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ANALYSIS OF VARIOUS METAL IONS IN SOME MEDICINAL PLANTS USING ATOMIC ABSORPTION SPECTROPHOTOMETER

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ABSTRACT

Metal ions such as iron , lead, copper, nickel, cadmium , chromium and zinc were investigated in medicinally important plants *Alstonia scholaris, Tabernaemontana coronariae, Asparagus racemosus, Mimosa pudica, Leucas aspera and Adhatoda vasica* applying atomic absorption spectrophotometer techniques. The purpose of this study was to standardize various metal ion Contamination in indigenous medicinal plants. Maximum concentration of lead was present in *Leucas aspera* and *Adhatoda vasica* followed by *Alstonia scholaris*, *Tabernaemontana coronariae* and *Asparagus racemosus*. The concentration of lead in *Mimosa pudica* was below the detectable level. The maximum concentration of zinc was detected in *Adhatoda vasica* followed by *Leucas aspera*, *Asparagus racemosus*, *Tabernaemontana coronariae*, *Alstonia scholaris* and *Mimosa pudica*. The concentration of Cadmium, nickel and chromium was below the detectable level.

INTRODUCTION: Although heavy metals are present in food in very minute quantities the very human existence is due to their role in body metabolism. It has been established that whatever is taken as food might cause metabolic disturbance if it does not contain the permissible upper and lower limits of heavy metals. Thus both deficiency and excess of essential micronutrients (e.g., iron, zinc, chromium etc.) may produce undesirable effects [1, 2, 3].

Beside, several organic compounds, it is now well established that many trace elements play a vital role in general well being as well as in the cure of diseases [4, 5]. Copper is essential metallo element, like essential amino acids, fatty acids, vitamins required for normal metabolic process.

The approximate daily dietary requirement of iron is 8 to 13 mg for children, 15 to 20 mg for menstruating women and 10 to 15 mg for men. Significant amount of zinc at concentration around 10µm are present in mammalian brain either protein bound or chelatable form [6, 7, 8].

Zinc permanganate has greater astringent activity compared to the potassium salt. So it is used for rural irrigation. They are corrosive and have weak antiseptic properties. Lead acts as an astringent because of the formation of lead proteinate. Mercury is used as antiseptic and preservative spermicides and diuretics. 1% solution of cadmium sulphide is used in treating seborrhoeic dermatitis and dandruff.

In the present study, six indigenous medicinal plants *Adhatoda vasica*, *Alstonea schlolaris*, *Mimosa pudica*, *Tabernaemantana coronariae*, *Asperagus racemosus* and *Leucas aspera* were studied for their bioactivity and the presence of metal ions.

Methods:

Plant material and preparation of the extract: Fresh leaves of *Adhatoda vasica*, *Alstonea schlolaris*, *Mimosa pudica*, *Tabernaemantana coronariae*, *Asperagus racemosus* and *Leucas aspera* were free from disease were collected from local areas in Davanagere district, Karnataka. Freshly collected leaves are shade dried and then powdered using a mechanical grinder.100 grams of pulverized leaf materials were soaked in 500 ml of methanol (LR grade, Merck, India) and kept on a rotary shaker for 24 h. At the end of extraction, extract was filtered under vacuum through a Whatman No. 1 filter paper and the process repeated until all soluble compounds had been extracted. The filtrate obtained was concentrated *in vacuo* using a Rotavapor (Buchi Flawil, Switzerland). A portion of the residue was subjected to screening for metal ion analysis.

Analysis of Metal ions: 1 gm of the residue after solvent extraction is digested with 10 ml of sulphuric acid and the solution is filtered and the volume is made up to 100 ml using volumetric flask. Atomic absorption Spectrophotometer was used to detect the heavy metals.

1. **Determination of chromium:**

   Preparation of Chromium Standard Calibration Solutions: 2.8290 gm of potassium dichromate ($K_2Cr_2O_7$) is dissolved in distilled water and acidified with 2ml of concentrated nitric acid and made up the volume to 1,000 ml in a volumetric flask. This is 1,000 parts per million (ppm) chromium. 2ml of 10 ppm solution is taken in a volumetric flask and diluted to 100 ml to get 0.2 ppm chromium solution. 0.4 ppm, 0.6 ppm, 0.8 ppm chromium solutions are prepared by taking 4 ml, 6 ml and 8 ml of chromium 10 ppm solutions and diluting it to 100 ml. AAS 201/203 is optimized with chromium hollow cathode lamp and 0.5 ppm solution was checked to produce a minimum of 0.6nm absorbance. The absorbance for 0.2 ppm, 0.4 ppm, 0.6 ppm and 0.8 ppm solutions are recorded and then the absorbance for the sample solution is checked.

