

## Heavy Metal Levels in Commonly used Traditional Medicinal Plants

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**Summary:** In the present study a survey of 24 commonly used medicinal plants of Indian subcontinent origin was carried out to evaluate their levels of heavy metals by electrothermal atomic absorption spectroscopy. The results showed that the highest mean value for Cd ( $12.06 \mu\text{g.g}^{-1}$ ), Cr ( $24.50 \mu\text{g.g}^{-1}$ ), Cu ( $15.27 \mu\text{g.g}^{-1}$ ), Pb ( $1.30 \mu\text{g.g}^{-1}$ ), Fe ( $885.60 \mu\text{g.g}^{-1}$ ), Mn ( $90.60 \mu\text{g.g}^{-1}$ ), Ni ( $9.99 \mu\text{g.g}^{-1}$ ) and Zn ( $77.15 \mu\text{g.g}^{-1}$ ) were found in *Lawsonia inermis*, *Murraya koenigii*, *Mentha spicata*, *Beta vulgaris* Linn, *Mentha spicata*, *Lagenaria sicerana standl*, *Lawsonia inermis*, *Embolia officinalis*, respectively. The mean and maximum levels of Cd in plant samples were found higher than the recommended values of the Joint Expert Committee on Food Additives of the Food and Agriculture Organization of the United Nations and the World Health Organization and may constitute a health hazard for consumers. All other heavy metals in medicinal plants were found below the recommended tolerable limits.

### Introduction

Large populations worldwide rely primarily on traditional medicine (largely herbs) to meet their primary healthcare needs [1, 2]. Consequently, the global demand for herbs is large and growing at a faster pace. Factors contributing to the growth in demand for traditional medicine include the increasing human population and the inadequate provision of health facilities in developing countries (Table-1). Traditional medical systems are especially concentrated in Africa and Asia. Some of the more widely familiar traditional medical systems in Asia are Chinese Traditional Medicine, Tibetan Medicine, Ayurveda, Siddha, Unani, and Western Herbal Medicine.

Table-1: Ratios of doctors and traditional medical practitioners (TMPs) (practicing largely plant-based medicine) to patients in East and Southern Africa [28].

Country	Doctor : Patient	TMP: Patient
Ethiopia	1 : 33,000	
Kenya	1 : 833 (urban)	1 : 987 (urban)
Malawi	1 : 50,000	1 : 138
Mozambique	1 : 50,000	1 : 200
South Africa	1 : 17,400	1 : 700-1200
Swaziland	1 : 10,000	1 : 100
Tanzania	1 : 33,000	1 : 350-450
Uganda	1 : 25,000	1 : 708

Various functions of cellular and sub-cellular levels require one or more minerals for the proper functioning of various biological processes. Heavy metals have both a curative and preventive

role in combating diseases. Therefore, it is a major interest to establish the levels of heavy metals in common herbal plants because, at elevated levels, these metals can also be dangerous and toxic [3]. The magnitude of herbs uses during recent decade for the cure of various ailments is evident from a report by the World Health Organization [4], which indicated that many people in developing countries still rely on herbal medicine for treatment of various ailments. Medicinal plants today are cultivated commercially in polluted environments, where soil [5], water [6], and air [7], contain rather high levels of pollutants. Therefore, the environment required for growth and synthesis of these plants is affected drastically, and the possibility that toxic pollutants are deposited in the plants cannot be disregarded. The possibility that toxic pollutants can be translocated to humans and animals through the use of herbs grown in polluted zones has concern for scientists who promote use of herbal medicines. We decided, therefore, to assess the levels of some heavy metals (*i.e.* Cd, Cr, Cu, Fe, Pb, Mn, Ni, & Zn) in commonly used herb's parts *i.e.* root, stem, fruits *etc.*, growing in the Indian sub continent of South-East Asia and also to compare the status of these heavy metals with established international guideline/ recommended values.

### Results and Discussion

Medicinal-herbal plants analyzed for heavy metals, local name, parts used and medicinal

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Table-2: Medicinal-herbal plants under investigation (name, parts studied and medicinal properties).

