Effect of Heavy Metals on *Lepidium sativum*, Growing in Various Polluted Areas of Peshawar Pakistan

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Summary: Heavy metal like Cr, Pb, Cu, Cd, Fe, Ni, and Mn were investigated in the medicinal plant *Lepidium sativum* as well as in their soils using atomic absorption spectrophotometer. The plant samples were collected from three different areas i.e. polluted, less polluted and non-polluted area of Peshawar situated at a distance of 3 and 4 km from the main polluted area. The plant parts including roots, stem, leaves and seeds were found to have more heavy metal than less polluted and non-polluted area. The main purpose of the study was to make awareness among the public about the use of such medicinal plants containing high level of heavy metals and their adverse health affects.

Introduction

Environmental contamination and exposure to heavy metals such as mercury, lead and cadmium are serious growing problems throughout the world. Human exposure to heavy metals has risen dramatically in the last 50 years as a result of an exponential increase in the use of heavy metals in industrial processes and products.

The use of herbal drugs in the recent years has tremendously increased due to their lesser side effects and acceptability to the majority of the third world countries. The wide spread distribution of heavy metals in soil, due to geoclimatic conditions and environmental pollution is inevitable; therefore the assimilation in plants is obvious. Heavy metals along with pollutants are discharged into the environment through industrial activity, automobile exhaust, heavy-duty electric power generators, municipal wastes, refuse burning and pesticides used in agriculture etc. Men, animals and plants through air, water and food take up these metals from the environment. Medicinal plants, which are the raw materials for many of the herbal formulations and popular nutrient supplements are sold all over the country. In the recent past, there has been a steady growth in the number of patients visiting practitioners of complementary system of medicine. This growing popularity is partly due to the popular concept that "being natural in origin, herbs are safe".

Heavy metals also have great significance due to their tendency to accumulate in human organs over a prolonged period of time. The presence of heavy metals beyond the allowed upper and lower limits can cause metabolic disturbance. Thus both the deficiency and excess of essential micronutrients (e.g. iron, zinc, copper) may produce undesirable effects [1]. Effects of toxic metals (Cd, Cr, Pb, Ni, etc.) on human health and their interaction with essential trace elements may produce serious consequences [2]. World Health Organization (WHO) 1998 recommends that medicinal plants which form the raw materials for the finished products may be checked for the presence of heavy metals, pesticides, bacterial or fungal contamination [3]. Environmental impact of heavy metals such as Cd, Pb, Hg and As, as well as their health effects have been the source of major concern. Outbreak of Itai-itai in Japan due to the consumption of rice, containing high levels of Cd, [4] the minimata disease [5] caused after eating methyl murcury contaminated fish are some of the examples of ill effects of environmental pollution due to toxic metals. Cadmium is reported to cause osteomalacia and pyelonephrites and Pb may cause renal tumors and other carcinoma [6]. The aim of the present study was to investigate the effects of heavy metals on the cellular and acellular parts of L. sativum growing in polluted, less polluted and non-polluted areas of Peshawar Pakistan. The study will also be useful for public awareness about the use of L. sativum as a local medicine for abortion purposes having high concentration of heavy metals.

Results and Discussion

Cress or *L. sativum* is a very important medicinal plant, its leaves are antiscorbutic, diuretic

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and stimulant [7-8]. The plant is administered in cases of asthma, cough with expectoration and bleeding piles [8]. The roots are used in the treatment of secondary syphilis and tenesmus [8]. The seeds are galactogogue. When boiled with milk it is used to procure an abortion. The plant has been applied as a poultice to cure pains and wound and have also been used as an aperient [8]. The seeds contain 58% of edible oil that can also be used for lightning purposes [9]. Keeping in view the importance of this medicinal plant and its possible impact by heavy metals, a study was carried out to determine the concentrations of heavy metals in plant and soil samples and the analytical data is depicted in Table-2.

Chromium

Soil samples collected from three different spots (Table-1) showed significantly different amount of chromium. High concentration of chromium was found in plants collected from spot 1 and spot 2 (Table-1). For example in case of spot 1 high concentration was found in their leaves 0.13 mg/ kg followed by roots 0.09 mg/ kg. Although chromium was also present in the roots, stem, leaves and seeds in spot 2 and 3, however their concentration is not significant i.e. chromium is present in low concentration (Table-1). Thus in general the concentration in the three spots was in the order spot 1 > spot 2 > spot 3, while among the plant parts leaves > root > stem > seeds.

