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Contamination of soil with Heavy metals from industrial effluents and their translocation in green vegetables of Peshawar Pakistan

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Abstract

Heavy metal pollution is a serious problem in countries like Pakistan, claiming soil, water and food resources rapidly due to negligence. Accumulation and bio-concentration of seven heavy metals in vegetables growing in mixed industrial effluent irrigated agricultural field near Hayatabad, Peshawar, Pakistan was determined by atomic absorption spectrometer. Ranges of concentration of copper, cobalt, iron, lead, chromium, manganese, zinc and nickel was 0.044-0.504, 0.009-0.085, 0.243-7.737, 0.496-0.474, 0.005-0.033, 0.019-2.019, 0.045-0.703 and 0.017-0.108 ppm respectively, in the edible parts of different vegetables growing in the area. Iron (Fe) accumulated to the highest level in all vegetables ($P < 0.05$). In general root was heavily contaminated with the metals, showing significantly greater accumulation than other parts ($P < 0.05$). A significant amount of the metals absorbed by the vegetables was Translocated to the edible parts, indicating health concerns. Cultivation of vegetable crops with such industrial effluents is not recommended on the studied land without certain remediation.

Key words: Industrial effluent, Irrigation, Vegetables, Heavy metals, Bioconcentration

1. Introduction

Metals are intrinsic components of the earth crust; however, today soil contamination with heavy metals is an environmental problem on a global scale and is becoming increasingly important as industrialization increases¹. Industrial activities such as mining, electroplating and manufacturing of essential commodities produce huge volume of waste water as effluent that contains heavy metals and other toxicants discharged². Being non-degradable, heavy metals stay persistently in the environment, accumulating at toxic levels in soil, water and food chain. Metals can also accumulate in the soil at toxic levels due to long term application of wastewater³. Apart from discharges from several industrial areas, metals continuously enter into the water source consumed by human beings and animals, endangering their growth and health^{4,5}. In many areas these discharges are used by local farmers for irrigating their crops, thus introducing these pollutants to the crops⁶. Very often plant chemical composition is modified without damage being easily visible, and plants grown in contaminated soils contain higher quantities of metals than plants grown in uncontaminated soils^{7,8}. In this changing scenario reuse of domestic and industrial wastewater in agriculture

for irrigating crops appears to be a lucrative option due to appreciable amount of plant nutrients present in this water⁹. A number of studies from developing countries have reported heavy metal contamination in wastewater and wastewater irrigated soil^{2, 10, 11}. In many developing countries, including India, farmers are irrigating their crop plants with industrial effluents^{2, 9, 12, 13} having high^{2, 13, 14} due to the non-availability of alternative sources of irrigation water. Several researchers have been documented use of diluted industrial effluents/sludge has growth and productivity enhancing effects on crop plants^{13, 15-17}. However, loading of heavy metals often leads to degradation of soil health and contamination of food chain mainly through the vegetables grown on such soils⁹. The process of metal uptake and accumulation by different plants depend on the concentration of available metal in soils, solubility sequences and the plant species growing on these soils¹⁸.

Long term wastewater irrigation may lead to the storage of heavy metals in agricultural soils and plants. Food safety issues and potential health risks make this as one of the most serious environmental concerns¹⁹. Vegetables store heavy metals in their edible and non-edible parts. Although some of the heavy metals such as Zn, Mn, Ni and Cu act as micro-nutrients in lower concentrations, they become toxic at higher concentrations. Health risk due to heavy metal contamination of soil has been widely reported^{20, 21}. Crops and vegetables grown in soils contaminated with heavy metals have a greater accumulation of heavy metals than those grown in non-contaminated soil^{22, 23}. Intake of vegetables is an important way of heavy metal toxicity to human being. Bioavailability of Cd, Cu, Zn and Mn in the human gastrointestinal tract from the edible part of vegetables using an in vitro gastrointestinal (GI) extraction technique was assessed by²⁴. Lettuce and radish were found to be more responsible than other vegetables for the accumulation of heavy metals in humans through the edible portion²⁴. The absorption capacity of heavy metals depends upon the nature of vegetables and some of them have a greater potential to accumulate higher concentrations of heavy metals than others.

