Assessment of Heavy Metals Concentration in Some Selected Medicinal Plants Collected from BCSIR, Chittagong Cultivation Area in Bangladesh

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This survey was carried out with the aim to assess the level of heavy metals i.e. Lead (Pb), Cadmium (Cd), Chromium (Cr), Cobalt (Co), Nickel (Ni), Copper (Cu), Zinc (Zn) and Manganese (Mn) in different medicinal plants grown at BCSIR, Chittagong cultivation area in Bangladesh during 2010-2011. Atomic absorption spectrometry was used to estimate and evaluate the levels of these metals. The results of this survey showed that the average concentrations detected were ranged from 0.01 to 3.25, 0.01 to 2.01, 0.88 to 5.30, 3.09 to 27.58, 0.41 to 3.50, 0.0 to 8.5, 4.5 to 24.5 and 3.14 to 16.22 mg/kg for Pb, Cd, Cr, Mn, Ni, Co, Zn and Cu respectively. The highest levels of Pb, Cd, Cr, Mn, Ni, Co, Zn and Cu were detected in Ashoka (*Ashok*), Malabar nut (*Basok*), Wild basil (*Kalo Tulsi*), Indian buttercup (*Sarpoganda*), Asparagus (*Satomuli*), and Wild basil (*Kalo tulsi*) respectively. The results of the investigation will be guideline for the users, collectors and practitioners of medicinal plants obtained from the polluted areas for human consumption.

Keywords: Medicinal plants, Heavy metals, Proximate analysis, Toxicity, BCSIR.

Introduction

Human beings are encouraged to take herbal medicines to recover from diseases, these herbs are considered in general to have no side effect. So medicinal plants are consumed worldwide and such plants are also an important source of raw material for pharmaceutical industries. However, these plants contain heavy metals over a wide range of concentrations. Many medical herbs used in formulating these medicines can present a health risk due to the presence of toxic ingredients like heavy metals. The toxicity of heavy metals depends upon the chemical form of elements. Heavy metals are dangerous in the form of their cations and are highly toxic when bonded to the short chains of carbon atoms (Kirmani *et al.,* 2011). Plants may absorb heavy metals from soil, water or air. Medicinal herbs may be easily contaminated during growing and processing. The ability of plants to selectively accumulate essential elements is different for different species and is subjected to certain geochemical characteristics depending on the type of soil (Bin *et al.,* 2001). Usually soil is subjected to contamination through atmospheric deposition of heavy metals from point sources including metalliferous mining, smelting and different industrial activities. Some other sources of soil contamination involve use of fertilizers, pesticides, sewage sludge and organic manures (Singh *et al.,* 1997). Plants readily assimilate such elements through the roots. Metallic ions get dissolved in water and retained. Additional sources of these elements for plants are rainfall, atmospheric dusts and plant protection agents, which could be adsorbed through the leaf blades. The publicity regarding the high level of heavy metals in the environment has created a certain apprehension and fear in the public as to the presence of heavy metal residues in their daily food. Heavy metal toxicity should have been received special attention globally due to neurotoxin, carcinogenic and several other impacts arising from their consumption even at lower contents. However, the public is confused and alarmed about their drug safety.

Heavy metals, such as cadmium, copper, lead, chromium and mercury are important environmental pollutants. Heavy metals, in general, are not biodegradable, have long biological half-lives and have the potential for accumulation in different body organs leading to unwanted side effects (Jarup, 2003). Lead and cadmium are among the most abundant heavy metals and are particularly toxic. The excessive content of these metals in food is associated with etiology of a number of diseases, especially with cardiovascular, kidney, nervous as well as bone diseases (Kudirat *et al.,* 2011). In addition, they are also implicated in causing carcinogenesis, mutagenesis and teratogenesis (Sobukola *et al.,* 2010). Other metals such as copper and zinc are essential for important biochemical and physiological functions and necessary for maintaining health throughout life. The knowledge of Zn toxicity in humans is minimal and the most important information reported is its interference with Cu metabolism. The symptoms that an acute oral Zn dose may provoke include: tachycardia, vascular shock, dyspeptic nausea, vomiting, diarrhoea, pancreatitis and damage of hepatic parenchyma (Adefila *et al.,* 2010). Zinc deficiency results in a variety of immunological defects whereas copper deficiency is characterized by anemia, neutropenia and skeletal abnormalities (Prentice, 1993; Linder *et al.,* 1996).