Lead

High lead concentration was found in the aerial parts (stem, leaves and seeds) of the plants collected from spot 1 and 2 respectively (Table-1). In case of spot 1, high concentration was found in the stem 0.46 mg/ kg followed by seeds 0.23 mg/ kg and leaves 0.20 mg/ kg. Thus lead concentration was in the order stem > seeds > leaves > roosts. However, high lead was found in the leaves 0.20 mg/ kg followed by seeds 0.16mg/ kg and stem 0.15 mg/ kg in the plant collected from spot 2. While considerably lower concentration of lead was found in the roots, stem, leaves and seeds in the plant collected from spot 3.

Due to soil and air pollution, lead concentration in plant parts was in the order stem > seeds > leaves. Obviously the high lead concentration in the above ground parts is due to air borne lead [10].

The plants from the three different environments accumulated different amount of lead. The most sensitive was being spot 1 and least one was spot 3.

Copper

The concentration of copper was found high in the soil from spot 1 than spot 2 and spot 3 respectively. Plants grown on the three spots contained significantly different amount of copper, due to the difference in the concentration of copper in the soils of the three spots. The level of copper concentration in roots, stem, leaves and seeds were found to be higher in spot 1 than spot 2 that has infect has more concentration than spot 3. Leaves accumulated significantly more copper 0.65 mg/kg followed by stem, which is 0.50 mg/kg. A similar

Table-1: Concentrations of Heavy Metals in Plant and soil Samples. mg/kg

Spot1	Cr	Pb	Cu	Cd	Fe	Ni	Mn
Root	0.09±0.04	0.13±0.01	0.34±0.04	nd	15.90±0.08	0.08±.01	3.50±0.01
Stem	0.06±0.05	0.46±0.06	0.50±0.00	0.06±0.02	15.92±0.08	0.04±0.03	2.42±0.03
Leaves	0.13±0.01	0.20±0.06	0.65±0.01	0.02±0.01	29.74±0.01	0.17±0.02	6.27±0.04
Seeds	0.04±0.00	0.23±0.02	0.37±0.00	0.01±0.00	17.83±0.24	0.15±0.08	2.04±.0.04
Soil	0.12±0.01	0.30±0.20	0.32±0.21	0.03±0.02	10.38±0.16	0.06±0.08	1.22±0.83
Spot2							
Root	0.04±0.01	0.12±0.04	0.32±0.00	nd	14.11±0.18	0.07±0.01	1.79±0.06
Stem	0.04±0.01	0.15±0.03	0.38±0.00	0.01±0.00	9.77±0.01	0.02±0.04	1.84±0.01
Leaves	0.09±0.02	0.2±0.02	0.39±0.00	nd	25.16±0.08	0.06±0.03	6.02±0.04
Seeds	0.03±0.00	0.16±0.06	0.27±0.01	nd	8.71±0.20	0.08±0.01	1.69±0.04
Soil	0.06±0.08	0.18±0.12	0.22±0.01	0.01±0.01	8.26±0.43	0.04±0.01	0.98±0.05
Spot3							
Root	0.04±0.04	0.09±0.04	0.26±0.00	nd	13.52±0.69	0.05±0.02	1.67±0.03
Stem	0.02±0.01	0.04±0.03	0.38±0.00	nd	7.28±0.1	nd	1.52±0.01
Leaves	0.08±0.06	0.18±0.06	0.28±0.04	nd	22.62±0.45	0.05±0.00	4.42±0.03
Seeds	0.01±0.03	0.14±0.08	0.24±0.01	nd	5.64±0.06	nd	1.46±0.01
Soil	0.05±0.01	0.14±0.02	0.28±0.02	nd	10.60±0.50	0.03±0.01	0.78±0.07

nd = not detected , ± = Standard Deviation.

WHO permissible limits for Pb: 10 mg/kg; Cd: 0.3 mg/kg (WHO 1998)

FDA permissible limits for Cr; 120 ug (RDI); Ni: 0.1 mg/l (FDA 1993, [12], 1999 [13]).

trend was found in spot 2, in which the leaves have 0.39 mg/kg and stem 0.38 mg/kg while the concentration of copper was found lower in spot 3 (Table-1). Thus the copper concentration in plant parts was in the order leaves > stem > seeds > roots.

Cadmium

In case of Cadmium, Surprisingly, small amount of was detected in the stem, leaves, and seeds of the plant sample collected from polluted area. This may be due to the polluted air from the surrounding area. Luckily no cadmium was detected in the plant samples collected from spot 2 and spot 3.

