Investigation of Metal Contents in Some Medicinally Important Plants Using Atomic Absorption Spectroscopy

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Summary: A large number of indigenous plants used as foods and medicines, around the world. Only few of these plants have been analyzed for their metal contents whereas several remain unanalyzed. Present study has been undertaken to investigate micronutrient and macronutrient metals among some hypoglycemic and non-hypoglycemic plants using atomic absorption spectroscopy. Chromium present at ppb level, was studied by graphite furnace atomic absorption spectroscopy equipped with hollow cathode lamp and Zeeman background correction. The selected herbs resulted 0.132-1.8475mg/g Fe, 0.0043-0.0227mg/g Cr, 0.0278-0.1108mg/g Zn, 0.0102-0.0345mg/g Cu and 0.0095-0.6123mg/g Mn as micro-nutrients. However these metals are present in significant quantities but under the safe and adequate limits. The water extract of these samples have rather low quantities.

Introduction

Transition metals play significant role in various metabolic processes in human body. There is evidence that a disturbance in the concentration of trace metals can produce various psychiatric symptomatology [1]. The essential and the toxic metals may both be present in a variety of substance around us, such as water, soil, air, food etc.

A balanced diet provides sufficient intake of mineral salts. Those supplied by the medicinal plants are associated with the other active principles and the medicinal action of such mineral constituents has supplementary importance [2-3]. Trace elements are crucial to virtually all biochemical and physiological processes in plants, animals and human beings. Out of these iron, zinc, manganese, copper, chromium, magnesium, calcium, sodium and potassium are now thought to be essential for animal's life [4-6]. Medicinal properties have been attributed to a large variety of plants [7-8] cultivated or naturally grown in different parts of Asian countries.

Herbal medicines are beneficial having less side affects [9-10]. In order to understand the use of medicinal plants and their modes of action, it is necessary to know some thing about their active constituents [11-12]. The World Health Organization estimates that 4 billion people, or 80% of the world's population, use herbal medicine for some aspect of primary health care [13-14].

It is imperative to analyze the plants for their trace metal contents, which has healing power for mankind in numerous ailments and disorders [15-16]. One of such disorder causes diabetes, a widespread disease in the world [17]. The highest rate of diabetes occurs in America where it is the sixth leading cause of death [18]. Pakistan is facing a similar threat. The result of survey conducted by the Diabetic Association of Pakistan, shows that 11.2% of the population above the age of 25 is suffering from diabetes [19]. Alternative treatments for diabetes have become increasingly popular during the last few years, including medicinal herbs, nutritional supplementation, acupuncture, and hot tub therapy [20].

Three classes of herbs are chosen for the investigation of above said metal ions. One, those which are famous as antidiabetic(#1 to 4), 2nd group is moderate antidiabetic(#5 to 9) and third are those which have no known antidiabetic activity, chosen for comparison (Table-1).

Results and Discussion

All the 12 samples, which are mentioned in present study were analysed for the selected metals. (Table-1).

Metals were classified into three groups according to their degree of effectiveness. One which

Table-1:Description of Selected Samples [27-38]

Short name	Botanical Names	General Name	Family	Part of Plant	Medicinal Use	Way of using	Dosage mg/day
ACV	Adiantum capillus veneris	Avenca	Adiantacea	Stems	Antdiabetic	WE	400-600
MC	Momardica charancia	Bitter Melon/ karela	Cucurbitaceae	Fruits	Antdiabetic	Powder	300-1500
EJ	Eugenia jambolana	Jambul/ jaman	Myrtaceae	Seeds	Antdiabetic	powder	(200-500
GS	Gymnema sylvestre	Gurmar	Asclepiadaceae	Leaves	Antdiabetic	WE	400-600
AS	Allium sativum	Garlic	Lilaceae	Cloves	Moderate Antdiabetic	Fresh / oil	2-4g/8mg oil
TF	Trigonella foenm-graecm	Fenugreek	Leguminaceae	Seeds	Moderate Antdiabetic	powder/ WE	10-100g
PG	Pongamia glabra	Karanjwa	Leguminaceae	Seeds	Moderate Antdiabetic	WE	N.R
SH	Sphaeranthus hirtus	Paneer herb	Asteracea	Leaves+Fruits	Moderate Antdiabetic	WE	–
CI	Clerodendrom inerme	Dum Dum	Verbenaceae	Leaves	Moderate Antdiabetic	WE	–
EC	Ervatamia coronarie	Chandni	Apocynaceae	Leaves	Non Antdiabetic	WE	–
PL	Polyalthia longifolia	Ulta Ashoak	Annonaceae	Leaves	Non Antdiabetic	WE	–
HR	Hibiscus rosasinensis	China Rose	Malvaceae	Leaves	Non Antdiabetic	WE	250

WE= Water extract

are thought to be effective in diabetes e.g. Cr, Zn, Cu and Mn. Second those which are non-effective and may cause hindrance in the cure of this disease like iron and third those metals which are found in bulk, e.g. non transition metals (i.e. alkali and alkaline earth metals) [21-23].