Sample No	Plant species	Local name	Part used	Medicinal properties
H01	<i>Abelmoschus esculantus</i>	Bhindi	Fruit	Venereal diseases, pneumonia, bronchitis, pulmonary tuberculosis
H02	<i>Achyranthes aspera</i>	Safed aghedo	plant	Laxative, anti-periodic in malaria, stomachic
H03	<i>Agastache foeniculum</i>	Aniseed	Seed	Carminative, pectoral, chest infection
H04	<i>Allium sativum</i> Linn	Lahsan	Root	Antihypertensive, astringent
H05	<i>Aloe vera</i>	Aloe	Leaves	Burns, skin rashes, insect bite, healing wound
H06	<i>Andrographis paniculata</i>	Kalmegh	Whole plant	Stomachic, bactericidal, laxative, tonic, alterative
H07	<i>Avicennia marina</i>	Mangroves	Leaf	Leprosy, headache, rheumatism, hemorrhages, Ulcers
H08	<i>Azadirachta indica</i>	Neem	Leaves	Antiseptic, tonic, spermicidal
H09	<i>Beta vulgaris</i> Linn	Chuqandar	Root	Cooling, diaphoretic
H10	<i>Datura alba</i>	Dahtoora	Leaf	Sedative, hypnotic
H11	<i>Embllica officinalis</i>	Amal	Fruit	Revitalizer
H12	<i>Ferula foetida</i>	Heing	Root-gum	Carminative and intestinal anti-septic
H13	<i>Gentiana kurroo</i>	Nilkhanth	Root	Anthelmintic
H14	<i>Helianthus annuus</i>	Sun Flower	Flower, seed and leaves	Bronchial, laryngeal, pulmonary infection, cough and cold
H15	<i>Ipomoea hederacea</i>	Kaladana	Seed	Narcotic, psychedelic
H16	<i>Lagenaria sicerana standl</i>	Lauki	Fruit	Antibacterial, analgesic and sedative
H17	<i>Lawsonia inermis</i>	Henna	Leaf, seeds	Astringent, jaundice, leprosy, small pox, infection of skin.
H18	<i>Mangifera indica</i>	Aa'm	Leaf	Antidiabetic
H19	<i>Mentha spicata</i>	Mint	Leaves	Carminative, neuralgia, diarrhea
H20	<i>Morus nigra</i>	Mulberry	Leaves, fruit	Infection of mouth, diarrhea, high fever
H21	<i>Murraya koenigii</i>	Curry patta	Leaves	Carminative, pain in kidney, skin infection, diabetes.
H22	<i>Ocimum basilicum</i>	Niazboo	Flower	Stimulant, styptic, diuretic and carminative
H23	<i>Pongamia glabra</i>	Tukham-karanjwa	Seed	Antihyperglycaemic
H24	<i>Zingiber officinale</i>	Drak	Root	Stimulant and used to cure diarrhea

properties are given in Table-2. A total of 8 metals (Cd, Cu, Cr, Pb, Fe, Mn, Ni, and Zn) were determined in the medicinal plant samples by GFAAS. Fig. 1 shows the mean concentration ( $\mu\text{g}\cdot\text{g}^{-1}$  dry weight) of trace metals in the medicinal herbal plants under study. The presence of certain heavy metals at trace levels in medicinal plants may be correlated with therapeutic properties against various health disorders. Whereas, increased levels of heavy metals have been known to cause various health disorders. Joint Expert Committee on Food Additives of the Food and Agriculture Organization of the United Nations and the World Health Organization has recommended provisional tolerable weekly intake (PTWI) and provisional maximum tolerable daily intake (PMTDI) values for certain elements. In present study these recommended values were also compared for the selected plants. The PTWI values were calculated based on estimated daily consumption of selected part of the plant and the mean, median and maximum amount of selected heavy metal [8]. The observed mean, median, and maximum amount was used for the per kg consumption of 60 kg person for 7 days (Table-3).