There are no guidelines to establish a permissible level of metals in herbs. By monitoring the level of metals in medicinal plants one can be able to indicate the level of environmental pollution in that area (Kirmani *et al.,* 2011). The present study is concerned with the assessment of the metal content of some medicinally important herbs and plants. This study has been focused to estimate the concentrations of some toxic or heavy metals ions in the medicinal plants namely *Saraca asoca, Adhatoda zeylanica, Terminalia bellirica, Phyllanthus emblica, Ocimum americanum, Terminalia chebula, Andrographis paniculata, Azadirachta indica, Rauvolfia serpentina, Asparagus racemosus* which were collected from BCSIR, Chittagong, Bangladesh. Eight heavy metals (Cd, Cr, Co, Cu, Pb, Mn, Ni, Zn) have been chosen on the basis of their ability to cause health disorders.

Materials and Methods

The heavy metal contents were determined by AAS using standard analytical procedure. Sample collection is important stage for metal analysis. Samples were generally carefully handled to avoid contamination. Glassware was properly cleaned and the reagents were of analytical grade. Double distilled deionised water was used throughout the study. Reagents blank determinations were used to correct the instrument readings. For validation of the analytical procedure, a recovery study was carried out by spiking and homogenizing several already analyzed samples with varied amounts of standard solutions of the metals. The techniques for sample collection, pretreatment, Standard Preparation, and procedure for metal analyses have been briefly described below:

(A) Sampling: At first ten medicinal plants were collected from BCSIR, Chittagong cultivation area in Bangladesh. Then the samples were first oven dried at 105°C for 24 h. The dried samples were crushed and powdered. Then 50 g of crushed and powdered portion from each item were taken in China dish and heated in a Muffle furnace for 6 h at 600°C for ashing. After ashing, ashes were taken to 250 ml beaker and added 50 ml water and 15 ml concentrated nitric acid. Then sample was returned to a hot plate and kept heated, adding additional acid as necessary until digestion was completed. Then filtrated into a 250 ml volumetric flask and then each sample was made up to the mark with deionised water (Bakar *et al.,* 2011).

(B) Preparation of Standard: Every metal standard solution was prepared for calibration, the instrument for each element being determined on the same day as the analyses were performed due to possible deterioration of standard with time (Cantle, 1982). All samples were prepared from chemicals of analytical grade with double distilled water. 1 gm of metal cadmium, cobalt, nickel and zinc dissolve in aqua regia (1:3) HCl and HNO_3, made up to 1 liter in volumetric flask, thus stock solution of 1000 ppm of Cd, Co, Ni, Zn were prepared. Stock solution of lead is prepared by 1.59 g Pb $(NO_3)_2$ (=1 g Pb) dissolve in 1% HNO_3. Thus a stock solution of 1000 ppm was prepared, similarly stock solution of chromium is prepared by 2.82 g $K_2Cr_2O_7$ (=1 g Cr) dissolve in 1% HNO_3 and diluted to 1 liter (Kumar *et al.,* 2009.). Then 100 ml of 0.05, 0.1, 0.25, 0.5 and 1.0 mg/l of working standards of each metal were prepared from these stock using micropipettes in 5 ml of 2N Nitric acid. Reagent blank was prepared in the same manner of sample preparation without sample to avoid reagents contamination.

(C) Analysis of sample: The atomic absorption instrument was set up and flame condition and absorbance were optimized for the analyte. Then blanks(deionized water), standards, sample blank and samples were aspirated into the flame in AAS (Model – iCE 3000, Thermo Scientific). The calibration curves obtained for concentration vs. absorbance. Data were statistically analyzed using fitting of straight line by least square method. A blank reading was also taken and necessary corrections were made during the calculation of concentration of various elements.

Result and Discussion

In all ten medicinal plants namely *Saraca asoca* (Ashoka), *Adhatoda zeylanica* (Malabar nut), *Terminalia bellirica* (Beleric Myrobalan), *Phyllanthus emblica* (Embolic myrobalan), *Ocimum americanum* (Wild basil), *Terminalia chebula* (Gall nut), *Andrographis paniculata* (Cret), *Azadirachta indica* (Indian lilac), *Rauvolfia serpentina* (Indian buttercup) and *Asparagus racemosus* (Asparagus) were investigated for their heavy metal load. In this respect concentration levels of Cd, Cr, Co, Cu, Pb, Mn, Ni and Zn in units of ppm have been determined. The Table 1 shows the heavy metals concentration in some medicinal plants in previously published results from other parts of the world. The results of our investigation are being presented through the Table 3. These results show that highest concentration of Lead (Pb) was found to be >2 ppm in the plant samples of *Saraca asoca*, *Ocimum americanum*, *Andrographis paniculata* and *Azadirachta indica.*

