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Research Article

POPULATION BASED STUDY OF HEAVY METALS IN MEDICINAL PLANT, *CAPPARIS DECIDUA*

ALI REHMAN, HUSSAIN ULLAH, RIZWAN ULLAH KHAN, IJAZ AHMAD*

Department of Chemistry, Kohat University of Science & Technology (KUST), Kohat-26000, Khyber Pakhtunkhwa, Pakistan.
Email: drijaz_chem@yahoo.com

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ABSTRACT

Objectives: The main goal of this study was to create awareness among the people about the use of such widely diverse medicinal plants having high concentration of heavy metals.

Method: The plant samples were collected from three different spots i.e. populated (spot 1) less populated (spot 2) and non-populated (spot 3) areas of Karak, Khyber Pakhtunkhwa Pakistan. Essential heavy metals Fe, Cu, Zn, Co and Mn and non-essential heavy metals like Cd and Pb were analyzed in different parts of the medicinal plant *Capparis decidua* as well as in soil sample where the plant was grown by using Flame atomic absorption spectrophotometer.

Results: Heavy metals concentration in different spots were found to be in the order of Spot 3 > Spot 1 > Spot 2 with the concentration rang of different heavy metals Fe 7.51-871.39 mg/kg, Ni 5.15-48.72 mg/kg, Cr 2.37-34.65 mg/kg, Zn 5.37-70.10 mg/kg, Mn 6.19-287.50 mg/kg, Cu 3.09-17.20 mg/kg, Co 1.00-16.15mg/kg, Cd 0.46-2.90 mg/kg and Pb 1.37- 21.80 mg/kg. Ash and Moisture composition were also determined in the selected medicinal plant samples.

Conclusion: Surprisingly, the plants as well as the soil samples of non-populated area have more heavy metals than populated and less populated samples. The high content of heavy metals in plant samples of non-populated (spot 3) area may be due to the geological strata.

Keywords: Medicinal plant, Ash, Moisture, Heavy metals, Population, Atomic Absorption Spectrophotometer.

INTRODUCTION

Plants are used by man for many purposes since the beginning of human culture. According to WHO about 80% of world population relies on herbs and on traditional healing system for both curatives and prophylactic therapies and the main goal of their use is a positive interaction with biochemical reactions of the body. Their active components must be taken by the body for giving the required benefits. When they are absorbed in the blood, they circulate and influence our whole system of the body [1]. The medicinal and toxicity of plants depend on the chemical constituents including metal ions. The metals play a vital role for the formation of bioactive compounds in medicinal plants [2]. The investigation of heavy metals in medicinal plants are therefore of supreme importance in protecting the human from the hazards of these metals ions. Metals like chromium, iron, copper, cobalt and zinc are essential nutrients but they are toxic at high concentration [3]. When the levels of heavy metals become greater in plants and animals, it can produce a variety of acute and chronic effects in organisms of various ecosystems [4]. Heavy metals are found generally at low level in soil and vegetation; living organisms need these elements in micro level. However, these have deadly effect on organism at high concentration. The toxicity of heavy metals has an inhibitory effect on plant growth, stoma function, enzymatic activity and photosynthesis activity and also damages the root system [5].

Capparis decidua belongs to Capparidaceae family commonly known as karir and is indigenous to the tropical and subtropical regions of Pakistan, India, Africa, Arabia and Egypt [6]. Karir is a densely branched shrub that reaches a height of 6-10 m. It has the ability to survive in various habitats and generally grows in dry, exposed habitats such as wastelands and foothills [7]. Its leaves are very minute with a very short life time on young shoots, so that plant looks leafless. Flowers of the plant are found in small groups along the shoots in the axils of the spine while Fruit is of berry size and shape of cherry [8].

In the traditional system of medicine, the plant is widely used by local physicians for different ailments like the bark of *Capparis decidua* is used in the treatment of cough, asthma, ulcer and inflammation; roots and buds are used in treatment of fever and to cure boils respectively. Root past is applied on scorpion bite, leaves act as appetizer, helps in cardiac troubles; Root bark is used as purgative and anthelmintic; wood coal used in muscular injuries. The fruit of *Capparis decidua* has antidiabetic action. The plant flowers extract has potent activity in preventing plaque formation, while its flower buds are eaten to relieve stomach pain. The young leaves and top shoots in powder form are used as a blister treatment, they are also used in cure of boils, eruptions, swellings and as an antidote to poison, and they are very useful in relieving in toothache when chewed [9,10].