Vegetable constitute an important component of human diet since they contain carbohydrates, proteins, as well as vitamins minerals and trace elements²⁵. Vegetables constitute essential components of the diet, by contributing protein, vitamins, iron, calcium and other nutrients which are usually in short supply. They also act as buffering agents for acid substances obtained during the digestion process. However, these plants contain both essential and toxic elements over a wide range of concentrations. One important dietary uptake pathway could be through vegetable crops irrigated with metal contaminated waste water. In Hayatabad, Peshawar, Pakistan a 100 Km long channel has been constructed for the discharge of effluents from several industrial units specially pharmaceuticals, pigments, petrochemicals, dye, paints, pesticides, chemical, lubricants, etc. The effluent channel is designed for effluent collection from various industrial estates and finally discharged into the Arabian Sea. Local farmers across the channel use this waste water to irrigate their agricultural fields for growing crops due to the scarcity of fresh water for irrigation. The

vegetables generally grown in the area of Hayatabd are Spinach (*Spinacia oleracea* L), Radish (*Raphanus sativus*), Tomato (*Lycopersicon esculentum*), Cress (*Lepidium sativum*), Dill (*Peucedanum graveolens*), Coriander (*Coriandrum sativum*), (*Capsicum annum*), Cabbage (*Brassica oleracea* var capitata), Brinjal (*Solanum melongena*) and Okra (*Abelmoschus esculentus*) by local farmers. However, there is very limited empirical information from Pakistan for heavy metal contents in soil and irrigation water and its accumulation and translocation to crop plants especially in vegetables form such metal contaminated agricultural field. It is of prime importance to know the degree of translocation of heavy metals from soils to plants used as food crops, and studies on the absorption of metals by food plants grown on soils to a safe level as not to cause phytotoxicity symptoms are of great practical interest. Indeed, edible plants produced from such soils could expose unknown consumers to the risk of ingesting high doses of metals that exceed the law and that, in the long-term, could cause cases of sub-acute or chronic intoxication¹.

The present study was selected in this agriculture field to establish a direct relationship of level of metals in such contaminated agricultural field and the vegetable crops growing there in The main objective of the present studies is to quantify the level of metals concentration in soil and their translocation in vegetables to evaluate health hazards and which may be helpful in making policies for growing safe vegetables in the present study area of Peshawar.

2. Materials and methods

2.1. Study area

The area selected for the present study was around effluent discharge channel located in Hayatabad, Peshawar, Pakistan. A number of industrial units are located in the adjacent area. Most of the treated and untreated industrial effluents are being discharged through the effluent channel. Several acres of agricultural land is irrigated by channel effluent water and local farmers cultivate various types of crop of economic importance, including seasonal vegetables. As per the information given by the local farmers, we have identified the above area where wastewater irrigation has been a common practice for many years and irrigation water has been the treated or untreated industrial effluent.

2.2. Sampling and analysis of industrial effluent and tube well water

For physico-chemical and metal analysis, samples of industrial effluents and tube well were collected in clean acid washed glass bottles of 500 ml each. 2 ml of concentrated HNO₃ was added to the water to avoid microbial utilization of heavy metal and other nutrients. The bottle containing samples were brought to the laboratory and digestion was completed within a week.

2.3. Soil sampling and analysis

Soil samples were collected from seven different sites of the Hayatabad Peshawar, irrigated with industrial effluents and tube well water. The samples were dried in the sun for four days, the samples

were then reduced to laboratory samples by using the tabling process. The samples were then dried in an oven at 110 °C for about five hours to remove the moisture completely. The samples were then ground into fine powder (80 mesh).