#### Cadmium (Cd)

The Cd concentration varies between 0.05 to 12.06  $\mu\text{g}\cdot\text{g}^{-1}$  with *Lawsonia inermis* (H17) had the

highest and *Azadirachta indica* (H08) the lowest Cd level (Fig. 1). Mean cadmium level of *Agastache foeniculum*, *Aloe vera*, *Avicennia marina*, *Beta vulgaris* Linn, *Helianthus annuus*, *Lagenaria sicerana standl*, *Lawsonia inermis*, *Mentha spicata*, *Morus nigra*, and *Murraya koenigii* exceeded the WHO recommended limit of 0.30  $\mu\text{g}\cdot\text{g}^{-1}$  [9]. Also the estimated maximum weekly intake of cadmium in all the medicinal plants under study are found many fold higher than the JECFA (Joint Expert Committee on Food Additives of the Food and Agriculture Organization of the United Nations and the World Health Organization) PTWI (Provisional Tolerable Weekly Intake) value of 7.0  $\mu\text{g}/\text{kg}$  body weight [10], except for *Aloe vera*, *Lagenaria sicerana standl* and *Zingiber officinale* (Table-3). The estimated mean weekly intake were also found higher than the PTWI for *Azadirachta indica*, *Beta vulgaris* Linn, *Helianthus annuus*, *Ipomoea hederacea*, *Lawsonia inermis*, *Morus nigra*, *Murraya koenigii* and *Ocimum basilicum*. These estimations were calculated by multiplying the maximum recommended dose of respective parts of medicinal plant and their mean and the maximum levels of metal found in the plant [11]. Similar nature of higher cadmium levels in medicinal plants, vegetables and edibles were also found in other parts of the world Thailand [12] Egypt [13] Turkey [14] Mexico and USA [15] India [16]. Higher level of cadmium poses serious