TABLE 1

Heavy Metals Concentration in Some Medicinal Plants in Previously Published Results from Other Parts of the World

S. No.	Name	Result as ppm								
		Lead (Pb)	Cad-mium (Cd)	Chro-mium (Cr)	Manga-nese (Mn)	Nickel (Ni)	Cobalt (Co)	Zinc (Zn)	Copper (Cu)	Reference
01	*Saraca asoca*	2.35	*BDL	14.38	—	1.79		8.93	3.2	Sathiyamourthy *et al.,* India
06	*Terminalia chebula*	4.08	*BDL	12.93	—	13.44		4.68	4.0	Sathiyamourthy *et al.,* India
08	*Azadirachta indica*	12.35	0.041	1.704	20.91	3.726	1.126	34.89	23.14	Kirmani *et al.,* 2011, Pakistan

*BDL – Below Detectable Limit.

Lead (Pb) content was found high in *Saraca asoca* (3.25 ppm), while *Adhatoda zeylanica* showed low concentration of Pb (0.01 ppm). Most of the accumulated Lead is sequestered in the bones and teeth. This causes brittle bones and weakness in the wrists and fingers. Lead that is stored in bones can reenter the blood stream during periods of increased bone mineral recycling (i.e., pregnancy, lactation, menopause, advancing age, etc.). Lead has been shown to have toxic impact on a variety of metabolic processes essential to plant growth and development, including photosynthesis, transpiration, DNA synthesis and mitotic activity (Wierzbicka, 1999). In soil Pb tightly binds itself to organic soil particles which may decrease the mobility of lead in most soils and may reduce uptake by plants (Kirmani *et al.,* 2011). It has been suggested that the mobility of lead and copper is greater in sandy soils, which tend to lack organic matter than in organic soils. Pb content was found high in Saraca asoca (3.25 ppm). The permissible limit set by FAO/WHO (1984) in edible plants was 0.43 ppm. However, for medicinal plants limit was 10 ppm set by China, Malaysia, Thailand and WHO (Jabeen *et al.,* 2010). In the present investigation, the value of Cadmium (Cd) ranges from 0.01 to 2.01 ppm in various medicinal plants. The maximum concentration (2.01 ppm) of Cd was recorded in *Adhatoda zeylanica*, while minimum concentration (0.01 ppm) was registered in *Terminalia chebula*. The permissible limit set by FAO/WHO (1984) in edible plants was 0.21 ppm. However, for medicinal plants the permissible limit for Cd set by WHO, China and Thailand was 0.3 ppm.

TABLE 2

List of Important Medicinal Plants, Parts Used for Analysis and Traditional Use

S.No.	Plant Name	English name	Local name	Parts used	Traditional uses
01	*Saraca asoca*	Ashoka	*Ashok*	Bark, Leaves	Uterine disorders, Blood purifier, Indigestion, Ulcer etc.
02	*Adhatoda zeylanica*	Malabar nut	*Basak*	Leaves	Cough, Respiratory troubles etc.
03	*Terminalia bellirica*	Beleric Myrobalan	*Bohera*	Fruit	Laxative, Tonic, Cough
04	*Phyllanthus emblica*	Emblic Myrobalan	*Amloki*	Fruit	Hair tonic, Revitalizer, Digestive, Laxative, Vitamin C, etc.
05	*Ocimum americanum*	Wild Basil	*Kalo Tulshi*	Leaves	Colds, Cough, Bronchitis, Dysentery, Indigestion etc.
06	*Terminalia chebula*	Gall nut	*Haritaki*	Fruit	Indigestion, Constipation, Jaundice, Piles, Menstruation, etc.
07	*Andrographis paniculata*	Crete	*Kalomegh*	Whole plant	Liver Diseases, Fever
08	*Azadirachta indica*	Indian Lilac	*Neem*	Leaves	Malaria, Gum and Skin diseases
09	*Rauvolfia serpentina*	Indian Buttercup	*Sarpagondha*	Root	Hypertension
10	*Asparagus racemosus*	Asparagus	*Shatamuli*	Root	Diuretic, Urinary problems

