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**RESEARCH ARTICLE**

# Quantitative Analysis of Minerals by ICP-MS and Flame Photometer in Herbal Tablets

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## Abstract

Use of herbal medicines is growing worldwide because of their minimal side effects and easily available in different forms. Herbal medicines and herbal formulation requires implementation and good manufacturing practices and effective quality control methods. In addition, various parameters such as dosage, stability, toxicity, chemical factors such as pesticides residue, aflatoxin content and heavy metals contamination should be taken in to consideration. Minerals are naturally occurring elements present in herbal medicines. The daily requirement of dietary minerals is in micrograms, but there is no proper method to detect the accurate concentration of minerals present in herbal medicines. Diseases occur due to their deficiency and toxicity due to overdose. These are categorized as essential, probably essential and non-essential. These minerals can bind to vital cellular components and interfere with their normal functions. In human being, these minerals can cause severe physiological and health effects. Hence in the present study, some important marketed herbal medicines are scanned. Herbal medicines contain many minerals like Ca (Calcium), Na (Sodium), K (Potassium), etc. In the present study, these minerals are determined quantitatively by using modern technique ICP-MS and Flame photometer which should be incorporated in routine quality control parameters.

**Keywords:** Herbal medicines, mineral analysis, inductively coupled plasma mass spectrometry, flame photometer.

## Introduction

The use of herbs as medicines in *Ayurveda* is the oldest science of health and healing practiced by ancient Aryans which is based on Atharvaveda, one of the oldest scriptures of Hindus about 6000 years old. The object of Ayurveda is to counteract the imbalance of three essential elements air, bile and phlegm. It is one of the most trusted systems of medicine in the world in which Vaidya's (Physician) used to select medicinal plants personally and were giving utmost thought to purity of the medicines. According to hypothesis of Ayurveda, the entire universe is composed of five basic elements called "Panchtatva" (five principals) and includes akasha, vayu, teja, jal and prithvi. Over the years, herbal products become commercial and many pharmaceutical companies started manufacturing different herbal medicines on large scale at the cost of inadequate quality control. In the preparation of herbal medicines, various parts of the plant such as roots, stem, bark, leaves, flowers, seeds, buds, fruits etc. in dry form or in original form are used as a raw material as single or in combination. After passing through many processes like collection, drying, powdering, extraction, separation, purification, clinical screening, formulation and clinical trials, they are converted into finished herbal medicines (Kamat and Suryawanshi, 2015). World Health Organisation states that around 85-95% of the world population uses traditional herbal medicines.

Most of the people use herbal medicines which are now available in different forms like tablets, elixirs, powders and bhasma (asses) (Lozak *et al.,* 2012). Herbal medicines have become more popular as alternative and supplementary medicines in recent years. Concentration of Calcium, Sodium, potassium etc., is of major concern because of their effects on kidney and liver function and BP is of utmost important and affects Glomerular Filtration Rate. However, some herbal medicines do contain minerals as essential ingredients. The poor quality control of these medicines causes health hazards. Some medicines may present unusually high concentrations of minerals that could lead to fatality if consumed for a longer time. Therefore, it is thought necessary to study the levels of minerals being consumed by patient per tablet so that their repercussions can be evaluated. Patients are not aware about their contents and standards. World Health Organisation gives some guidelines (WHO, 2007) for the preparation of herbal medicines and listed some methods for the standardization of herbal medicines (WHO, 2011) and also gives maximum permissible limit of heavy metals (The Merck Index, 1989) and quality controlled norms however, there is no mention regarding minerals in herbal medicines. Therefore, it is imperative to label mineral quantities on these medicines.

Table 1. Tablet name with the company name and plants as per label.

| Code | Brand and company name | Product name | Plants as per label* |
|---|---|---|---|
| A | Baidyanath (Mfg. Lic. No. ND/AYU/4) | Sarpagandha | Sarpagandha powder |
| B | Safe life (Mfg. Lic. No. NKD/AYU 82) | Cardiol vati | Suthi, Arjun ghan, Punarnava, Bringrajn, Abhrak bhasma, shuddha shiljit, Amalki ghan, Guduch ghan, Gokshur ghan, Akik pisti. |
| C | Safe life (Mfg. Lic. No. NKD/AYU 82) | Hemiplus vati | Amalaki, Haritaki, Bibhitaki, Sunthi, Pipali, marich, Vidang, Suvarna makshik bhasma, kasis bhasma. |
| D | Safe life (Mfg. Lic. No. NKD/AYU 82) | Medomine vati | Pipali, Marich, Amalki ghan, Haritaki ghan, Bibhitaki ghan, Trmad churn, Loha bhasma, Shuddha shilajit, Kitatika, Guduchi, Gugul, Sunthi. |
| E | Safe life (Mfg. Lic. No. NKD/AYU 82) | Arthowin vati | Rasna mool, Sunthi, Gokshur, Erand mool, Ashwagandha, Guggul, Guduchi. |
| F | Peekay pharma (Mfg. Lic. No. 25D/10/88) | B.P.C capsule | Sarpgandha, Lahasun, Arjun chhal Ex, Guggul Ashwag Jatamansi, Naandha, Isabgol, Brahmi, Jatamansi, Nagarmotha, Shankpushi, Kapoor kachri, Badi ilaichi. |

*Data collected from the labelled contain with the tablets.

