered in the calculations (calculated F-values are greater than the tabulated F-values). Except samples taken from Addis Ababa, there was also a significant difference in the metal content of different plant species collected from the same area. On the other side, there was no significant difference in the metal content of the same plant species collected from different areas except *D. angustifolia* samples where significant difference was observed.

References

Abou-Arab AA, Abou Donia MA (2000) Heavy metals in Egyptian species and medicinal plants and the effect of processing on their levels. J Agric Food Chem 48:2300–2304

Agoramoorthy G, Chen FA, Hsu MJ (2008) Threat of heavy metal pollution in halophytic and mangrove plants of Tamil Nadu, India. Environ Pollut 155:320–326

Ang HH, Lee EL, Matsumoto K (2003) Analysis of lead content in herbal preparations in Malaysia. Hum Exp Toxicol 22:445–451

Barthwal J, Nair S, Kakkar P (2008) Heavy metal accumulation in medicinal plants collected from environmentally different sites. Biomed Environ Sci 21:319–324

BBC News (Wednesday, 15 March 2006) http://news.bbc.co.uk/ accessed on 18 March, 2006

Bloniarz J, Zareba S, Rhnama M (2001) Cadmium and lead content in some herbs, herbal preparation and herbal infusions used by children and adults. Przegl Lek 58:39–43

Caldas ED, Machado LL (2004) Cadmium, mercury and lead in medicinal herbs in Brazil. Food Chem Toxicol 42:599–603

Chuang IC, Chen KS, Huang YL, Lee PN, Lin TH (2000) Determination of trace elements in some natural drugs by atomic absorption spectrometry. Biol Trace Elem Res 76: 235–244

Gasser U, Klier B, Kuhn AV, Steinhoff B (2009) Current findings on the heavy metal content in herbal drugs. Pharmeur Sci Notes 1:37–50

Gross R, Auslitz J, Schramel P, Payer HD (1987) Concentrations of Pb, Cd, Hg and others in seeds of *Lupinus mutabilis* and other legumes. J Environ Pathol Toxicol Oncol 7:59–65

Jarup L (2003) Hazards of heavy metal contamination. Br Med Bull 68:167–182

Kadir MM, Janjua NZ, Kristensen S, Fatmi Z, Sathiakumar N (2008) Status of children's blood lead levels in Pakistan: implications for research and policy. Public Health 122:708–715

Królak E (2003) Accumulation of Zn, Cu, Pb and Cd by Dandelion in environments with various degrees of metallic contamination. Pol J Environ Stud 12:713–721

Kwon KY, Jang JH, Kwon SY, Cho CH, Oh HK, Kim SP (2003) Cadmium induced acute lung injury and TUNEL expression of apoptosis in respiratory cells. J Kor Med Sci 18:655–662

Naithani V, Kakkar P (2006) Effect of ecological variation on heavy metal content of some medicinal plants used as herbal tea ingredients in India. Bull Environ Contam Toxicol 76:285–292

Rai V, Agarwal M, Khatoon S, Rawat AK, Mehrotra S (2001) Estimation of Co and Mn in some medicinal plants. Bull Environ Contam Toxicol 66:427–432

Rai V, Agnihotri AK, Khatoon S, Rawat AK, Mehrotra S (2005) Chromium in some herbal drugs. Bull Environ Contam Toxicol 74:464–469

Street RA, Kulkarni MG, Stirk WA, Southway C, Van Staden J (2008) Variation in heavy metals and microelements in South African medicinal plants obtained from street markets. Food Addit Contam Part A Chem Anal Control Expo Risk Assess 25:953–960

WHO (2005) Working document QAS/05.131/Rev.1, Quality control methods for medicinal plant material, Geneva, Switzerland, pp 20–27

Wong MK, Tan P, Wee YC (1993) Heavy metals in some Chinese herbal plants. Biol Trace Elem Res 36:135–142

Zaccaroni A, Amorena M, Naso B, Castellani G, Lucisano A, Stracciari GL (2003) Cadmium, chromium and lead contamination of *Athene noctua*, the little owl of Bologna and Parma, Italy. Chemosphere 52:1251–1258