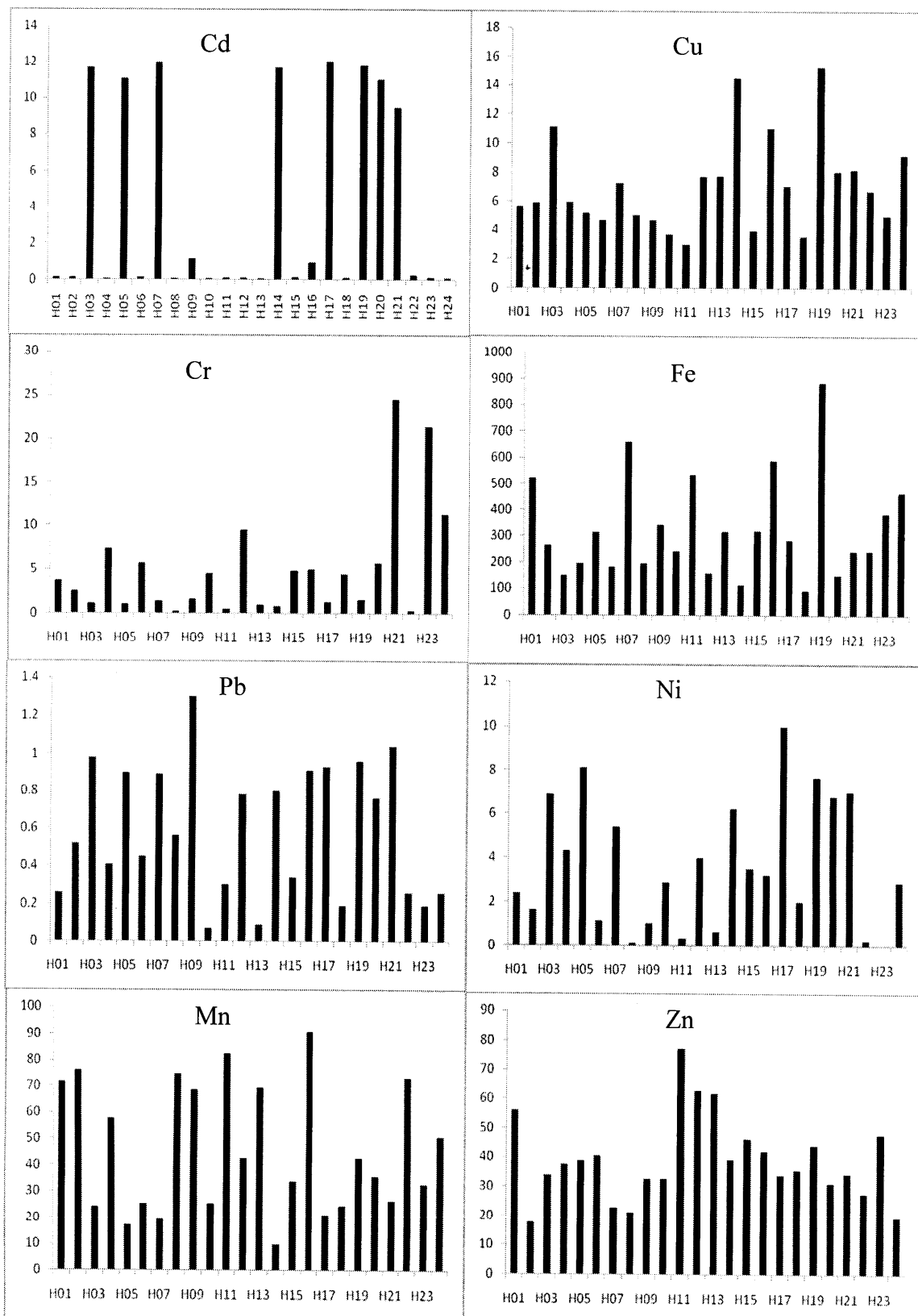


Fig. 1: Heavy metal contents ( $\text{Mean } \mu\text{g.g}^{-1}$  dry weight) from the commonly used herbal medicinal plants.

Table-3: Estimated weekly intake values for selected medicinal herbs in the present study.