Similarly, permissible limits in medicinal plants for Cd set by Canada were 0.3 ppm in raw medicinal plant material and 0.006 mg/day in finished herbal products (WHO, 2005) (Jabeen *et al.,* 2010). Acute doses (10-30 ppm/day) of Cadmium can cause severe gastrointestinal irritation, vomiting, diarrhoea and excessive salivation. Doses of 25 mg of Cd/kg body weight can cause death. Low-level chronic exposure to Cd can cause adverse health effects including gastrointestinal, hematological, musculoskeletal, renal, neurological and reproductive effects. The main target organ for Cd following chronic oral exposure is the kidney. Intake of Cd can double if one smokes cigarettes because each cigarette contains about 2 mg Cd (Kumar *et al.,* 2007). Chromium is one of the known toxic pollutants in the world. At an elevated concentration it is toxic for both plant and animals. The problems that are associated with chromium involve skin rashes, stomach ulcer, kidney, liver damages, lungs cancer and ultimate death. All of our medicinal plant samples bear the concentration level of chromium that fall into the range of 0.88-5.30 ppm. It has been reported that daily dietary intake of 0.05-0.2 mg (Table 4) of chromium contributes to well being of the human. The permissible limit set by FAO/WHO (1984) in edible plants was 0.02 ppm. However, for medicinal plants the WHO (2005) limits have not yet been established for Cr. Although in medicinal plants, permissible limits for Cr set by Canada were 2 ppm in raw medicinal plant material and 0.02 mg/day in finished herbal products (WHO, 2005) (Jabeen *et al.,* 2010).

TABLE 3

Heavy Metals Concentration in Some Medicinal Plants at BCSIR, Chittagong Cultivation Area

S. No.	Name	Result as ppm							
		Lead (Pb)	Cad-mium (Cd)	Chro-mium (Cr)	Manga-nese (Mn)	Nickel (Ni)	Cobalt (Co)	Zinc (Zn)	Copper (Cu)
01	*Saraca asoca*	3.25	0.34	2.88	17.44	2.35	N/D**	20.5	4.14
02	*Adhatoda zeylanica*	0.01	2.01	5.30	25.84	1.95	N/D**	24.50	3.14
03	*Terminalia bellirica*	0.28	0.14	0.94	15.50	1.1	N/D**	10.4	7.12
04	*Phyllanthus emblica*	0.32	0.165	0.88	23.43	2.05	1.2	4.5	3.30
05	*Ocimum americanum*	2.15	1.36	4.50	27.58	2.90	0.1	22.35	16.22
06	*Terminalia chebula*	0.01	0.01	0.89	3.09	0.93	2.5	8.2	5.94
07	*Andrographis paniculata*	2.69	0.16	2.99	10.65	0.41	8.5	19.1	14.90
08	*Azadirachta indica*	2.18	0.21	2.35	25.04	3.01	0.5	17.13	4.33
09	*Rauvolfia serpentina*	1.31	0.14	2.27	27.44	3.50	0.15	12.6	8.13
10	*Asparagus racemosus*	0.76	0.05	1.95	9.14	1.2	6.1	20.1	4.28
11	Average	1.30	0.46	2.49	18.52	1.94	1.36	15.94	7.15

**N/D – Not detectable.

Manganese is one of the major minerals, which is related to the carbohydrate and fat metabolism. Our studies established that concentration of manganese is in the range of 3.09 ppm to 27.58 ppm in selected medicinal plants. The major sources of manganese in soil are fertilizers, sewage sludge and ferrous smelters. Highest concentration of manganese was recorded for *Ocimum americanum*. It is of the order of 27.58 ppm. Whereas the daily dietary intake of 2-5 mg manganese is considered to be suitable for human adults as shown in Table 4. The permissible limit set by FAO/WHO (1984) in edible plants was 2 ppm. However, for medicinal plants the WHO (2005) limits have not yet been established for Mn (Jabeen *et al.,* 2010).

