Short Communication

Heavy and essential metals contents of Artemisia annua L. and Pyrus pashia Buch. Ham

J. S. Negi, V. K. Bisht, A. K. Bhandari and R. C. Sundriyal*

Herbal Research and Development Institute, Mandal, Gopeshwar (Chamoli) - 246 401, Uttarakhand, India.

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Artemisia annua and Pyrus pashia are used to relieve and treat many human diseases due to their low side effects. They are rich source of biologically important elements, which may play an important role in the observed therapeutic uses of these plants. It is important to have a good quality of medicinal plants in order to protect consumers from contamination. Therefore, the aim of the present study was to carry out a comparative evaluation of heavy and essential metals contents in *A. annua* and *P. pashia*. The heavy and essential metals (As, Cd, Zn, Ni, Pb and Hg) contents ranged between $1.82\pm0.54 - 1.86\pm0.22$, $0.30\pm0.07 - 0.36\pm0.15$, $18.00\pm3.32 - 27.00\pm2.53$, $1.22\pm0.44 - 3.02\pm0.55$, $1.24\pm0.54 - 1.84\pm0.42$ and $1.60\pm0.84 - 1.92\pm0.35$ mg/kg respectively in *A. annua*, while for *P. pashia* these varies from $2.65\pm1.83-2.81\pm0.25$, $0.02\pm0.06 - 0.05\pm0.25$, $13.50\pm3.13 - 25.20\pm1.18$, $0.21\pm0.82 - 2.42\pm0.24$, $0.38\pm0.03 - 1.05\pm0.08$ and $1.48\pm0.78 - 1.82\pm0.46$ mg/kg respectively. The highest Cd, Zn, Ni, Pb and Hg content was detected in roots of *A. annua*, while highest concentration of As was found in leaves of *P. pashia*. The contents were well within the permissible limits therefore, it can be used in the cure system.

Key words: Heavy metals, essential elements, ICP-MS, Microwave digestion system.

INTRODUCTION

Traditional medicinal plants produce and their extracts play an important role in human community. Artemisia annua (Asteraceae) is one of the common medicinal plants used for antipyretic, skin allergy, insecticide and as tonic, likewise, ripe fruits of Pyrus pashia (Rosaceae) used in digestive disorders (Gaur, 1999). Both species are rich source of many essential nutrient elements and phytochemicals in bio-available form (Park et al., 2007; Khandelwal et al., 2008; Brisibe et 2009: Ajah and Eteng, 2010: al., http://www.hort.purdue.edu/newcrop/parmar/20.html;

Ashraf et al., 2010). Mineral elements play an important role in humans as well as, plant physiology but these elements particularly, heavy metals when present in excess amount causes toxicity (Schumacher et al., 1991; Nurul et al., 2011). The heavy metal contents in medicinal plant products are studied particularly in view point of toxicity and bioavailability. Heavy metals such as arsenic (As), cadmium (Cd), lead (Pb) and mercury (Hg) may be introduced into medicinal plants in a variety of ways including contamination during cultivation, processing, and storage (Bober and Chen, 2005). The investigation of heavy metals in herbal products is widely fascinated. Inductively Coupled Plasma Mass Spectrometry (ICP-MS) has been proved to be the technique of choice to analyze herbal medicines for metals (Krachler et al., 1996).

High contents of As, Cd, Pb and Hg in plants may also be accredited to the uptake of these elements from polluted soil primarily due to anthropogenic activities. In general, all medicinal products for human and animal use must meet regulatory guidelines for their quality, safety and efficacy. It is a major interest to establish the level of these heavy metallic elements in medicinal plants produces, because at high level, these metals act as toxicant and may cause several physiological disorders (Schumacher et al., 1991). According to Hussain et al. (2011), eighty one elements are present in living organism; their excess or deficiency affects at least 235 diseases or functions of the body. Thus, determination of

^{*}Corresponding author. E-mail: negijs@yahoo.com, sundriyalrc@yahoo.com. Tel: +919997239903. Fax: +91-1372-254273.

Sample	Plant part	Contents mg/kg					
		As	Cd	Zn	Ni	Pb	Hg
A. annua	Leaves	1.82±0.54	0.30±0.07	18.00±3.32	1.22±0.44	1.24±0.54	1.60±0.84
	Roots	1.86±0.22	0.36±0.15	27.00±2.53	3.02±0.55	1.84±0.42	1.92±0.35
P. pashia	Fruits	2.65±1.83	0.02±0.06	13.5±3.13	0.21±0.82	0.38±0.03	1.48±0.78
	Leaves	2.81±0.25	0.05±0.25	25.2±1.18	2.42±0.24	1.05±0.08	1.82±0.46

Table 1. Concentration of heavy metals and essential elements (mg/kg).

heavy metals in medicinal plants will establish their purity, safety and efficacy as par with WHO recommendations (WHO, 1992). It is therefore, crucial to explore the present status of *A. annua* and *P. pashia* in terms of selected four heavy metals and two essential elements. These plants are commonly used by local people to prepare infusions for phytotherapeutical purposes. This will establish whether the use of *A. annua* and *P. pashia* is safe for the consumers or not according to the world health standards.