Calculations;

$$Cr\% = (ppm) \times \frac{100}{10} \times \frac{100}{10} \times \frac{100}{10} \times 100 \times \frac{100}{1.0} \times 10^{-6}$$

2. **Determination of copper**:

   Preparation of standard calibration solutions: 0.3927 gm of $CuSO_4.5H_2O$ is weighed and dissolved in 5ml of concentrated hydrochloric acid. It is diluted to 100 ml with distilled water in a volumetric flask. This is 1,000 ppm copper solution. 10 ml of this solution is diluted to 100 ml with distilled water to get 1 ppm copper solution. Similarly 2ml, 3ml, 4ml and 5 ml of the 100 ppm solution is diluted to 100 ml in volumetric flasks to get 2 ppm, 3 ppm, 4 ppm and 5 ppm copper solutions. The AAS 201/203 is optimized with copper hollow cathode lamp and checked with 5 ppm copper solution to produce a minimum of 0.6 nm absorbance.

Calculations:

$$Cu\% = (ppm) \times \frac{100}{10} \times \frac{100}{10} \times 100 \times \frac{100}{1.0} \times 10^{-}6$$

(1.0 is the weight of the sample in solution A)

3. **Determination of iron**:

   Preparation of standard calibration solutions: 0.7021 gm of ferrous ammonium sulphate is dissolved in 10 ml of concentrated hydrochloric acid and the volume is made up to 100 ml in a volumetric flask with the distilled water. This is 1,000 ppm of iron. 10 ml of the above solution is diluted with distilled water to make up 100 ml in a volumetric flask. This is 100 ppm of iron. 2ml of 100 ppm iron solution is added with 2 ml of concentrated hydrochloric acid and made up to 100 ml to get 2 ppm of iron solution. 4 ml, 6 ml, 8 ml and 10 ml of iron 100 ppm is added with 2ml of concentrated hydrochloric acid and made up to 100 ml with distilled water to get 4, 6, 8, and 10 ppm solution respectively. The AAS 201/203 is optimized with iron lamp and checked with 10 ppm of iron solution to produce minimum of 0.5nm absorbance.

Calculations:

$$\% \text{ Iron} = (\text{ppm}) \times \frac{100}{10} \times \frac{100}{10} \times 100 \times \frac{100}{1.0} \times 10^{-6}$$

1.0 is the weight of the sample in solution 'A'.

4. **Determination of zinc**:

   Preparation of zinc standard calibration solutions as follows: 0.4390 gm of $ZnSO_4.7H_2O$ is weighed and dissolved in 10 ml concentrated hydrochloric acid and made up to 100 ml with distilled water in a volumetric flask. This is 1000 ppm of zinc. 0.2ppm, 0.4 ppm, 0.6 ppm, 0.8 ppm zinc solutions were prepared by taking 4 ml, 6 ml and 8 ml of zinc 10 ml solution and diluting it to 100 ml. AAS 201 / 203 is optimized with zinc hollow cathode lamp and checked with 0.8 ppm solution to produce a minimum of 0.6nm absorbance.

Calculation:

$$\text{Zn } \% = (\text{ppm}) \times \frac{100}{5} \times \frac{100}{10} \times \frac{100}{10} \times 100 \times \frac{100}{1.0} \times 10^{-6}$$

5. **Determination of Nickel**: 0.44576 gm of nickel sulphate is dissolved in 10 ml of concentrated hydrochloric acid and made up to 100 ml with distilled water in a volumetric flask. This is 1000 ppm of nickel. 0.2ppm, 0.4 ppm, 0.6 ppm and 0.8 ppm nickel solutions are prepared by taking 4 ml, 6 ml and 8 ml of nickel 10 ppm solution and diluting it to 100 ml. AAS 201 /203 is optimized with nickel hollow cathode lamp and check with 0.8 ppm solution to produce a minimum of 0.6 nm absorbance.