Table 2. Sample weight and dilution.

| Samples | Weight (g) | Dilution |
|---|---|---|
| Sarpagandha | 0.37287 | 100 mL in 1% $HNO_3$ |
| Cardiol vati | 0.45548 | 100 mL in 1% $HNO_3$ |
| Hemiplus vati | 0.2527 | 100 mL in 1% $HNO_3$ |
| Medomin vati | 0.47809 | 100 mL in 1% $HNO_3$ |
| Arthowin vati | 0.41698 | 100 mL in 1% $HNO_3$ |
| B.P.C capsule | 0.11798 | 100 mL in 1% $HNO_3$ |

Table 3. Multi-Elemental standards and mercury analysis.

| Concentration | Yttrium (1 ppm) | MES | MES + Hg (20 ppb) | Final volume (mL) |
|---|---|---|---|---|
| Std .05 ppb | 750 µL | - | 75 µL | 30 |
| Std 0.5 ppb | 750 µL | - | 750 µL | 30 |
| Std 1.0 ppb | 750 µL | - | 1500 µL | 30 |
| Std 2.0 ppb | 750 µL | - | 3000 µL | 30 |
| Std 5.0 ppb | 750 µL | 150 µL | - | 30 |
| Std 20 ppb | 750 µL | 600 µL | - | 30 |
| Std 50 ppb | 750 µL | 1500 µL | - | 30 |
| Std 100 ppb | 750 µL | 3000 µL | - | 30 |
| Std 200 ppb | 750 µL | 6000 µL | - | 30 |

Various instrumental methods like XRPD (Sunil, 2009-10) and ICP-MS was carried out (Kamat and Suryawanshi, 2015). Standardization of herbal medicines maintained the quality and contains well defined constituents required for reliable beneficial therapeutic effects. Most of the herbal medicinal medicines are not labeled appropriately in their contents. Keeping above points in view, the determination of various minerals in the herbal medicines were done by ICP-MS methods which has high degree of sensitivity and specificity and flame photometer which is very simple, inexpensive and less time consuming.

## Materials and methods

*Chemicals:* Yttrium as internal standard, deionized water solution of 0.5% nitric acid and 2 ppm gold (Thermo- fisher ICP-MS icap model).

*Sampling:* In the present study, the marketed herbal tablets Sarpagandha, Cardiol vati, Hemiplus vati, Medomine vati, Arthowin vati and B.P.C capsules are selected for the analysis. The brand names of the products, license number and the plants used as per company's label are included (Table 1).

*Experimental design:* Code numbers namely A to F was assigned for Sarpagandha, Cardiol vati, Hemiplus vati, Medomine vati, Arthowin vati and B.P.C capsules. By taking the weight of each tablet on digital balance, each tablet sample was gently ground to fine powder using mortar and pestle and packed in butter paper until analysis. The dilution is given in Table 2. Quantitative multi-elemental analysis by inductively coupled plasma (ICP) [Icap-Q] mass spectrometry depends on complete digestion of solid samples.

Table 4. Sample weight and dilution for flame photometer.

| Samples | Weight (g) | Dilution | Final volume |
|---|---|---|---|
| Sarpagandha | 0.336 | 25 mL in 1% $H_2SO_4$ | 1 mL in 25 mL de-ionized $H_2O$ |
| Cardiol vati | 0.443 | 25 mL in 1% $H_2SO_4$ | 1 mL in 25 mL de-ionized $H_2O$ |
| Hemiplus vati | 0.228 | 25 mL in 1% $H_2SO_4$ | 1 mL in 25 mL de-ionized $H_2O$ |
| Medomin vati | 0.484 | 25 mL in 1% $H_2SO_4$ | 1 mL in 25 mL de-ionized $H_2O$ |
| Arthowin vati | 0.409 | 25 mL in 1% $H_2SO_4$ | 1 mL in 25 mL de-ionized $H_2O$ |
| B.P.C capsule | 0.468 | 25 mL in 1% $H_2SO_4$ | 1 mL in 25 mL de-ionized $H_2O$ |

Table 5. Accuracy of mineral concentrations in ppm and percentage by ICP-MS and Flame photometer.

| Samples | Estimated by ICP-MS | | Estimated by Flame photometer | | | |
|---|---|---|---|---|---|---|
| | Calcium (ca) | | Sodium (Na) | | Potassium (K) | |
| | in ppm | % by weight | in ppm | % by weight | in ppm | % by weight |
| Sarpagandha | 0.00132 | 0.000000132 | 2.3 | 0.00023 | 4 | 0.0004 |
| Cardiol vati | 0.0019 | 0.00000019 | 2.9 | 0.00029 | 5.6 | 0.00056 |
| Hemiplus vati | 0.00141 | 0.000000141 | 2.4 | 0.00024 | 2.6 | 0.00026 |
| Medomin vati | 0.00223 | 0.000000223 | 2 | 0.0002 | 4.6 | 0.00046 |
| Arthowin vati | 0.00135 | 0.000000135 | 2.3 | 0.00023 | 4.5 | 0.00045 |
| B.P.C capsule | 0.00554 | 0.000000554 | 2.8 | 0.00028 | 7 | 0.0007 |