For sample preparation Nitric-perchloric acid digestion method²² was performed. 1 g of a sample was placed in 250 ml digestion tube and 10 ml of concentrated HNO₃ was added. The mixture was boiled for 30-45 min to oxidize all easily oxidizable matter. After cooling, 5 ml of 70% HClO₄ was added and the mixture was boiled gently till the appearance of dense white fumes. The contents were cooled and 20 ml of distilled water was added, and re boiled to stop the release of any fumes. The solution was cooled again, filtered off through Whatman No.42 filter paper and transferred to 50 ml volumetric flask. The volume was made up to the mark with distilled water. Blank solution was prepared by the same procedure, except the addition of soil sample. Heavy metals were studied using atomic absorption spectrophotometer. For the variability of results, all experiments were carried out in triplicate

2.4. Collection and analysis of vegetable crops

Five vegetable crops, like onion (*Allium cepa*), Tomato (*Lycopersicon esculentum*), Chili (*Capsicum annum*), Brinjal (*Solanum melongena*) and okra (*Abelmoschus esculentus*), seasonally grown in the agricultural field of Hayatabad by local farmers were collected. For each species five samples were collected randomly at their maturity stage and seasons in plastic bags and brought to the laboratory. The collected vegetable samples were washed with distilled water to remove dust particles. The samples were then cut to separate the roots, stems, leaves and fruits with the help of a knife. Different parts (roots, stems, leaves and fruits) of vegetables were dried in air for two days and then placed in oven for 6-8 hours at 110 °C. Dried samples of different parts of vegetables were ground into a fine powder (80 mesh) using a commercial blender and stored in polyethylene bags, until used for acid digestion.

1g of ground, oven dried plant sample was taken in a small beaker. 10 mL of concentrated HNO₃ was added and allowed to stand overnight. The mixture was carefully heated on a hot plate until the production of red NO₂ fumes was ceased. The contents of the beaker were cooled and small amount (2-4 mL) of 70% HClO₄ was added. The beaker was heated again and the contents were allowed to evaporate to a small volume. The solution was filtered using what man No.42 filter paper. The filtrate was diluted to 50 ml with distilled water using volumetric flask. All the samples were prepared using the same procedure. Heavy metals concentration was studied using atomic absorption spectrophotometer. For the variability of results, all experiments were carried out in triplicate.

2.5. Analysis of heavy metals

All samples, including soil and plants were analyzed using atomic absorption spectrophotometer. The standards for all elements were used from PERKIN ELMER. The samples and blank were studied under the specified Conditions for atomic absorption spectrophotometer given in table 1.

2.6. Data Analysis

Translocation factor (TF)

Translocation factor was calculated as the ratio of metal concentration in the edible part of the plant and metal concentration in plant root, as follow,

$$TF (\text{Edible}) = \frac{C_{\text{Edible}}}{C_{\text{Root}}}$$

Where C_{stem} , C_{root} and C_{edible} is the concentration of metals in plant stem, root and edible part respectively.

Bio-concentration factor (BCF)

Bio-concentration factor is the ratio of metal concentration in plant tissue (root, stem etc.) and concentration of metals in agriculture field.

$$BCF (\text{Edible}) = \frac{C_{\text{Ediblepart}}}{C_{\text{Soil}}}$$

Where C (edible part), and C (soil) is the concentration of heavy metals in the edible part and soil respectively.

3. Results and discussion

3.1. Metal concentration:

The study site selected in Hayatabad, Peshawar, Pakistan, have a large number of industrial units, including chemicals, petrochemicals, pesticide, dye, dye intermediate, pharmaceuticals, Agrochemicals, etc. where high amounts of liquid and solid waste are present in the vicinity. Effluent from these industries dumped to a common drain, when analyzed by atomic absorption spectroscopy, was found to contain Ni, Zn, Pb, Co, Cr, Fe and Mn in concentration 0.04, 0.36, 0.18, 0.13, 0.05, 2.39 and 0.15 ppm respectively. Repeated irrigation of agriculture soil with industrial effluents has resulted in contamination of soil with heavy metals leading to high content of Zn, Pb, Ni, Co in these fields. It is clear from Figure 2, that the average concentration of heavy metals in the soil irrigated with industrial effluent water was 0.63, 1.87, 0.87, 0.12, 0.49, 3.54 and 4.77 ppm for Nickel, zinc, lead, cobalt, chromium, iron and manganese. It was noticed that in addition to other crops, farmers grow vegetables in these contaminated field, which may lead to serious risk in case of human consumption²⁶. To investigate the accumulation of heavy metals in green vegetables, we determined the concentration of these metals in the root, stem, leaves and edible parts of vegetables (onion, tomato, brinjal, okra and chili) grown in the affected fields. For the