Medicinal plant having such common valuable and rich medicinal use must be evaluated for heavy metal determination before its use.

The determination of heavy metals in medicinal plant and crops is therefore of great importance in protecting the public from the hazards of these heavy metals ions.

MATERIALS AND METHODS

Samples Collection

Plant parts as well as soil samples were collected from three different areas of Tehsil Karak, i.e. populated (Spot 1), less populated (Spot 2) and non-populated (Spot 3) areas. The spot 3 was selected in hilly and rocky area. Soil samples were taken from the upper 8-10 cm of soil. Plants parts particularly roots were washed in tape water to remove dust and dirt, and then rinsed properly with deionized water. The rinsed plant parts were then dried under shed. The dried samples were grinded by using pistol and mortar and then stored in clean, dried plastic bottles. Heavy metals like Fe, Zn, Cu, Pb, Ni, Cr, Mn and Co were determined in roots, root bark, stem bark, flower and fruit as well as in soil of this plant.

Analysis of Samples

(a) Acid Digestion of Soil Samples

For heavy metals determination, all the soil samples were crushed lightly and were sieved to pass through 2-mm mesh. One gram of soil sample was treated with 10 ml of HNO_3 for 24h in china dish at room temperature, then 5 ml of $HClO_4$(70%) was added. The contents of the china dish were heated on a hot plate until the volume of contents was reducing to 2 mL. The contents of the china dish were cooled and filtered through whatman (#42) filter paper into 25 mL volumetric flask and diluted to the mark with distilled water [11]. Heavy metals like Fe, Zn, Cu, Pb, Ni, Cr, Mn and Co were analyzed with Flame Atomic Absorption Spectrophotometer [FAAS] (Perkin Elmer 400).

(b) Determination of Moisture

Five grams of each fresh plant part was taken in a Petri dish and placed in oven at 90^{0}C for 3hours. It was then removed, cooled in desiccators and weighed. The sample was further heated in an oven for another 2 hours and the process was repeated until a constant weight was achieved. The percent moisture content was determined.

(c) Ashing

One gram of crushed and powder portion from each part of plant like root, root bark, stem bark, flower and fruit was taken in crucible for heating in an oven at 105^{0}C to remove moisture. Then the dried sample after charring was placed in furnace. The furnace temperature was gradually increased from room temperature to 550^{0}C in 1 hr. The sample was ashed for about 4 hr until a white or grey ash residue was obtained. The content of crucible were cooled in desiccators and weighed. The percent ash composition was calculated by the formula reported in literature [12].

(d) Acid digestion of plant samples

After cooling and weighing of samples, 5 mL of HNO_3 (25%, v/v) solution was added into crucible and, when necessary, the mixture was heated slowly to dissolve its contents. The solution was filtered through whatman (#42) filter paper into 25 mL flask and diluted to the mark [13, 14].

Estimation of heavy metals was carried out on Flame Atomic Absorption Spectrophotometer [FAAS] (Perkin Elmer 400 was used).

RESULTS AND DISCUSSION

The various parts of *Capparis decidua* plant collected from three different spots, populated, less populated and non-populated areas were analyzed for moistures, ash and heavy metals. The percent moisture, percent ash composition and concentration of heavy metals are appended in tables 1 and 2 respectively.

Interestingly, the plant parts including roots, root bark, stem bark, flowers and fruits as well as in the soil of non-populated area were found to have more heavy metals than populated and less populated areas. This may be because of geological strata of the studied area.

Iron

Iron is the most abundant and an essential constituent for all plants and animals. It is also an essential component of hemoglobin for oxygen transport in blood and other wide variety of functions, including DNA synthesis and electron transport; it helps in the oxidation of carbohydrates, fats and protein to control body weight, which is very necessary factor in diabetes. High concentration of Fe may cause tissue damage, due to the formation of free radicals. The dietary limit of iron in the food is 10-60 mg/day [15]. Low iron content causes nose bleeding and gastrointestinal infection.