	Pb			Cd			Cr			Cu		
	(µg/Kg/Wk)			(µg/Kg/Wk)			(µg/Kg/Wk)			(µg/Kg/Wk)		
	Mean	Median	Maximum	Mean	Median	Maximum	Mean	Median	Maximum	Mean	Median	Maximum
<i>Abelmoschus esculantus</i>	0.69	0.63	1.52	4.60	0.17	14.07	5.89	3.64	28.58	8.26	7.40	17.82
<i>Achyranthes aspera</i>	0.55	0.50	1.21	3.68	0.14	11.26	4.71	2.91	22.87	6.61	5.92	14.25
<i>Agastache foeniculum</i>	0.41	0.38	0.91	2.76	0.10	8.44	3.53	2.18	17.15	4.96	4.44	10.69
<i>Allium sativum</i> Linn	0.69	0.63	1.52	4.60	0.17	14.07	5.89	3.64	28.58	8.26	7.40	17.82
<i>Aloe vera</i>	0.07	0.06	0.15	0.46	0.02	1.41	0.59	0.36	2.86	0.83	0.74	1.78
<i>Andrographis paniculata</i>	0.34	0.32	0.76	2.30	0.08	7.04	2.94	1.82	14.29	4.13	3.70	8.91
<i>Avicennia marina</i>	0.96	0.88	2.12	6.44	0.24	19.70	8.24	5.10	40.02	11.56	10.36	24.94
<i>Azadirachta indica</i>	4.81	4.41	10.62	32.18	1.18	98.49	41.20	25.48	200.08	57.81	51.78	124.71
<i>Beta vulgaris</i> Linn	2.75	2.52	6.07	18.39	0.68	56.28	23.54	14.56	114.33	33.04	29.59	71.26
<i>Datura alba</i>	0.48	0.44	1.06	3.22	0.12	9.85	4.12	2.55	20.01	5.78	5.18	12.47
<i>Emblica officinalis</i>	0.69	0.63	1.52	4.60	0.17	14.07	5.89	3.64	28.58	8.26	7.40	17.82
<i>Ferula foetida</i>	0.48	0.44	1.06	3.22	0.12	9.85	4.12	2.55	20.01	5.78	5.18	12.47
<i>Gentiana kurroo</i>	0.76	0.69	1.67	5.06	0.19	15.48	6.47	4.00	31.44	9.08	8.14	19.60
<i>Helianthus annuus</i>	5.16	4.73	11.38	34.48	1.27	105.53	44.14	27.30	214.38	61.94	55.48	133.61
<i>Ipomoea hederacea</i>	2.06	1.89	4.55	13.79	0.51	42.21	17.66	10.92	85.75	24.78	22.19	53.45
<i>Lagenaria sicerana</i> standl	0.28	0.25	0.61	1.84	0.07	5.63	2.35	1.46	11.43	3.30	2.96	7.13
<i>Lawsonia inermis</i>	1.72	1.58	3.79	11.49	0.42	35.18	14.71	9.10	71.46	20.65	18.49	44.54
<i>Mangifera indica</i>	0.55	0.50	1.21	3.68	0.14	11.26	4.71	2.91	22.87	6.61	5.92	14.25
<i>Mentha spicata</i>	0.69	0.63	1.52	4.60	0.17	14.07	5.89	3.64	28.58	8.26	7.40	17.82
<i>Morus nigra</i>	1.72	1.58	3.79	11.49	0.42	35.18	14.71	9.10	71.46	20.65	18.49	44.54
<i>Murraya koenigii</i>	2.06	1.89	4.55	13.79	0.51	42.21	17.66	10.92	85.75	24.78	22.19	53.45
<i>Ocimum basilicum</i>	1.72	1.58	3.79	11.49	0.42	35.18	14.71	9.10	71.46	20.65	18.49	44.54
<i>Pongamia glabra</i>	1.38	1.26	3.03	9.19	0.34	28.14	11.77	7.28	57.17	16.52	14.79	35.63
<i>Zingiber officinale</i>	0.28	0.25	0.61	1.84	0.07	5.63	2.35	1.46	11.43	3.30	2.96	7.13
<i>Abelmoschus esculantus</i>	381.34	320.18	1033.20	53.27	45.77	105.70	4.29	3.55	11.66	45.37	42.67	90.01
<i>Achyranthes aspera</i>	305.07	256.14	826.56	42.62	36.61	84.56	3.43	2.84	9.32	36.30	34.14	72.01
<i>Agastache foeniculum</i>	228.80	192.11	619.92	31.96	27.46	63.42	2.57	2.13	6.99	27.22	25.60	54.01
<i>Allium sativum</i> Linn	381.34	320.18	1033.20	53.27	45.77	105.70	4.29	3.55	11.66	45.37	42.67	90.01
<i>Aloe vera</i>	38.13	32.02	103.32	5.33	4.58	10.57	0.43	0.35	1.17	4.54	4.27	9.00
<i>Andrographis paniculata</i>	190.67	160.09	516.60	26.64	22.88	52.85	2.14	1.77	5.83	22.69	21.34	45.00
<i>Avicennia marina</i>	533.87	448.25	1446.48	74.58	64.08	147.98	6.01	4.97	16.32	63.52	59.74	126.01
<i>Azadirachta indica</i>	2669.36	2241.26	7232.40	372.89	320.38	739.90	30.03	24.83	81.59	317.60	298.70	630.06
<i>Beta vulgaris</i> Linn	1525.35	1280.72	4132.80	213.08	183.07	422.80	17.16	14.19	46.62	181.49	170.68	360.03
<i>Datura alba</i>	266.94	224.13	723.24	37.29	32.04	73.99	3.00	2.48	8.16	31.76	29.87	63.01
<i>Emblica officinalis</i>	381.34	320.18	1033.20	53.27	45.77	105.70	4.29	3.55	11.66	45.37	42.67	90.01
<i>Ferula foetida</i>	266.94	224.13	723.24	37.29	32.04	73.99	3.00	2.48	8.16	31.76	29.87	63.01
<i>Gentiana kurroo</i>	419.47	352.20	1136.52	58.60	50.35	116.27	4.72	3.90	12.82	49.91	46.94	99.01
<i>Helianthus annuus</i>	2860.03	2401.35	7749.00	399.53	343.26	792.75	32.17	26.60	87.41	340.29	320.03	675.06
<i>Ipomoea hederacea</i>	1144.01	960.54	3099.60	159.81	137.31	317.10	12.87	10.64	34.97	136.12	128.01	270.03
<i>Lagenaria sicerana</i> standl	152.53	128.07	413.28	21.31	18.31	42.28	1.72	1.42	4.66	18.15	17.07	36.00
<i>Lawsonia inermis</i>	953.34	800.45	2583.00	133.18	114.42	264.25	10.72	8.87	29.14	113.43	106.68	225.02
<i>Mangifera indica</i>	305.07	256.14	826.56	42.62	36.61	84.56	3.43	2.84	9.32	36.30	34.14	72.01
<i>Mentha spicata</i>	381.34	320.18	1033.20	53.27	45.77	105.70	4.29	3.55	11.66	45.37	42.67	90.01
<i>Morus nigra</i>	953.34	800.45	2583.00	133.18	114.42	264.25	10.72	8.87	29.14	113.43	106.68	225.02
<i>Murraya koenigii</i>	1144.01	960.54	3099.60	159.81	137.31	317.10	12.87	10.64	34.97	136.12	128.01	270.03
<i>Ocimum basilicum</i>	953.34	800.45	2583.00	133.18	114.42	264.25	10.72	8.87	29.14	113.43	106.68	225.02
<i>Pongamia glabra</i>	762.67	640.36	2066.40	106.54	91.54	211.40	8.58	7.09	23.31	90.74	85.34	180.02
<i>Zingiber officinale</i>	152.53	128.07	413.28	21.31	18.31	42.28	1.72	1.42	4.66	18.15	17.07	36.00