All metals were examined by Atomic absorption Spectroscopy whereas, chromim that is present at ppb level, was investigated by Graphite furnace Atomic absorption Spectroscopy, which has accuracy equivalent to ICP (Inductive Coupled Plasma, the most modern technique for analyses of ultra trace quantities of metals) [24].

It is observed that three of these medicinal plants were found to be very rich in all metals, they were Adiantm capillus veneris(ACV), gymnema sylvester(GS) and sphaeranthus hirtus(SH) and somewhat trigonella foenum graecum(TF) also. They are considered as good medicinal plants. Even in their water extract, they have remarkable amount of Cr, Zn, Cu and Mn, especially in GS (Table-2 & 3). So their water extract as well as the whole may be a good source of metals.

Bulk metals do not show any regular or notable sequence. Mg was found high in those samples which were selected in the form of leaves. Calcium was found high in those, selected as stem except GS, which showed a high concentration of Ca (these metals were found in high amount in acid digested samples as compared to water extract (Table-4 & 5).

The selected herbs contained 0.132-1.8475mg/g Fe, 0.004325-0.0227mg/g Cr, 0.0278-0.1108mg/g Zn, 0.0102-0.0345mg/g and 0.0095-0.6123mg/g Mn as micro-nutrients. Whereas 0.2-29mg/g Na, 2.5-55mg/g, 0.1-9mg/g Mg and 0.07-10.6mg/g Ca were found.

Table-2:Micro-Nutrient Metals in Acid Digested Samples of Vegetables and Herbs

Sample ID	Fe mg/g±SD	Cr mg/g±SD	Zn mg/g±SD	Cu mg/g±SD	Mn mg/g±SD
ACV	0.7383±0.036	9.225E-03±0.002	0.0620±0.004	0.0102±0.007	0.0565±0.004
MC	0.3993±0.040	6.975E-03±0.001	0.0723±0.007	0.0172±0.020	0.0363±0.004
EJ	0.2348±0.000	6.100E-03±0.001	0.0278±0.001	0.0168±0.008	0.0095±0.001
GS	1.3835±0.040	2.003E-02±0.016	0.1070±0.003	0.0293±0.015	0.6123±0.003
AS	0.1320±0.009	6.175E-03±0.003	0.0573±0.002	0.0102±0.010	0.0178±0.018
TF	0.7023±0.030	2.268E-02±0.016	0.1108±0.013	0.0267±0.002	0.0280±0.013
PG	0.2330±0.008	1.233E-02±0.005	0.0968±0.008	0.0345±0.014	0.0425±0.008
SH	1.8475±0.030	1.(193E-02±0.02	0.0680±0.012	0.0223±0.012	0.1010±0.000
CI	1.2050±0.001	1.008E-02±0.003	0.0690±0.001	0.0309±0.016	0.1520±0.001
EC	1.1313±0.060	4.325E-03±0.003	0.0485±0.007	0.01(19±0.004	0.3070±0.016
PL	0.6440±0.006	6.025E-03±0.007	0.0703±0.003	0.0155±0.013	0.1275±0.003
HR	0.5788±0.035	5.425E-03±0.004	0.0570±0.004	0.0107±0.004	0.0948±0.004

Table-3:Micro-Nutrient Metals in Water Extract of Vegetables and Herbs

Sample ID	Fe	Cr	Zn	Cu	Mn
	mg/g±SD	mg/g±SD	mg/g±SD	mg/g±SD	mg/g±SD
ACV	0.0014±0.006	6.827E-05±0.09	0.0062±0.006	0.0004±0.002	0.0095±0.007
MC	0.0147±0.001	2.089E-04±0.39	0.0189±0.015	0.0027±0.004	0.0077±0.004
EJ	0.0035±0.001	1.112E-04±0.27	0.0045±0.001	0.0020±0.001	0.0030±0.006
GS	0.0110±0.003	2.108E-04±0.016	0.0116±0.008	0.0031±0.002	0.0701±0.020
AS	0.0125±0.009	4.170E-05±0.05	0.0171±0.002	0.0013±0.001	0.0085±0.002
TF	0.0018±0.001	9.400E-05±0.46	0.0016±0.002	0.0011±0.002	0.0011±0.004
PG	0.0087±0.022	1.356E-04±0.86	0.0148±0.016	0.0024±0.003	0.0106±0.005
SH	0.0135±0.004	6.609E-04±0.76	0.0080±0.000	0.0035±0.001	0.0106±0.012
CI	0.0059±0.001	1.427E-04±0.03	0.0058±0.001	0.0014±0.004	0.0270±0.013
EC	0.0052±0.001	2.122E-04±0.07	0.0105±0.001	0.0014±0.003	0.0474±0.028
PL	0.0017±0.003	1.082E-04±0.18	0.0037±0.004	0.0010±0.003	0.0208±0.044
HR	0.0029±0.002	4.595E-04±0.93	0.0029±0.001	ND	0.0085±0.001