As revealed by the analysis of samples (figure- 2), the soil samples collected from different spots show significantly high concentration of Fe. In soil, the iron level occurs in the range of (283.75-871.39 mg/kg). The highest level of iron was found in spot 3 (871.39 mg/kg) followed by spot1 (421.46 mg/kg) and 2 (283.75mg/kg). The soil of spot 3 was found more contaminated in iron. Among the plant samples, the level of iron content was in the range: (7.51-94.57 mg/kg). In general, the lesser amount of iron level was recorded in flowers and fruits. The stem bark showed the comparatively high concentration of iron.

Nickel

Nickel is essential element. Nickel forms the phospholipids and also regulates the lipid level in tissue. Its deficiency results in the disorder of liver. The most common disease arising from nickel is an allergic dermatitis known as nickel itch and also adversely affects lungs and nasal cavities [16]. Although nickel is required in very small quantity for body and is mostly present in pancreas and hence take part in the production of insulin. EPA (Environmental Protective Agency) has recommended that daily intake of nickel should be less than 1mg beyond which it is toxic [17].

It is clear from table 2 that Nickel level occurs in the range of (21.00-48.72 mg/kg). The highest level of Nickel occurred in the soil of spot 3 (48.41 mg/kg). In plant parts, the level of Nickel was found in the range of (4.19-12.20 mg/kg). The Stem bark of plants collected from Spot 3 have high nickel concentration (12.20 mg/kg) followed by fruits (8.89mg/kg) and flowers (7.80 mg/kg). In case of plant parts from spot 2, lesser amount of nickel concentration was found then spot 1. In general, the nickel concentration in the three spots was in the order: spot 3> spot 1>spot 2.

Chromium

Chromium is one of the most known environmentally health hazard chemical pollutants in the world. The toxic effects of chromium intake is nose irritations, bleeds, skin rash, upset stomach, liver and kidney damage, nasal itch ,respiratory problems, weakened immune system and lung cancer [18]. The daily intake of chromium 50-200 µg has been recommended for adults by National Academy of Science [19].

Chromium level was determined in three different spots and was found in the range of (22.07-34.65 mg/kg).The highest level was found in spot 3 (34.64mg/kg) followed by spot2 (29.63 mg/kg), spot 1 (22.07 mg/kg). Among the plant parts, the level of chromium occurred in the range of 2.37 -9.58 mg/kg. In spot 1 the level of chromium was high in the root 8.26 mg/kg followed by root bark 7.55mg/kg, fruit 5.29mg/kg, flowers 5.11mg/kg, stem bark 4.85mg/kg. In spot 2 the chromium concentration was in the range of (2.37 -7.87 mg/kg). The concentration of chromium in different plant parts of spot3 was in tune of stem bark 9.24 mg/kg, flowers 6.89 mg/kg, Root bark 5.83 mg/kg, root 5.02 mg/kg and fruit 4.49 mg/kg.

Zinc

Zinc is prominent trace element for plants and animals growth and also plays a significant role in various cell processes like normal growth, bone formation, brain development and wound healing. Zinc is a neurotoxin in vitro when present in high level. The dietary limit of zinc is 100mg/kg [5].

Zinc level in the soil of selected spots was in the range of 50.28 - 70.10 mg/kg. In plant parts high amount of zinc was found in the flower of spot 3 (96.64 mg/kg) followed by the flower of spot1 (58.12 mg/kg) and spot 2 (50.28 mg /kg). Surprisingly, zinc level in flowers was greater than their respective spots. The less concentration of zinc was recorded in the fruits of all the selected spots. As compared to other elements like Ni, Cu, Co, Pb, Cd and Cr, the concentration of Zn was higher in all the plant samples

Manganese

Manganese is one of the essential elements which are related to the carbohydrate and fat metabolism. Deficiency of Mn in human beings causes myocardial infarction and disorder of bony cartilaginous growth in mammals. High intake of Manganese produces adverse effects on the brain and lungs. The daily recommended dietary intake in adults is 11 mg/day [19].