PTWI= Provisional Tolerable Weekly Intake; PMTDI= Provisional Maximum Tolerable Daily Intake

toxicological impacts on human health and ingestion via food, especially plant-based foodstuffs. The kidney, especially the renal tract, is the critical organ of intoxication after exposure to Cd. Excretion is slow, and renal accumulation of Cd may result in irreversible impairment in the re-absorption capacity of renal tubules [17].

#### Chromium (Cr)

Concentration of Cr in *Murraya koenigii* (H21) was found to be highest (24.50 µg.g<sup>-1</sup>) among the analyzed medicinal plants and *Azadirachta*

*indica* (H08) contains the lowest amount of Cr (0.18 µg.g<sup>-1</sup>). In general, Cr concentration ranges between 0.18 and 11.30 µg.g<sup>-1</sup>, except for *Pongamia glabra* (H23) and *Murraya koenigii*. The JECFA and WHO has not recommended a PTWI for Cr. Most of the Cr present in food is in the trivalent form [Cr(III)] which is an essential nutrient. The hexavalent form of Cr [Cr(VI)] is more toxic but is not normally found in food [18]. In present study highest mean, median and maximum weekly intake for Cr was estimated 44.14 µg/kg/wk, 27.30 µg/kg/wk and 214.38 µg/kg/wk respectively in *Helianthus annuus* (Table-3).

### Copper (Cu)

Cu concentration varied between 2.98 and 15.27  $\mu\text{g.g}^{-1}$ . *Mentha spicata* (H19) contains the highest level of Cu and *Emblica officinalis* (H11) contains the lowest. JECFA has recommended a PMTDI (Provisional Maximum Tolerable Daily Intake) of 500  $\mu\text{g/kg}$  bodyweight, which is equivalent to 30000  $\mu\text{g/day}$  for a 60.0 kg person. The results of the present investigation have shown that copper concentration is not a matter of concern from the toxicity point of view for the observed herbs.

### Lead (Pb)

It is appeared from the observed data that the Pb concentration varies between 0.07 to 0.96  $\mu\text{g.g}^{-1}$  in general, except for (H21) *Murraya koenigii* (1.04  $\mu\text{g.g}^{-1}$ ) and (H09) *Beta vulgaris* Linn (1.30  $\mu\text{g.g}^{-1}$ ). *Datura alba* (H10) contain the lowest Pb level and *Beta vulgaris* Linn the highest. The levels of Pb were comparable in *Abelmoschus esculantus*, *Emblica officinalis*, *Ocimum basilicum*, and *Zingiber officinale*. These dietary exposures in the selected herbs are less than the JECFA PTWI of 25  $\mu\text{g/kg}$  bodyweight (0.21 mg/day for a 60.0 kg person). Exposure to Pb is of concern mainly because of its acute toxicity even at trace levels and numerous studies have revealed that it can adversely affect the central and peripheral nervous system, growth and development, cognitive development, renal system, blood circulation, mental retardation, reproductive health and even can cause death [19-23].