Table-4: Macro-Nutrient Metals in Acid Digested Samples of Vegetables and Herbs

Sample ID	Na mg/g	K mg/g	Mg mg/g±SD	Ca mg/g±SD
ACV	1.3750	6.5000	2.3075±0.004	9.7925±0.056
MC	(19.375	52.500	3.9875±0.017	3.0700±0.015
EJ	3.1250	18.750	0.3723±0.001	0.0838±0.006
GS	6.6250	55.000	9.0650±0.003	10.6400±0.006
AS	0.4000	24.375	1.3125±0.002	0.2950±0.006
TF	7.2500	23.750	2.5313±0.006	1.0600±0.008
PG	4.8750	39.375	2.3000±0.004	1.0700±0.006
SH	7.5000	55.000	4.1725±0.006	2.4825±0.005
CI	21.250	39.375	4.6475±0.004	4.1250±0.003
EC	0.6375	8.2500	0.3645±0.018	0.2108±0.002
PL	4.5000	36.250	4.4000±0.008	1.3375±0.001
HR	29.375	17.500	5.0375±0.004	2.0925±0.008

Table-5: Macro-nutrient Metals in Water Extract of Vegetables and Herbs

Sample ID	Na mg/g	K mg/g	Mg mg/g±SD	Ca mg/g±SD
ACV	0.4933	2.5158	0.1833±0.011	0.0730±0.002
MC	5.4186	20.689	1.07(19±0.017	0.1892±0.007
EJ	1.2(192	6.3400	0.5735±0.000	0.2907±0.004
GS	1.7964	11.477	1.3832±0.011	0.5758±0.004
AS	0.2780	8.9360	0.4031±0.002	0.3882±0.007
TF	1.3056	4.4763	0.1533±0.002	1.8838±0.016
PG	1.6759	12.433	0.6649±0.004	0.4563±0.010
SH	2.1834	15.232	0.5910±0.006	1.1089±0.013
CI	6.7755	12.583	1.3348±0.001	1.5351±0.016
EC	0.4769	17.882	1.8935±0.003	0.5285±0.001
PL	0.2454	8.8358	0.9248±0.005	1.5070±0.021
HR	3.7708	3.7708	0.8946±0.008	0.1131±0.001

Finally it can be concluded that even these medicinal plants are good source of metals. These metals may be effective or toxic.

Diabetes is a disease which actually can not be cured but controlled. Therefore the herbs or vegetables related to its treatment are generally continuously utilized and may produce toxicity or even over dosage of essential metals. Therefore their continuous use should be avoided.

Experimental

Reagents and Glassware

All the chemicals and reagents were of analytical-reagent grade, purchased from Merck and BDH Laboratory Supplies. Appropriate quality assurance procedures and precautions were carried out to ensure reliability of the results. Samples were generally carefully handled to avoid any contamination. Glasswares were properly cleaned, and the reagents (Nitric acid and distilled water) were of analytical grade.

Standard Solutions

Reagent blank determinations were used to correct the instrument readings. Calibration standards were made by dilution of the high purity commercial BDH metal standards for atomic absorption analysis.

Sampling

Samples of herbs were purchased from local markets of Karachi, Pakistan. Samples were cleaned, smashed and oven-dried at 105°C [25] and sub-sampled for heavy metal analysis. Accurately weighed samples were mineralized (wet ashing) according to the recognized method [26].

Graphite Furnace Atomic Absorption Spectroscopy

A Hitachi model Z 8000 Atomic Absorption Spectrophotometer equipped with hollow cathode lamp and Zeeman background correction and microprocessor control was used in the flame version. Iron, Magnesium, Manganese and Zinc were analyzed, using air-acetylene flame, by standard calibration technique (Table-6) whereas Chromium was investigated using argon and standard addition method (Table-7).

Table-6: Standard Conditions for Hitachi Z-8000 Atomic Absorption Spectrophotometer

S. #	Metals	λ (nm)	SBW (nm)	Sensitivity Check (ppm)	Flame Gases	Max. Lamp Current mA
1.	Fe	248.3	0.2	5.0	*A – Ac	10
2.	Cr	359.3	1.3	4.0	**Ar	7.5
3.	Zn	213.9	1.3	1.0	A – Ac	10
4.	Cu	324.8	0.7	4.0	A – Ac	10
5	Mn	279.6	0.4	2.5	A – Ac	7.5
6.	Mg	285.2	1.3	0.3	A – Ac	7.5
7.	Ca	422.7	0.7	4.0	A – Ac	06

*A – Ac = Air Acetylene Flame **Ar = Argon

Table-7:Conditions for Chromium Estimation in Water Extract by AAS Using Graphite Furnace

Process	Temperature °C	Time (sec)
	50-65	30
Drying	60-75	30
	80-180	30
Ashing	700	30
Atomization	2900	10
Cleaning	3000	02
Cooling	0	30

Flame Photometry

Sodium and potassium were analyzed in all plant samples by flame photometer model Corning 410. The intensity of these metals in sample solutions was recorded by using deionized distilled water as reagent blank. Concentrations were calculated with the calibration curve.

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