Elemental Analysis of *Paeonia emodi* and *Punica granatum* by Atomic Absorption Spectroscopy

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Abstract During present study *Paeonia emodi* and *Punica granatum* were selected for elemental analysis due to its wide application in the indigenous medicinal system and the importance of its chemical constituents. The plant samples were digested with concentrated Nitric acid (HNO₃), followed by concentrated Perchloric acid (HClO₄). The transparent solutions of these samples were analysed by Atomic Absorption Spectrophotometer. The analysis for various elements in the sampled medicinal plants indicated that Sodium, Potassium, Calcium, Magnesium, Copper, Zinc, Iron, Cobalt, Manganese, and Lead were present in all samples of both plant species in different concentration. In Paeonia emodi Calcium is found in high concentration (66.26 mg/L) while Cobalt is in low concentration (0.039 mg/L). Similarly in Punica granatum Potassium concentration is maximum (55.19 mg/L) while Cobalt concentration is minimum (0.031 mg/L).

Keywords Medicinal Plants, Elements, Constituents, Atomic Absorption, Battagram

1. Introduction

Ethnobotanical information on medicinal plants and their uses by indigenous cultures is useful in the conservation of traditional cultures, biodiversity, for community health care and drug development[1]. The authentic knowledge of the uses of medicinal plants passed on from one generation to another, after refining and additions[2]. The folk recipes are prepared either from the whole plant or from their different organs, like leaves, stem, bark, root, flower, seed, prop roots etc and also from their secondary product such as gum, resins and latex[3]. Plant based recipes have been in use in the amelioration of various ailments ranging from common cold to cancer[4]. Plants supply the body with minerals, vitamins and certain hormone precursors in addition to protein and energy[5,6]. Relatively high levels of essential elements, such as Fe, Mn, Zn, and Ca, have been demonstrated to influence the retention of toxic elements in animals and human beings [7,8].

In recognition of the important role that major and trace elements play in health and disease of human body, in the building up and restoration phenomenon, progress has occurred in this area of health sciences during the last few years[3].

Elements research has definitely been part of this explosion of scientific knowledge[9]. Trace elements are required

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Published online at http://journal.sapub.org/ajb

in very small quantities for plant life[10]. The quantitative estimation of various element concentrations is important for determining the effectiveness of the medicinal plants in treating various diseases and also to understand their pharmacological action[4]. The imbalance in human health has been linked with the excess or deficiency of elements in soils, water, plants and animals[3]. The continuous intake of diets that are excessively high in a particular element can influence changes in the functioning, forms, activities of some organs or concentrations of such element in the body tissue and fluids can rise above the permissible limit[11].

District Battagram is located in the Western Himalayan Province between 34°25′ and 34°58'N and 72°54′ and 73°15'E with a total area of 1,301 km²[12,13]. District Battagram is bounded by the Kohistan valley in the north, by Siran valley in the east, by the Konsh and Agror valleys in the south and by the Black mountain and river Indus in the west[2]. The district consists of two sub-divisions i.e. Battagram and Allai. District Battagram can be categories into agricultural land, wasteland, forest and alpine meadows[14,15].

District Battagram is mostly mountainous ranging in altitude from 525 m at Thakot to 4690 m above mean sea level at Sukaisar. The area is generally rough and mountainous having variable slopes from gentle to precipitous. The climates varying from sub tropical at the base of the hills to "alpine" conditions prevailing in the higher reaches. Majority of the people are dependent on agriculture as a first source of livelihood, followed by pasture animal husbandry. They grow rice, maize, wheat, red beans and vegetables. All small streams coming from the sub valleys join two main streams

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Nandiar Khuwar and Allai Khuwar in District Battagram to join River Indus[2,13,15,16].

The diversity and distribution of Paeonia emodi and Punica granatum were discussed in District Battagram[13], their traditional medicinal uses[2] and conservation status were also discussed[17]. The comparative determination of trace elements from Allium sativum, Rheum australe and Terminalia chebula were also recently analysed[5]. The Physical and Chemical Properties of *Punica granatum*[18] and the chemical constituents of Paeonia emodi[19] were also studied. Keeping in mind the wide application Paeonia emodi and Punica granatum in traditional medicine were therefore analysed for further elemental contents.

2. Materials and Methods

The field survey was conducted during the year 2010 to collect the information and specimens of Paeonia emodi and Punica granatum from different parts of District Battagram for their element analysis. The fresh root of Paeonia emodi and fruit of Punica granatum were cleaned and oven dried to a constant weight at 60°C for 72 h. After drying, the plant materials were ground into fine powder using an electric grinder. The powder of both plant species were weighted 0.5g separately and put in a separate 100ml conical flasks and 30ml nitric acid (HNO₃) were added. Each flask were placed on magnetic stirrer heater in fume hood for four hours at 250C° and then colour solution were changed to milky solutions which were cooled for 10 minutes and then 15ml concentrated Perchloric acid (HClO₄) were added and heat the solutions until colourless solutions were obtained. The colourless solutions were filtered to remove the impurities for trace elements analysis by Atomic Absorption Spectroscopy.