### Iron (Fe)

Fe levels varied significantly between the samples (Fig. 1). For example the highest level of Fe was found in (H19) *Mentha spicata* (885.60  $\mu\text{g.g}^{-1}$ ) and lowest in (H18) *Mangifera indica* (90.58  $\mu\text{g.g}^{-1}$ ). Such large variations in Fe contents may arise due to the soil conditions, transportation, treatment and preservation conditions at various points.

### Manganese (Mn)

Similar to that of Fe, Mn shows a wide variation from 9.66 to 90.60  $\mu\text{g.g}^{-1}$  in selected herbs. *Helianthus annuus* (H14) contains the lowest level of Mn (9.66  $\mu\text{g.g}^{-1}$ ) and *Lagenaria sicerana standl* (H16) contains highest (90.60  $\mu\text{g.g}^{-1}$ ). JECFA and WHO has not proposed guidelines for the consumption of Fe and Mn through food chain.

### Nickel (Ni)

The concentration levels of Ni occurred highest in (H17) *Lawsonia Inermis* (9.99  $\mu\text{g.g}^{-1}$ ) and the lowest were in (H08) *Azadirachta indica* (0.12  $\mu\text{g.g}^{-1}$ ) among the selected herbs. The Ni concentration varied from 0.20 to 7.0  $\mu\text{g.g}^{-1}$  in general, except for *Mentha spicata*, *Aloe vera* and *Lawsonia inermis*. The concentration of Ni is comparable in *Zingiber officinale*, *Datura alba*, *Mangifera indica*, and *Abelmoschus esculantus*. The same is being true for *Ipomoea hederacea* and *Lagenaria sicerana standl*. The JECFA has not recommended a PTWI or PMTDI for dietary exposure to Ni, but the WHO has recommended a TDI (Tolerable daily Intake) of 5.0  $\mu\text{g/kg}$  body weight/day (equivalent to 0.30 mg/day for a 60.0 kg adult). Contact with Ni (e.g. from long-term contact with nickel-containing jewellery) can cause dermatitis in sensitised individuals [24].

### Zinc (Zn)

The Zn concentration varied between 17.54 and 77.15  $\mu\text{g.g}^{-1}$  and most of the samples contain Zn concentration in the range of 20.0 to 50.0  $\mu\text{g.g}^{-1}$ . *Achyranthes aspera* (H02) had the lowest Zn concentration and *Emblica officinalis* (H11) the highest. The concentration of Zn were comparable in *Beta vulgaris* Linn and *Datura alba*, and in *Ipomoea hederacea* and *Pongamia glabra*. Zinc is an essential element for humans and the Committee on Medical Aspects of Food Policy, UK [25] has recommended Reference Nutrient Intake (RNIs) of 9.50 mg/day and 7.0 mg/day for adult males and females, respectively. However, high levels of exposure to Zn can be harmful and JECFA has recommended a PMTDI for Zn of 1.0 mg/kg bodyweight which is equivalent to 60.0 mg/day for a 60.0 kg person. The observed concentrations of Zn in selected herbs were found well under the prescribed values.

### Experimental

All the materials and glassware were used of Analytical grade  $\text{HNO}_3$ ,  $\text{HClO}_4$ , and HF of Suprapure<sup>®</sup> quality (Merck). Ultrapure water obtained from Merck was used throughout the study. All glassware and plastic ware were precleaned with 2% v/v nitric acid and soaked in 5% solution of nitric acid for 24 h and then rinsed with ultrapure water. Metal standard solution of Cd, Cu, Cr, Pb, Fe, Mn, Ni, and Zn, containing 1000 mg. L<sup>-1</sup> of each metal

Table-4: Mean concentration ( $\mu\text{g.g}^{-1}$  dry weight, except Fe as % wt), percent recovery (%REC) and percent relative standard deviation (% RSD) of the certified reference materials SRM 1570a SRM 1515 and the two spiked solutions.