In all the three selected spots, Manganese level was estimated, which was found highest after iron. Manganese level occurred in the range of 221.87-287.50 mg/kg. The highest level was found in the soil of spot 3 (287.50mg/kg) followed by spot 1 (264.00 mg/kg) and spot 2 (221.87mg/kg). Among the plant parts of all the three spots, the stem bark accumulated significant level of manganese. The roots contain least amount of manganese. Near equal concentration of manganese has been found in both flowers and fruit of each spot. The table 2 shows that generally the upper parts of plant contain higher level of Manganese than the underground parts.

Copper

Although copper is an essential enzymatic element for plant growth and development but can be toxic in high concentration [20]. Its high concentration in Plants can cause Phytotoxicity. High levels of copper may cause metals fumes fever wit flue like symptoms, dermatitis, irritation of the upper respiratory tract, hair and skin decoloration, metallic taste in the mouth and nausea. Deficiency of copper results an anemia. The recommended daily dietary intake of copper is 2-3 mg/day [16].

Copper level was determined in all the studied samples and was in the range of 9.60-17.20 mg/kg. High level of copper was found in the soil of spot 3 followed by the soil of spot 1. The plant samples collected from spot 3 showed high level of copper as compared to the plant samples collected from spot 1 and spot 2. From the table 2, it is clear that root bark of each spot contains the high level of copper. It is also clear from the table 3, that in case of each spot, the aerial parts of plants contain high level of copper then the underground parts.

Cobalt

Cobalt is useful for humans because it is a part of vitamin B12, Which is important for humans' health. It is beneficial for anemia in pregnant women, as it activate the production of red blood cells. The high concentration of cobalt may cause asthma and pneumonia, vision problems, heart problems, nausea and thyroid damage. The recommended daily dietary dose of cobalt is about 0.04 mg by a person weighing 70 Kg [21].

The concentration of Co in the analyzed soil samples ranges from (9.6-16.15 mg/kg) while in plant sample the range is from 1.00- 2.20 mg/kg. The Co level in plant samples of spot 1 was comparatively higher then plant samples of spot 2 and spot 3.

Cadmium

Cadmium is highly toxic non-essential trace element to humans even at very low concentration. It causes neurological disorders such as hyperactivity and learning disabilities and badly affects the cardio vascular system, liver and kidney function [22].

The result shows high concentration of cadmium in the soil as well as in plant parts collected from spot 1 as compared to spot 2 and 3. In all the three selected spot, the cadmium was found to be in the range of 1.85-2.90 mg/kg as shown in table 3. In plant parts cadmium was recorded in the range of 0.46 mg/kg to 2.78 mg/kg. High level of Cd was recorded in the underground plant parts of all the selected spots. Comparatively lesser amount was found in the upper parts.

Lead

Lead is another non-essential highly toxic element having functions neither in humans nor in plants. It induce various toxic effects in humans especially in microorganism at low doses It cause toxic impact on various metabolic process like plant growth, DNA synthesis, transpiration and photosynthesis. The main symptoms' of lead intakes in humans are anemia, colic, headache, brain damage and central nervous system disorder [23].

The mean concentration of Lead in the analyzed soil and plant samples ranges from (10.42-21.80 mg/kg) and (0.37 -15.52 mg/kg) respectively. The concentration of lead was highest in spot 1 followed by spot 2 and spot 3. In plant parts, the stem bark of spot 1 contain the highest level of lead followed by roots of spot 2 and 3. The concentration of lead in the stem bark of spot 3 was high (9.86 mg/kg). The lesser amount of lead (0.37 mg/kg) was found in the flowers of spot 3. The lead concentration was in the tune; stem bark > root bark > fruit > flowers > root. A relatively lower concentration of lead was recorded in the root, flowers and fruits in the plants collected from spot 3. Generally the underground parts contain the least amount of lead. In all the three selected spots, the level of lead was different. The dietary intake limit is 3 mg/week [24].