3. Results

3.1. 1. Paeonia emodi Wall. ex Hook. f.

Paeonia emodi locally known as Mamekh belongs to family Paeoniaceae.

3.1.2. Description

Perennial herbs, to 70 cm tall. Stems glabrous. Proximal leaves 2-ternate; some leaflets segmented; leaflets and segments up to 15, oblong-elliptic or oblong-lanceolate, 9--13 \times 2-3.5 cm, both surfaces glabrous, base cuneate, decurrent, apex acuminate. Flowers 2--4 per shoot, both terminal and axillary, single, 8--12 cm wide, all or only terminal one fully developed. Bracts 3--6, leaf like, lanceolate. Sepals ca. 3, suborbicular, ca. 1.5 \times 1.5 cm, apex caudate. Petals white, obovate, ca. 4.5 \times 2.4 cm. Filaments 1.5--2 cm. Disc annular. Carpel 1(or 2), pale yellow to mentose, rarely glabrous. Follicles ovoid, 2--3.5 \times 1--2 cm. Seeds black, globose. Fl. May-Jun.

3.1.3.Traditional uses

The infusion of dried flower is used in diarrhea. Rhizome is used to increase milk production in livestock, also used as tonic.

3.2.1. Punica granatum Linn

Punica granatum commonly known as Pomegranate and is locally called as Narsaway belongs to family Punicaceae.

3.2.2. Description

Deciduous shrub to small tree up to 6 m high, with thin smooth grey bark. Leaves are mostly opposite, short-petiolate, blades oblong-elliptical, up to 8 cm long. Flowers showy and up to 6 cm broad, bisexual, 5-8 petals, reddish and up to 2.5 cm long, numerous stamens surrounding a conspicuous hypanthial tube, he flowers usually occuring terminally or in axils. Fruit a red spherical berry up to 13 cm broad, with a leathery rind enclosing numerous seeds surrounded by edible juicy, tart, bright red "kernels". Fl. Apr.-May.

3.2.3. Chemical Constituents

Apigenin glucoside, betulinic acid, callistephin, tannins, chrysanthemin, coniine, coumestrol, cyanidin and its diglucoside, cyanin, daid zein, daid zin, delphin, delphinid in and its glucosides, ellagic acid and its derivatives, estrone, friedelin, gallic acid, genistein, genistin, lipids, hygrine and norhygrine, luteolin glycosides, mannitol, pelargonin, pelletierine and its derivatives, piperidine derivatives, polyphenols, sedridine, xanthoxylin, estradiol.

3.2.4. Traditional Uses

A decoction of seed is used to treat syphilis. Its juice of seeds is used to treat jaundice and diarrhea. Juice of the fruit is used to treat jaundice and diarrhoea. The rind of the fruit is ground in water and drunk every morning by diabetics. The fruit together with the juice of Cynodon dactylon leaves is used for runny noses and colds. The juice of the flowers is used to treat nose bleeds. The fruit pulp and the seed are a stomachic. The root and stem bark have astringent and anthelmintic properties.

3.3. Elemental Analysis

The analysis for various elements in the sampled medicinal plants indicated that Sodium, Potassium, Calcium, Magnesium, Copper, Zinc, Iron, Cobalt, Manganese, and Lead were present in all samples of both plant species which are responsible for curing various diseases. The plant samples were analysed by Atomic Absorption Spectroscopy. These elements play a vital role in the formation of secondary metabolites which are responsible for pharmacological actions of medicinal plants. The average comparative concentration of these elements in both *Paeonia emodi* and *Punica granatum* are presented in table 1 along with standard deviation.

3.3.1. Elemental analysis of Paeonia emodi

The elemental analysis of *Paeonia emodi* through Atomic Absorption Spectroscopy shows different concentration of various elements such as Sodium (1.235 mg/L), Potassium (33.38 mg/L), Calcium (66.26 mg/L), Magnesium (9.525 mg/L), Copper (0.329 mg/L), Zinc (0.956 mg/L), Iron (2.667 mg/L), Cobalt (0.039 mg/L), Manganese (0.082 mg/L), and Lead (0.735 mg/L). These results show that the concentration of Calcium is maximum and Cobalt concentration is minimum in sampled *Paeonia emodi* plant species.

3.3.2. Elemental analysis of Punica granatum

The analysis for various elements in the sampled *Punica* granatum indicated that the concentration of Sodium (1.279 mg/L), Potassium (55.19 mg/L), Calcium (1.650 mg/L), Magnesium (3.721 mg/L), Copper (0.231 mg/L), Zinc (0.430 mg/L), Iron (0.466 mg/L), Cobalt (0.031 mg/L), Manganese (0.033 mg/L), and Lead (1.119 mg/L) were present in all samples of both plant species which are responsible for curing various diseases.