comparison concentration of metals in vegetables grown in normal fields, irrigated with tube well water was also determined. In most of the cases greater accumulation of heavy metals was recorded in vegetables grown on contaminated soil than the vegetables grown in normal soil. The difference was statistically significant (Duncan multiple range test; $p < 0.05$). Metals accumulation was greatest in the roots of all plants studied with maximum retention of heavy metals in this part (Figure 2). Nevertheless, stem, leaves and fruits also accumulated significant amounts of the contaminant metals translocated from the roots. In green vegetables, iron (Fe) accumulated to the greatest level, although its concentration in the soil and industrial effluent was lesser than other metals. The concentration of this metal in the edible parts of onion grown on contaminated soil was 10 fold greater than the control onion. Irrigation with contaminated water resulted in more than two fold greater accumulation of Mn in the edible parts of onion. The concentration of other metals was also significantly increased in the edible parts of this vegetable ($p < 0.05$). Tomato fruits grown on soil irrigated with industrial effluent were also heavily contaminated with heavy metals, iron being the most serious contaminant showing over 24 fold enhanced accumulation than control. Roots, stem and leaves of the exposed crop also contained a significant amount of the metals (Figure 3). The fruits of brinjal and Okra exposed to industrial effluent accumulated significantly greater amount of Zn, Pb, Fe, Cr and Mn than their normally grown counterpart ($p < 0.05$). Concentration of Zn, Pb, Fe and Cr was significantly greater in the fruits of chili grown on contaminated soil than control fruits.. Accumulation of metals in the edible parts of onion was in the order of; Fe>Mn>Cr>Co>Pb>Zn>Ni. Concentration of different heavy metals in the fruits of tomato (the edible parts) was in the order of Fe>Co>Zn>Mn>Pb>Ni>Cr.

The contamination and accumulation of Cr, Mn and Co in the edible part of tested vegetable was higher than the permissible limits set by WHO²⁷, making the vegetables unsafe for human consumption. Different vegetable species accumulated different metals depending on environmental conditions, metal species and plant available with forms of heavy metals. Overall, the results on comparison revealed that metals in water had more impact on vegetation than the soil itself. Plants have a natural tendency to take up metals. Some metals like Co, Fe, Mn, Ni and Zn, are essential mineral nutrients, while the others like Cr and Pb, have no known physiological activity²⁸. Accumulation of heavy metals in plants has been shown to induce the production of phytochelatin (PCs), a family of thiol-rich peptides²⁹. Plants sustain metals at comparatively low concentrations by avoiding extreme uptake and transport metal^{28, 30} by different mechanisms, including chelation, compartmentalization, biotransformation and cellular repair mechanisms^{31, 32}. Plants have an extremely high capacity to uptake metals by the roots, translate and store them in shoots^{28, 30, 33}. The accumulation of heavy metal from soil to plant did not follow any particular pattern and varied with respect to metals, species and plant parts. Some metals like Cr and Pb, have very low solubility in soil and showed particularly strong barrier. If the heavy metals accumulate in the root,

they do not translocate significantly to the leaves, fruit or seed. For other elements, it is protected against transfer to the edible part of the crop is under the physiological control of the plant. There were large differences in the metal concentration between the root and aerial parts among the studied vegetables crop, which showed an important restriction of the internal transport of metals from root towards shoot. Similarly, Bose and Bhattacharyya¹⁵ reported higher concentration of heavy metals in root than shoot in wheat plant.

3.2. Translocation Factor (TF).

The translocation factor of heavy metals from root to an edible part in the vegetables irrigated with tube well water and industrial effluents mixed water is shown in Figure 4. Translocation factor varied from metal to metal and plant to plant in the studied vegetables and did not follow any specific sequence.