Iron

The soil samples collected from the three spots showed significant difference between the heavy metals (Table-1). The plant samples collected from the three spots have different amount of Fe. For example, high amount of iron was found in the leaves 29.74 mg/ kg followed by seeds 17.83 mg/ kg, stem 15.92 mg/ kg and roots 15.91 mg/kg in the plant samples collected from spot 1. In case of the plant sample from spot 2, high concentration was found in the leaves 25.16 mg/ kg followed by roots 14.11 mg/kg. A similar trend was found in the sample from spot 3 i.e. the leaves have 22.62 mg/ kg and roots 13.62 mg/ kg. Thus in general the order of iron was spot 1 > spot 2 > spot 3

Nickel

In the leaves of plant collected from spot 1 has high nickel concentration 0.17 mg/ kg followed by seeds 0.15 mg/ kg and roots 0.08 mg/k g. In the case of plant parts from spot 2, less amount of nickel was found Table-1 than spot 1. Surprisingly no nickel was detected in the stem and seeds of plant from spot 3, while equal amount was found in the roots and leaves 0.05 mg/kg. Thus in general the concentration of nickel in the three spots was in the order spot 1 > spot 2 > spot 3.

Manganese:

The leaves of spot 1 and spot 2 have almost equal amount of manganese 6.27 mg/kg than other parts of the plant. Thus the concentration level of manganese is well below the critical level and hence acceptable at this level, because it does not affect the plant growth nor will cause pollution.

Experimental

Lepidium sativum was collected from polluted, less polluted and relatively non-polluted areas, which are at a distance of 3 and 4 km from the main polluted area. Heavy metals like Cr, Pb, Cu, Cd, Fe, Ni, and Mn were determined in the roots, stems, leaves and seeds of the plant. Plant parts especially roots were washed in fresh running water to eliminate dust, dirt and possible parasites, and then they were washed with deionized water. Similarly soil samples were taken in plastic envelopes dried and stored. During all these steps of sample processing necessary measures were taken in order to avoid any loss or contamination with heavy metals.

Sampling areas

Spot 1 (Polluted area)

In this area the plant is exposed to both polluted soil and air pollution.

Spot 2 (less polluted area)

This area is situated at a distance of 3 km from polluted area; the plant is exposed to less polluted soil and air.

Spot 3 (Relatively non-polluted area)

This area is located at a distance of 4 km from polluted area; the plant is growing relatively in unpolluted air and soil.

Post harvest treatment of plant material

Acid digestion of soil samples (Aqua Regia Digestion)

Weighed 1 g of air-dried and sieved (< 2mm) soil was taken in a flask. 15 mL of Aqua Regia was added and swirled to wet the sample. It was kept overnight. The next day the flask was heated at 50 °C for 30 minutes. The temperature was raised to 120 °C and the heating was continued for 2 h. The flask was cooled and 10 mL of 0.25 M HNO_3 added [11].

The solution was filtered through a Whatman No. 542 filter paper. The flask and filter paper were washed with small aliquots of 0.25 M HNO_3. The filtrate and washings were transferred to a 50-mL flask and made up to the mark with 0.25 M HNO_3.

Acid digestion for plant samples

Weighed 1 g of crushed powdered part from each part of plant like root, stem, leaf and seed were taken in a china dish and heated in the furnace for 4 h keeping the temperature at 550 °C after charing. The contents of the china dish were cooled in a dessicator. Then 2.5 mL 6 M HNO_3 solution was added to the dish to dissolve its contents. The solution was transferred to a 20-mL flask and was diluted to the mark of 20-mL.

The analysis for heavy metals was done by flame atomic absorption spectrophotometer (Polarized Zeeman Hitachi-2000).

For the studied elements we established the following sensitivity and detection limits respectively of the used FAA apparatus.

Cr - 0.5 and 3.0 mg kg^{-1}

Pb - 0.2 and 1.0 mg kg^{-1}

Cu - 0.5 and 3.0 mg kg^{-1}

Cd - 0.2 and 1.0 mg kg^{-1}

Fe - 0.5 and 5.0 mg kg^{-1}

Ni - 0.5 and 4.0 mg kg^{-1}

Mn - 0.5 and 2.50 mg kg^{-1}

Conclusions

The study showed that plant grown on polluted area has high concentration of heavy metals than less polluted and unpolluted area. The population generally uses herbal medicine for prolonged period of time to achieve desirable effects. Prolong consumption of such herbal medicine might reduce chronic or subtle health hazards. Thus our finding indicate that the medicinal plant or plant parts used for different types of diseases must be checked for heavy metal contamination in order to make it safe for human consumption. In other words for local or pharmaceutical purposes, it should be collected from area not contaminated with heavy metals.

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