Table 1: Percent Ash and Moisture composition in various parts of *C. decidua*

S. No.	Plant Parts	% Ash	% Moisture
Spot 1	Roots	5.01	36.48
	Root Bark	9.93	38.71
	Stem Bark	12.81	20.1
	Flower	9.93	73.24
	Fruit	7.9	72.86
Spot 2	Root	4.34	34.63
	Root Bark	9	37.64
	Stem Bark	12.79	19.99
	Flower	8.64	71.01
	Fruit	6.89	70.32
Spot 3	Root	6.03	33.01
	Root Bark	10.21	36.51
	Stem Bark	14.21	18.67
	Flower	8.43	70.11
	Fruit	7.01	71.65

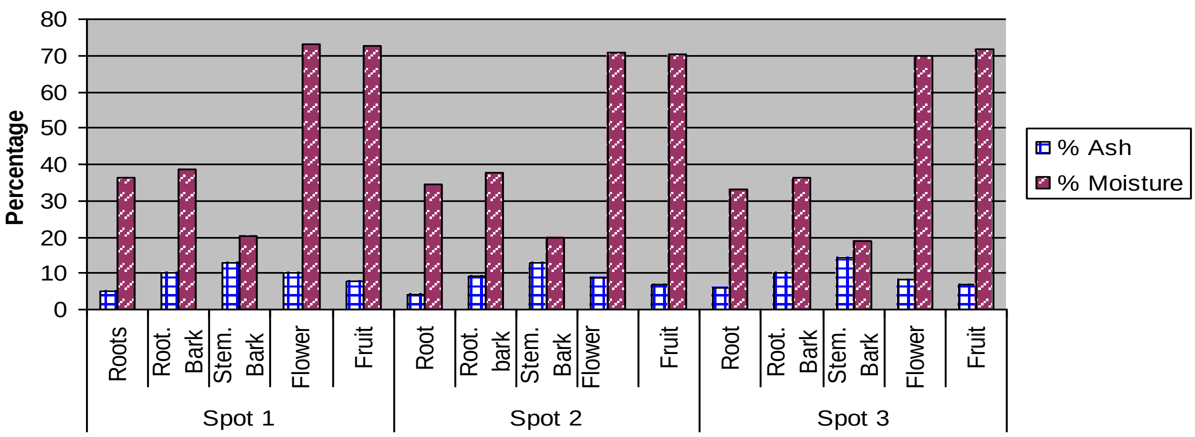

Fig. 1: Showing percentage of Ash and Moisture in Various parts of medicinal plant

Table 2: Heavy metals concentrations (mg/Kg) in various parts of *Capparis decidua* and Soil samples

Location	Sample	Fe	Ni	Cr	Zn	Mn	Cu	Co	Cd	Pb
Spot 1	Soil	421.46+1.90	33.41+0.49	22.07+0.16	58.12+0.89	264.00+1.49	11.25+0.09	10.46+0.21	2.90+0.08	21.80+0.14
	Root	37.75+0.42	5.150+0.09	8.26+0.19	22.52+0.34	8.43+0.08	4.14+0.08	1.25+0.01	1.61+0.01	3.52+0.05
	Root Bark	34.78+0.33	5.67+0.04	7.55+0.23	92.96+0.87	6.57+0.09	5.22+0.0.1	2.20+0.03	2.78+0.03	5.25+0.08
	Stem Bark	75.83+0.67	7.95+0.08	4.85+0.12	42.37+0.45	106.27+0.09	10.33+0.05	2.17+0.03	0.78+0.01	9.34+0.07
	Flower	15.09+0.09	7.97+0.21	5.11+0.43	89.87+0.86	12.25+0.20	7.23+0.11	1.66+0.02	0.77+0.01	2.41+0.02
	Fruit	7.51+0.13	6.39+0.11	5.29+0.12	5.37+0.20	15.25+0.15	7.22+0.09	1.76+0.01	0.91+0.02	3.50+0.02
Spot 2	Soil	283.75+1.70	21.00+0.24	29.63+0.17	50.28+0.23	221.87+1.62	9.65+0.15	9.6+0.21	1.85+0.02	12.98+0.13
	Root	31.37+0.45	4.19+0.08	7.07+0.22	32.16+0.56	6.19+0.22	3.09+0.01	1.72+0.02	1.47+0.03	2.77+0.01
	Root Bark	30.64+0.23	5.36+0.05	3.65+0.11	23.56+0.87	15.42+0.34	4.86+0.09	1.54+0.04	0.76+0.01	3.23+0.04
	Stem Bark	65.13+0.43	7.18+0.08	7.87+0.22	30.36+0.98	69.24+0.87	8.27+0.12	1.78+0.01	1.97+0.01	7.14+0.03
	Flower	15.48+0.12	6.51+0.05	4.58+0.10	80.17+0.69	11.23+0.12	6.17+0.11	1.70+0.02	0.93+0.01	4.41+0.01
	Fruit	12.27+0.09	5.43+0.07	2.37+0.0.9	6.39+0.31	3.92+0.11	6.31+0.08	1.43+0.02	0.87+0.03	6.21+0.01
Spot 3	Soil	871.39+1.95	48.72+0.67	34.65+0.22	70.10+0.13	287.50+1.81	17.20+0.19	16.15+0.11	2.08+0.07	10.42+0.10
	Root	44.28+0.56	6.37+0.09	5.02+0.21	21.31+0.43	11.97+0.26	4.55+0.06	1.00+0.01	1.79+0.04	2.03+0.02
	Root Bark	26.19+0.21	6.74+0.13	5.83+0.20	19.50+0.54	17.53+0.11	6.85+0.02	1.55+0.01	1.11+0.01	3.80+0.06
	Stem Bark	94.47+0.45	12.20+0.11	9.24+0.43	63.05+0.67	118.69+0.91	15.13+0.13	1.52+0.02	0.73+0.01	9.86+0.04
	Flower	18.88+0.11	7.80+0.23	6.89+0.23	96.64+0.90	16.75+0.09	7.69+0.21	1.08+0.03	0.48+0.02	1.37+0.05
	fruit	33.95+0.13	8.89+0.14	4.49+0.11	10.52+0.21	13.79+0.23	7.03+0.20	1.16+0.01	0.46+0.01	2.98+0.04