Calculate nickel content;

$$\text{Ni } \% = (\text{ppm}) \times \frac{100}{5} \times \frac{100}{10} \times \frac{100}{10} \times 100 \times \frac{100}{1.0} \times 10^{-6}$$

6. **Determination of Lead**: 1.5982 gm of lead nitrate solution is dissolved in 10 ml of concentrated nitric acid and made up to 100 ml with distilled water in a volumetric flask. This is 1000 ppm of lead solution. 0.2ppm, 0.4 ppm, 0.6 ppm and 0.8 ppm lead solutions are prepared by taking 4 ml, 6 ml and 8 ml of lead 10 ppm solution and diluting it to 100 ml. AAS 201/ 203 is optimized with lead hollow cathode lamp and checked with 0.8 ppm solution to produce a minimum of 0.6nm absorbance.

Calculate lead content;

$$\text{Pb\%} = (\text{ppm}) \times \frac{100}{5} \times \frac{100}{10} \times \frac{100}{10} \times 100 \times \frac{100}{1.0} \times 10^{-6}$$

RESULTS: Analysis of metal ions was done using Atomic Absorption Spectrophotometer. The concentration of iron in the leaves of *Adhatoda vasica* is about 5.498 mg/g, the concentration of zinc is about 1.850 mg/g, the concentration of lead is about 0.412 m/g and copper is 0.100 mg/g. The concentration of nickel, cadmium and chromium are below detectable level (**Table 1**).

Leucas aspera has maximum concentration of iron that is (8.206 mg/g), the concentration of zinc is about 1.793 mg/g, 0.412 mg/g of lead and the concentration of copper is about 0.129 mg/g. The concentration of nickel, chromium and cadmium is below detectable level (Table 1).

Mimosa pudica has about 6.458 mg/g of iron. The concentration of lead is below the detectable level. The plant has about 0.090 mg/g of copper, 0.947 mg/g of zinc. The concentration of nickel, chromium and copper are below the detectable level (Table 1).

Asparagus racemosus has about 6.680 mg / g of iron, 0.079 mg/g of lead, 0.085 mg/g of copper and 1.570 mg/g of zinc. The concentration of nickel, chromium and cadmium is below detectable level (Table 1).

Tabernaemontana coronariae leaves have about 6.320 mg/g of iron, 0.301 mg/g of lead, 1.544 mg/g of zinc, 0.020 mg/g of copper. The concentration of nickel, chromium and cadmium are below the detectable level. The leaves of *Alstonia scholaris* has about 5.069 mg/g of iron, 1.157 mg/g of zinc and the concentration of lead are about 0.0329 mg / g. The concentration of copper, chromium, cadmium and nickel is below detectable level (Table 1).

Maximum concentration of lead was present in *Leucas aspera* and *Adhatoda vasica* followed by *Alstonia scholaris*, *Tabernaemontana coronariae* and *Asparagus racemosus*. The concentration of lead in *Mimosa pudica* was below the detectable level. The maximum concentration of zinc was detected in *Adhatoda vasica*

followed by *Leucas aspera*, *Asparagus racemosus*, *Tabernaemontana coronariae*, *Alstonia scholaris* and *Mimosa pudica*. The concentration of cadmium, nickel, and chromium was below the detectable level.

The *Leucas aspera* has maximum concentration of iron (8.206 mg/g) followed by *Tabernaemontana coronariae* 96.320 mg /g), *Mimosa pudica* (6.458 mg/g), *Asparagus racehorses* (6.680 mg/g), *Alstonia scholaris* (5.069 mg/g) and *Adhatoda vasica* (5.498 mg/g).

TABLE 1: CONCENTRATION OF METAL IONS IN DIFFERENT PLANT RESIDUES (mg/g).

Name of the plant	Iron	Lead	Copper	Nickel	Cadmium	Chromium	Zinc
Adhatoda vasica	5.498	0.412	0.100	BDL	BDL	BDL	1.850
Leucas aspera	8.206	0.412	0.129	BDL	BDL	BDL	1.793
Mimosa pudica	6.458	BDL	0.090	BDL	BDL	BDL	0.947
Asparagus racemosus	6.690	0.079	0.085	BDL	BDL	BDL	1.570
Tabernaemontana coronariae	6.320	0.301	0.020	BDL	BDL	BDL	1.544
Alstonia scholaris	5.069	0.329	BDL	BDL	BDL	BDL	1.157

Abbreviation: BDL, below detectable level

DISCUSSION: In the present study iron, copper and zinc have been detected in the plant residues. However, No recent systematic investigations are available about the prevalence of heavy metal content of traditional Indian remedies on sale in developed countries. Thus, a considerable degree of uncertainty continues to surround this area. Obviously, heavy metals are not the only possible toxic ingredients in herbal remedies; contamination with herbicides, pesticides, micro-organisms or mycotoxins, insects or undeclared herbal constituents are other relevant possibilities [9, 10, 11, 12].