MATERIALS AND METHODS

Leaves and roots samples of A. annua (Cultivated) and leaves and fruits of P. pashia (wild) were collected from Herbal Garden of Herbal Research and Development Institute, Mandal, Gopeshwar, Uttarakhand, India. Each sample was collected from three places of the same population (nearly same maturity) for the estimation of heavy metals contents. Analytical grade concentrated HNO3, perchloric acid and Milli-Q water (Millipore, Bedford, USA) were used. Prior to analysis, samples were homogenized and prepared. The microwave digestion system (Aurora MW 800) was used for the decomposition and digestion of samples. 0.25 g of each sample was transferred to an acid washed Teflon PFA digestion tube separately. 8 ml of concentrated nitric acid (LR grade 72 to 90%) and perchloric acid was added (3:1), and the tube was heated in a microwave oven for about 25 min and then left for cooling for another 30 min. Vessels were opened in a fume cupboard and the digests were transferred into a 25 ml acid washed teflon tubes filled up with demineralized water (Millipore), filter and stored.

Preparation of standard solutions

Multi-element working standard solutions (1, 5, 10, 25 and 50 μ g/l) were prepared by appropriate dilution from the stock ICP multielement standard (10000 μ g/l; Merck, Germany). The calibration curves for each element were linear and the correlation coefficients ranged between 0.995 and 0.999.

Analysis

All analyses were performed on a Perkin Elmer NexION 300X ICP-MS using both Standard and Kinetic Energy Discrimination (KED) modes. The sample uptake rate was 0.5 ml/min; the flush delay, read delay and wash were 35, 90 and 40 s respectively. The sample run time was 3 min per sample. The plasma gas flow, auxiliary gas flow and nebulizer gas flow rate were 17.0, 3.0 and 0.98 L/min respectively. All samples were analyzed thrice.

RESULTS AND DISCUSSION

A total of six elements (As, Cd, Pb, Hg, Zn and Ni) were determined in leaves and roots of A. annua and fruits and leaves of P. pashia samples by ICP-MS. Table 1 shows the different concentration of selected elements in investigated plants. Each data represent means ± SD of three replicates of each sample. All results were calculated on a dry weight basis (mg/kg). From the study, it was revealed that all the metals were accumulated to different extents by A. annua and P. pashia. Levels of the essential metals in A. annua and P. pashia samples were found to be higher than those of the non-essential (heavy) metals. The concentrations of As, Cd, Zn, Ni, Pb and Hg in different samples of A. annua and P. pashia were found in the range of $1.82 \pm 0.54 + 2.81 \pm 0.25$, 0.02 ± 0.02 $0.06 - 0.36 \pm 0.15$, $13.5 \pm 3.13 - 27.00 \pm 2.53$, 0.21 ± 0.82 -3.02 ± 0.55 , $0.38 \pm 0.03 - 1.84 \pm 0.42$ and 1.48 ± 0.78 -1.92 ± 0.35 mg/kg, respectively. The lowest As content (1.82 ± 0.54 mg/kg) was found in leaves part of A. annua; however, it was found higher $(2.81 \pm 0.25 \text{ mg/kg})$ in leaves of P. pashia. The levels of Cd, Zn, Ni, Pb and Hg were found lowest in the fruits of *P. pashia* and highest in roots of A. annua. Highest Cd concentration was found 0.36 ± 0.15 in root samples of A. annua and leaves sample P. pashia (0.05 ± 0.25 mg/kg). The permissible limit of Cd for medicinal plants was recommended to be 0.3 ppm (WHO, 2005). After comparison of our findings, it was observed that most of the studied samples accumulate Cd below this limit except A. annua roots. The highest Zn content was found to be 27.00 ± 2.53 and 25.2 ± 1.18 mg/kg in root samples of A. annua and leaves samples of *P. pashia*. The permissible limit for Zn set by FAO/WHO (1984) for edible plants was 27.4 ppm and the content of Zn in the present study are within this limit. Therefore, the studied plants showed satisfactory level of Zn accumulation.

Highest concentration of Ni was found to be 3.02 ± 0.55 mg/kg in *A. annua* roots and 2.42 ± 0.24 mg/kg in *P. pashia* leaves. The permissible limit for this element set by FAO/WHO (1984) for edible plants was 1.63 ppm. After comparison, Ni limit in both studied plant samples was found above this limit. However, Ni toxicity in human is not very common because its absorption by the body is very low (Onianwa et al., 2000). Brisibe et al. (2009) reported 64.5, 46.2 and 75.9 ppm Zn in leaves, stems

and roots of *A. annua* while 52.8 mg/kg Zn and 3.60 mg/kg Ni were reported from leaves of *A. annua* by WEI Ji-qinetal.(http://en.cnki.com.cn/Article_en/CJFDTOTAL

AHNY200822104.htm). The permissible limit of Pb for medicinal plants was set at 10 ppm by FAO/WHO (1984). In the present study, highest concentration was found to be 1.84 \pm 0.42 mg/kg. The Pb content in the studied medicinal plants samples was found within this limit. The concentrations of Zn, Ni, Pb and Cd in *A. annua* have been reported in the range 32.38 \pm 0.49, 5.05 \pm 0.74, 3.95 \pm 2.63 and 0.98 \pm 0.84 ppm, respectively (Ashraf et al., 2010). The elemental studies of the plant samples showed that they contained large amounts of nutrients and are rich in Zn. The Hg uptake by both plant samples was almost equally distributed between roots, fruits and leaves parts, unlike As, which was concentrated preferentially in *P. pashia* leaves and fruits.

CONCLUSION

Different concentrations of toxic metals were estimated in the plant samples. The results showed that most of the studied toxic metals were found within the recommended limits. This study concluded that investigated *A. annua* and *P. pashia* samples accumulates significant amount of As, Cd, Pb, Hg, Zn and Ni. However, *A. annua* roots carry a slight high amount of Cd which can cause metal poisoning in humans. Therefore, special care must be taken during the administration of *A. annua* roots produce as safe remedy.

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