In present study it was observed that the concentration of Potassium is maximum in *Punica granatum* (55.19 mg/L) and the concentration of Cobalt is minimum (0.031 mg/L).

Table 1. Concentration of different elements in mg/L of Paeonia emodi and Punica granatum

S. No	Elements	Paeonia emodi (Conc.)	Punica granatum (conc.)
1	Sodium (Na)	1.235 mg/L ± 0.001	1.279 mg/L ± 0.003
2	Potassium (K)	33.38 mg/L ± 0.011	55.19 mg/L ± 0.021
3	Calcium (Ca)	66.26 mg/L ± 0.00	1.650 mg/L ± 0.00
4	Magnesium (Mg)	9.525 mg/L ± 0.002	3.721 mg/L ± 0.001
5	Copper (Cu)	0.329 mg/L ± 0.001	0.231 mg/L ± 0.003
6	Zinc (Zn)	0.956 mg/L ± 0.005	0.430 mg/L ± 0.001
7	Iron (Fe)	2.667 mg/L ± 0.002	0.466 mg/L ± 0.003
8	Cobalt (Co)	0.039 mg/L ± 0.003	0.031 mg/L ± 0.001
9	Manganese (Mn)	0.082 mg/L ± 0.001	0.033 mg/L ± 0.002
10	Lead (Pb)	0.735 mg/L ± 0.003	1.119 mg/L ± 0.002

± Standard deviation

4. Discussion

The elements play both curative and preventive role in combating diseases[5]. There is a vast scope to exploit the preventive medicinal aspects of various elements[20].

Sodium (Na) involves in the production of energy, transport of amino acids and glucose into the body cells, the average concentration Na was 1.235mg/L in Paeonia emodi and 1.279mg/L in Punica granatum.

Potassium (K) is helpful in reducing hypertension and maintaining cardiac rhythm. In the human body, Potassium play vital role in many physiological reactions and their deficiency or excess can affect human health[21]. The average concentration of Potassium was 33.38mg/L in Paeonia emodi and 55.19mg/L in Punica granatum.

Calcium (Ca) overcome the problems of high blood pressure, heart attack, premenstrual syndrome, colon cancer and keeping the bones strong and reduces the risks of osteoporosis in old age[3]. The average concentration Ca was 66.26 mg/L in Paeonia emodi and 1.650mg/L in Punica granatum.

Magnesium (Mg) improves insulin sensitivity, protect against diabetes and its complications and reduce blood pressure[5]. The average concentration Mg was 9.525mg/L in Paeonia emodi and 3.721mg/L in Punica granatum.

Copper (Cu) play important role in treatment of chest wounds and prevent inflammation in arthritis and similar diseases. The average concentration Cu was 0.329mg/L in Paeonia emodi and 0.231mg/L in Punica granatum.

Zinc (Zn) deficiency may contribute to arrested sexual maturation, growth retardation and hair loss, delayed wound healing and emotional disturbance[3]. The average concentration of Zn was 0.956 mg/L in Paeonia emodi and 0.430 mg/L in Punica granatum.

Iron (Fe) is an essential mineral to prevent anaemia and cough associated with angiotensin-converting enzyme (ACE) inhibitors. The average concentration Fe was 2.667mg/L in Paeonia emodi and 0.466mg/L in Punica granatum.

Cobalt (Co) is necessary in very small amounts in all mammals and is used to treat several different types of cancer in humans and treat anaemia but the intake of high amount can cause heart diseases. The average concentration Co was 0.039mg/L in Paeonia emodi and 0.031mg/L in Punica granatum.

Manganese (Mn) can help to assist the body in metabolizing protein, help the diabetic also metabolize carbohydrates and in treating diabetes. The average concentration Mn was 0.082mg/L in Paeonia emodi and 0.033mg/L in Punica granatum.

Lead (Pb) is toxic metal and non-essential element for human body as it causes a rise in blood pressure, kidney damage, miscarriages and subtle abortion, brain damage, declined fertility of men through sperm damage, diminished learning abilities of children and disruption of nervous systems[3,11]. The average concentration Pb was 0.735mg/L in Paeonia emodi and 1.119mg/L in Punica granatum.

The environmental factors including atmosphere and pollution, season of collection sample, age of plant and soil conditions in which plant grows effect the concentration of elements as it varies from plant to plant and region to region[5].

5. Conclusions

In this study 10 elements were determined in Paeonia emodi and Punica granatum. A mong the various elements detected in different medicinal plants in different concentration is there fore used in the treatment of various diseases. The data obtained in present study will be helpful in the synthesis of new modern drugs with various combinations of plants which can be used in the cure of many diseases ethno medicinally. However, more detailed analysis of chemical composition of these medicinal plants is required.

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