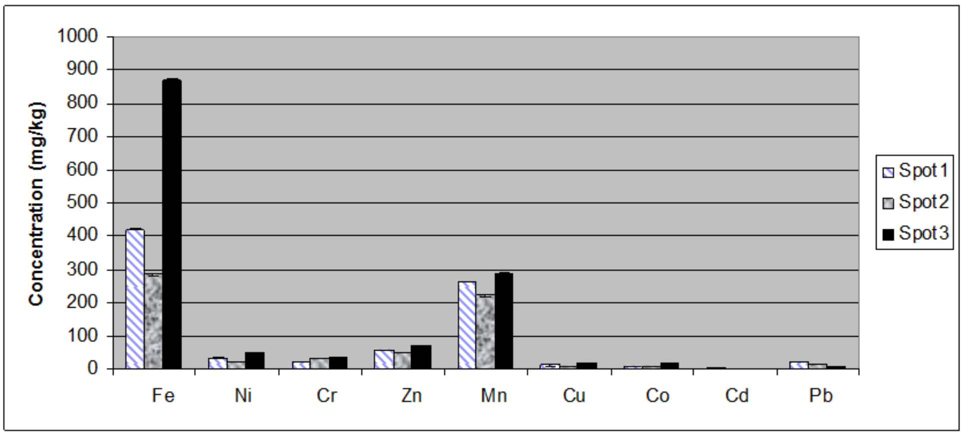

Fig. 2: Comparison of heavy metals concentration in soils of different spots

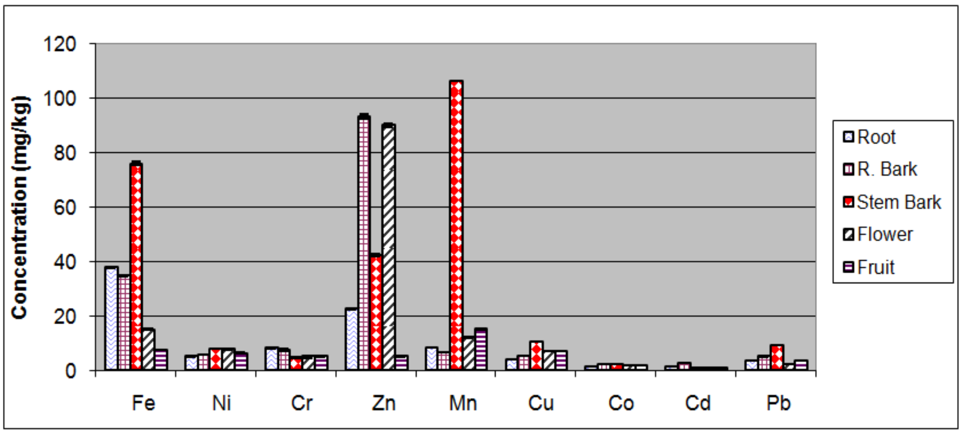

Fig. 3: Showing of heavy metals concentration in Populated area (spot 1)

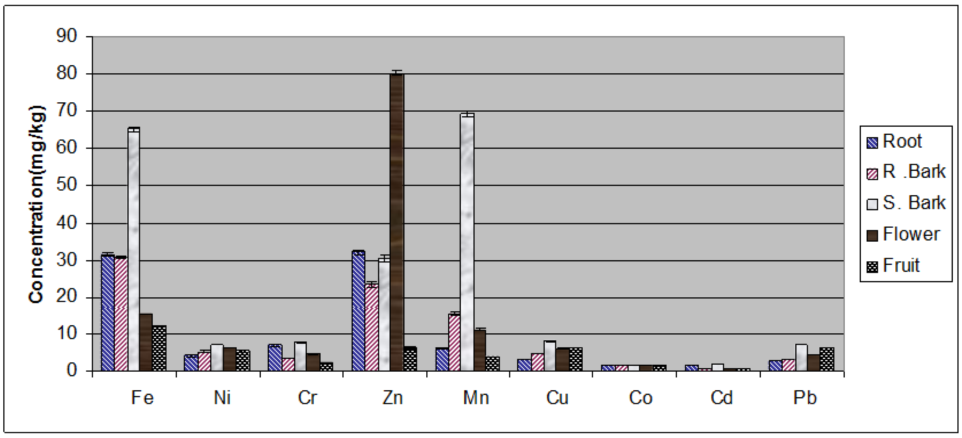

Fig. 4: Showing of heavy metals concentration in less populated area (Spot 2)

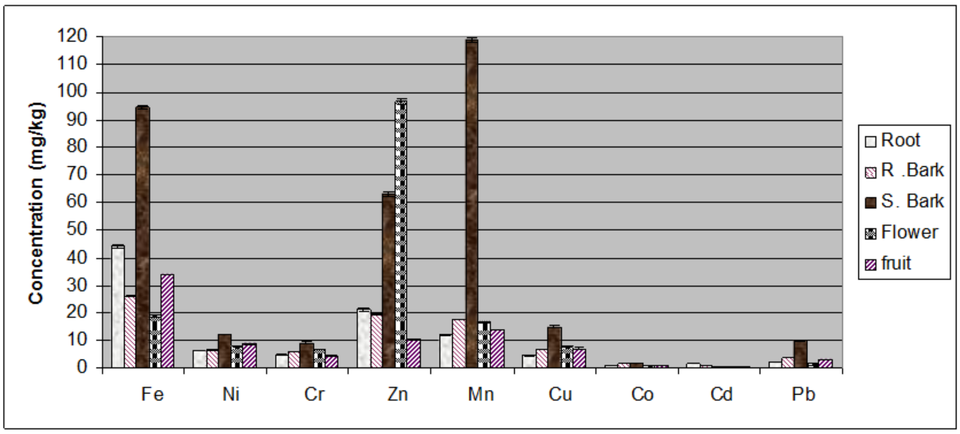

Fig. 5: Showing of heavy metals concentration in Non Populated area (Spot 3)

CONCLUSIONS

The study showed that soil collected from non-populated area has high concentrations of heavy metals than populated and less populated areas. It may be due to geological strata of the studied area; as a result the heavy metals uptake by plant in that spot was greater. Medicinal plants are traditionally used by local people for long period of time to achieve desirable results. Accumulation of heavy metals varied from spot to spot. These are lower in residential area than in non-residential area. Thus, it reiterates our belief that every medicinal plant should be analyzed for contaminant load before processing it for further pharmaceutical purposes or for local human consumption.

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