In human body iron is present in all cells and has several vital functions as a carrier of oxygen to the tissues from the lungs in the form of hemoglobin, as a transport medium for electrons with in the cell in the form of cytochromes, and as integral part of enzyme reactions in various tissues. Iron deficiency can delay normal infant motor function or mental function. Iron deficiency anemia during pregnancy can increase risk for small or early babies. The *Leucas aspera* has maximum concentration of iron (8.206 mg/g).

Copper is required for normal artery structure. Copper deficiency leads to weak aortic walls and rupture of the aorta. In the present study maximum concentration of copper was found in *Leucas aspera* (0.129 mg/g) followed by *Adhatoda vasica* (0.100 mg/g), *Mimosa pudica* (0.090 mg/g), *Asperagus racemosus* (0.085 mg/g) and *Tabernaemontana coronariae* (0.020 mg/g).

CONCLUSION: The analysis of metal ions was done by digesting known volume of the plant residue in concentrated sulphuric acid and making the volume up to 50 ml. Standard solutions were also prepared. Atomic absorption spectrometer was calibrated using different concentrations of the standard solutions. Then unknown solution was scanned. The plant residues showed the presence of iron, zinc, copper and lead.

REFERENCE:

1. Van Der Heijden, R, Threlfall, D.R, Verpoorte, R and Whitehead I.M. Studies on the regulation and enzymology of pentacyclic triterpenoid phytoalexin biosynthesis in cell suspension cultures of *Tabernammontana divaricata*. *Phytochemistry* 1989; 28: 2981 – 8.
2. Konofal, E, M. Lecendreux, I, Amulf and M.C, Mouren. Iron deficiency in children with attention-deficit/hyperactivity disorder. *Arch*. *Pediatr*. *Adolesc Med* 2004; 158: 1113-1115.
3. Koqak S, Tokusoglu O and Aycan S. Some heavy metal and trace essential element detection in canned vegetable food stuffs by differential pulse polarography (DPP). *Electronic J. Environ. Agric. Foodchem* 2005; 4:871-878.
4. Underwood E.J. Trace Elements in Human and Animal Nutrition, Edition 4, Academic Press, New York, 1977: 543.
5. Prasad A.S. Essential and Toxic Elements in Human Health and Disease: An Update, Wiley-Liss, New York 1993.
6. Fredrickson C.J. Detection of pathological zinc accumulation in neurons. *Int Rev. Neurobiol*. 1989: 31: 145 – 238.
7. Colin W Wright, David Allen, J, David Phillipson, Geoffrey C, Kirby, David, C, Warhurst, Georges Massiol and Louisette Le Menolvivie. *Alstonia* species: are they effective in malaria treatment? *Journal of Ethnopharmacology* 1993; 40 (1): 41 – 45.
8. Zerdahelyi P.S and Kasa P. Histochemistry of zinc and copper. *International Reviews on Cytology* 1984; 89: 1 -33.

9. Ernst E. Risks associated with complementary therapies. In: M.N.G. Dukes and J.K. Aronson, Editors, *Meyler's Side Effects of Drugs* (14), Elsevier 2000: 1649–1681.
10. Slifman N.R, Obermeyer W.R, Aloi B.K, Musser S.M, Correl W.A, Cichowicz S.M, Betz J.M and Love L.A. Contamination of botanical dietary supplements by *Digitalis lanata*. *New Engl. J. Med* 1998; 339: 806–810.
11. Halt M. Moulds and mycotoxins in herb tea and medicinal plants. *Eur. J. Epidemiol* 1998; 14: 269–274.
12. Lins Brandao M.G, Noélia Freire, Vianna-Soares C.D. Surveillance of phytotherapeutic drugs in the State of Minas Gerais. Quality assessment of commercial samples of chamomile. *Cad Saúde Pública, Rio de Janeiro* 1998; 14, 613–616.

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