Table 6. LD50 of the elements (The Merck Index, 1989).

| Elements | Compounds | LD 50 |
|---|---|---|
| Calcium (Ca) | Calcium acetate | 04.28 g/kg orally on rat |
| Sodium (Na) | Sodium arsenate dibasic | 14-18 mg As/kg in rat |
| | Sodium chloride | 3.75 g/kg orally in rat |
| | Sodium borate | 5.66 g/kg orally in rat |
| | Sodium benzoate | 4.07 g/kg orally in rat |
| Potassium (K) | Potassium carbonate | 1.87 g/kg orally in rat |
| | Potassium arsenite | 14 mg/kg orally in rat |

However, fast and thorough sample digestion is a challenging analytical task in modern multi-elemental analysis. To determine each elemental concentration as minerals, 0.125 mL internal standard and 4.675 mL of diluent was added in to 0.2 mL sample solution. Deionzed water solution of 0.5% nitric acid and 2 ppm gold was used as a diluent (Table 3). Elico Flame Photometer CL 378 was also used for the analysis of samples. Analysis made much simpler because of the multiple calibration curves saving facility. The instrument carries advanced features like automatic ignition, automatic gas shut off in case of a power failure. The dilution for FP is given in Table 4.

*Statistical analysis:* The obtained values were properly validated with standard deviation, standard error and coefficient variance. In addition to normal validation parameters, average weight equal to each tablet is analyzed from the crushed powder (3/4/5/6 tablets) as additional validation.

## Results and discussion

Research on mineral concentrations requires analytical techniques that are versatile, robust, highest sensitivity and capable of providing accurate and reliable information on concentrations and species identity.

With respect to most of these criteria, determination of minerals by ICP-MS is performing extremely well and is unchallenged by other MS techniques. On the other hand, it is a unique and outstanding advantage of ICP-MS to use inexpensive, unspecific, certified element standards, allowing a quantitative control on elemental losses, species decomposition or contamination in each single step of an experiment. Scanty work is reported regarding minerals even in human system and therefore their role is not yet clear in biochemical reactions. Even in Ayurvedic medicines, the minerals, trace elements, toxic elements, microelements are not reported with the daily intake of tablet, it is essential to know the amount of individual minerals consumed by patient. Results obtained from ICP-MS analysis of tablet samples detected the accurate concentration of calcium in ppm and result obtained from flame photometer of tablet sample detected the accurate concentration of sodium, and potassium. The detected accuracy of mineral concentration in selected samples by ICP-MS and flame photometer is given in Table 5.

*Sarpagandha:* In Sarpagandha sample, most abundant element was K-4.0ppm, whereas, Ca was found in lowest concentration (0.00132 ppm).

*Cardiol vati:* In Cardiol vati sample, most abundant element was K-5.60 ppm, whereas Ca was found in lowest concentration (0.0019 ppm).

*Hemiplus vati:* In Hemiplus vati sample, most abundant element was K-2.60 ppm whereas Ca was found in lowest concentration (0.00141 ppm).

*Medomine vati:* In Medomine vati sample, most abundant element was K-4.60 ppm whereas, Ca was found in lowest concentration (0.00223 ppm).

*Arthowin vati:* In Arthowin vati sample, most abundant element was K-4.50 ppm whereas Ca was found in lowest concentration (0.00135 ppm).

*BPC capsules:* In BPC capsule samples, most abundant element was K-7.0 ppm whereas, Ca was found in lowest concentration (0.00554 ppm).

Recently published FDA regulations hold supplement manufacturers or distributors responsible for the content of the dietary supplement which should only contain what they are labeled and not any harmful or undesirable substances, including pesticides and heavy metals (Avula, 2010). All these concentration of calcium, sodium and potassium showed less level in herbal medicines and detected below LD50 (Table 6).

## Conclusion

The content of calcium, sodium and potassium elements is not indicated on their label. Elemental analysis by ICP-MS is a recent technique, Detection limit of ICP-MS is Excellent for most elements and Analytical capability is Multi-element, which gives more accurate concentration of minerals contain in the Ayurvedic medicinal tablet which is not previously reported by researchers. Quantitative estimation of metals is done by atomic absorption spectrophotometer in bhasma only, not in tablets, therefore, the concentration of calcium, sodium and potassium elements are found below the hazardous levels. None of the tablets studied showed limits hazardous to human beings. Calcium, sodium and potassium concentration levels in the tablets showed that it will not pose any problem to patients having high BP, Cardinals and Kidney ailments.

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