Nickel regulates the mineral metabolism, enzyme activity and several other metabolic processes in plants. It is used as a fungicide but it is a well known toxic to the germination of some seeds. It causes mitotic disturbances in root tips of some plants. High concentrations of nickel cause severe chlorosis and necrosis in plants and a host of other growth abnormalities and anatomical changes. Generally Nickel and its salts do not affect the human body but in some cases it has been recorded to cause allergic problems as it comes in contact with moist skin. It also adversely affects the lungs and nasal cavities (Kirmani *et al.,* 2011). The highest concentration of nickel (3.5 ppm) was recorded for *Rauvolfia serpentina* and the lowest concentration of nickel (0.41 ppm) was found in *Andrographis paniculata*. The recommended daily dietary intake of nickel is about 0.025 mg per day as shown in Table 4. The permissible limit set by FAO/WHO (1984) in edible plants was 1.63 ppm. However, for medicinal plants the WHO (2005) limits have not yet been established for nickel. (Jabeen *et al.,* 2010). The Cobalt (Co) content varies from N/D to 8.50 ppm. The lowest concentration (<0.01 ppm) of Co was observed in *Saraca asoca, Terminalia bellirica* and *Adhatoda zeylanica*, which was not detectable because the instrument lowest detection limit was 0.01 ppm . On the other hand, *Andrographis paniculata* showed highest concentration of Co (8.5 ppm). Cobalt is beneficial for humans because it is a part of vitamin B12, which is essential for human health. Cobalt is used to treat anaemia in pregnant women because it stimulates the production of red blood cells. Deficiency of Co in diet results into pernicious anaemia, severe fatigue, shortness of breath and hypothyroidism, while overdose may lead to angina, asthma, cardiomyopathy, polycythemia and dermatitis. The safety limit for human consumption of Co is 0.04 mg/day in humans. However, too high concentrations of cobalt may damage human health. And it may cause asthma and pneumonia, vision problems, nausea, heart problems and thyroid damage. Like Cu, Zn is an essential element for plants and animals but slight increase in its levels may interfere with physiological processes. Sufficient Zn is essential to neutralize the toxic effects of Cd. Zinc has been well known to be an important trace element as a cofactor for insulin. In this study, maximum quantity of zinc was detected in *Adhatoda zeylanica* (24.50 ppm) while *Phyllanthus emblica* had the lowest concentration (4.50 ppm). Whereas the recommended daily dietary intake of zinc stands at about 15 mg. The permissible limit set by FAO/WHO (1984) in edible plants was 27.4 ppm. However, for medicinal plants the WHO (2005) limits have not yet been established for Zn (Jabeen *et al.,* 2010). Copper is necessary for body pigmentation in addition to Fe, the maintenance of a healthy central nervous system, prevention of anaemia and is interrelated with the function of Zn and Fe in the body. The acceptable limit for human consumption of Copper is 10 ppm (Nair *et al.,* 1997). The permissible limit set by FAO/WHO (1984) in edible plants was 3.00 ppm. However, for medicinal plants the WHO (2005) limits have not yet been established for Cu (Jabeen *et al.,* 2010). When Cu exceeds its safe level concentration, it causes hypertension, sporadic fever, uremias, coma etc. Generally, plants contain the amount of Cu, which is inadequate for normal growth of plants. Application of micronutrient fertilizers and copper based fungicides may sometimes increase it to the alarming levels. Present investigation reveals that Cu varies from 3.14 to 16.22 ppm. The highest amount was found in *Ocimum americanum* and the lowest in *Adhatoda zeylanica.*

TABLE 4
Daily Needs or the World's Daily Average Up-take of Elements by a Person Weighing 70 kg (Mahan and Scott-Stump, 1996)

S.No.	Element	Average Daily dietary intake (ADDIs) mg/day (Range)
01	Fe	15 (10-28)
02	Mn	2.8 (2-5)
03	Zn	15
04	Cu	2.5 (2-3)
05	Ni	0.025
06	Cr	0.05-0.2
07	Co	0.04
08	Pb	0.415
09	Cd	0.057

The present study provides additional data on heavy metals in selected medicinal plants and also can help in risk assessment of consumer exposure to the expected heavy metal levels. It is therefore suggested that regular survey of heavy metals should be done on all food commodities in order to evaluate whether any health risks from heavy metal exposure do exist, to assure food safety and to protect the end user from food that might be injurious for their health.

Conclusion

People generally use herbal medicine from long time to achieve desirable effects. Consumption of such herbal medicine might reduce various types of diseases. Thus our findings indicate that the medicinal plant or plant parts used for different diseases must be checked for heavy metals contamination in order to make it safe for human consumption.

It should be collected from area not contaminated with heavy metals for local or pharmaceutical purposes. The present study provides a base line data for our efforts directed towards maintaining a healthy life style.

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