Metal	Certified		Measured		% REC		% RSD		Spike 1	Spike 2
	1570a	1515	1570a	1515	1570a	1515	1570a	1515	% Recoveries	% Recoveries
Cd	2.89 ± 0.07	0.013 ± 0.002	3.12 ± 0.068	0.015 ± 0.002	107.96	115.38	2.2	13.3	99.4	104.2
Cr	--	0.3 <sup>a</sup>	--	0.31 ± 0.032	--	104.72	--	--	102.7	103.8
Cu	12.2 ± 0.60	5.64 ± 0.240	10.67 ± 0.469	5.33 ± 0.191	87.46	94.50	4.4	3.6	88.6	93.0
Pb	0.20a	0.47 ± 0.024	0.23 ± 0.029	0.40 ± 0.021	115.00	85.11	13.0	5.3	98.6	100.4
Fe	--	83.0 ± 5.0	--	82.67 ± 2.15	--	99.60	--	2.6	--	--
Mn	75.9 ± 1.90	54.0 ± 3.0	71.67 ± 2.65	53.1 ± 2.60	94.43	98.33	3.7	4.9	101.9	103.28
Ni	2.14 ± 0.10	0.91 ± 0.12	1.39 ± 0.109	0.94 ± 0.209	64.95	103.30	7.9	22.3	82.7	99.4
Zn	82 ± 3.0	12.50 ± 0.30	87.54 ± 10.67	11.8 ± 0.307	106.76	94.40	12.2	2.6	102.6	99.2

a) Non certified (information) values.

were obtained from Merck (Darmstadt, Germany). NIST Standard Reference Material<sup>®</sup> (SRM) 1570a trace elements in spinach leaves and 1515 apple leaves from national institute of standards and technology, USA were used to obtain data quality objectives (DQO).

Heavy metals (Cd, Cu, Cr, Pb, Fe, Mn, Ni, and Zn) were analyzed in plant samples by electrothermal atomic absorption spectrometry (ETAAS). The analysis was performed using Perkin Elmer AAnalyst 700 equipped with deuterium background correction and graphite furnace. 24 different medicinal herbs were purchased from local markets in Karachi and Lahore cities of Pakistan. The medicinal herbs, common name, parts used (leaves, flowers, fruits, *etc.*) their therapeutic use and activity are shown in Table-2. The method described by Gomez *et al.* [26] were used for the preparation of sample and analysis. Precisely, the samples were ground and sieved to ensure a particle size <200  $\mu\text{m}$ . The powdered samples were dried for 24 hrs at a temperature of 104 °C and stored in polyethylene bags prior to analyses. About 2 g dried powder sample of each herb was digested in a Teflon vessel with 20 mL of conc.  $\text{HNO}_3$  near to dryness and then 10 mL  $\text{HClO}_4$ , HF was added to digest the silica salt until white fumes were observed. The residue was quantitatively transferred to 25 mL volumetric flask and made upto volume with ultrapure water.

Prescribed quality assurance (QA) and quality control (QC) procedures were strictly followed throughout the study. The level of accuracy and QC for the determination of heavy metal concentrations comprised of measurement of SRMs, determination of spike recovery for selected heavy metals and measurements of duplicate for each batch of samples. Calibration standards of each element were obtained by appropriate dilution of the stock

solution, the qualified calibration curve correlation coefficients ( $R^2$ ) for each analytes were greater than 0.995. The coefficient was calculated using the inverse of the standard's concentration squared ( $1/X^2$ ) as the weighting factor. The SRMs, NIST standard 1570a and 1515 were used for the method validation. Percentage recoveries ranged between 64.95% and 115.20 % with % RSDs for mean recoveries below 13 % for SRM 1570a. For SRM 1515, percentage recoveries ranged from 85.11 % to 115.38 % whereas, %RSD below 13.3 except for Ni (Table-4). The spikes recoveries are generally within 85-115%, as specified by the EPA, two representative samples were spiked with known amount of analyte and their recoveries were within 82 to 105% limits (Table-4).

## Conclusions

Various locations of diverse nature and compositions of harvesting of herbs and medicinal plants in Indian sub continent of South-East Asia can expose to various levels of heavy metals causes *grounds* for concern because of the crucial role of heavy metals both as contaminant and as at the same time therapeutic agent. In the light of the present preliminary investigations and absence of any local food law for monitoring and regulation can cause serious health concerns as it is observed in case of Cd concentration levels. The WHO [4, 27] has provided guidelines for the quality assurance and quality control of herbs and/or medicinal plants and it can be adopted locally as a first step towards the health safety of the inhabitants. In case of all other heavy metals, it is found that all the herbs under study contain levels within the safe limits.

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