The concentration of metals in Plants is kept relatively low as compared to the edible parts by avoiding excessive metal uptake and transportation³⁰, due to chelation, compartmentalization, biotransformation and cellular repair mechanisms^{31, 32}. Plants have an exceedingly high capacity to take up metals by roots and translocate and store them in the shoots^{28, 30, 33}.

Certain metals like Cr and Pb, due to their low solubility in soils have shown particularly strong barrier. They accumulate at the root, and are not usually translocated significantly to the aerial parts of the plants like leaves, fruit or seed. For other elements, transfer to the edible part of the crop is protected under the physiological control of the plant. From many experiments, it is evident that metals can be ranked on the relative scale on the strength of this barrier; Pb, Cr, Hg>Cu>Ni>Zn>Cd>Mo>Ti. There were large differences between the root and aerial parts concentrations among the vegetable crop of all metals studied, which indicated an important restriction of the internal transport of metals from root towards shoot.

A general trend of high translocation factor (TF) from root to the edible parts of vegetables grown on contaminated water was observed (Figure 3) as compared to their control counterpart ($p > 0.05$). Cobalt (Co) showed the highest TF (1.39 ± 0.05) from the roots to the edible parts was found in okra grown in contaminated soil. In onion, tomato, brinjal and chili the higher TF from root to the edible parts was recorded for Mn (1.29), Zn (1.00), Ni (1.08) and Ni (0.77) respectively.

3.3. Bio-concentration factor (BCF).

The uptake factor from soil to plant is recognized as Bio-Concentration Factor (BCF). BCF in vegetables irrigated with industrial effluents is given in Figure 5. In general, BCF found in the root was more than that of stem, leaves and fruit for different metals with some exceptions. Maximum BCF was

recorded in case of Co (9.56), followed by Fe (4.40) and then Ni (2.33). High BCF in edible parts of vegetable irrigated with such metal loaded industrial effluent indicated high uptake and accumulation of toxic metals, which could easily enter into the food chain and may affect dangerously the human being and animals. It is reported that the use of treated and untreated waste water for irrigation has increased the contamination of Cd, Pb and Ni in edible parts of vegetables causing some potential risk in the long term²². The average level of Cr is 60 mg/day²⁷, Cu is 1.5–3.0 mg/day³⁴, Mn is 11mg/day. National Research Council recommended the dietary allowance of Iron for male is 10–12 mg and 15 mg for female during pregnancy and the recommended limit of Fe is up to 30 mg³⁵. Vegetables are indispensable part of the food taken both in cooked and uncooked forms. The required amount of vegetables in our daily diet must be 300–350 g per person²⁷.

Though, the determination of essential and toxic metals concentrations in vegetables is important from the view point of crop yield techniques, food nutrition values and health impacts. In event of their presence in excess, these metals enters into the body and may disturb the normal function of central nervous system, liver, lungs, heart, kidney and brain, produce hypertension, abdominal pain, skin eruptions, intestinal ulcer and different types of cancer³⁰.

Conclusions:

Heavy metals contamination in soil above the tolerable level has been found to be dangerous in many aspects, such as its direct effects on physical and chemical nature of the soil and indirect effects on plants grown in that soil. Plants grown in soil near to the industrial areas get a large amount of heavy metals and thus have adverse effects on living organisms.

The present study also concludes that the practice of routine irrigation with mixed industrial effluents has resulted increased metals concentration in the agricultural field and subsequently plants growing therein. The vegetable studied accumulate and translocate variable amounts of metals from the soil into their tissue by different extents. The vegetables like Onion, Tomato, Brinjal, okra and Chili irrigated with industrial effluents mixed water showed high accumulation and translocation of toxic metals (Pb, Zn, Pb, Co, Fe, Cr, Mn) in their edible parts. The results indicate a considerable risk and the hazardous impact of metal contaminating vegetable crops for human being as well as animals through the food chain. In order to evaluate the potential risk associated with the use of vegetable crops grown on metal contaminated soils, it is necessary to monitor the agricultural soils and crop plants for the presence of toxic metals. However, the vegetable crops restricting toxic metal in non- edible portion may be recommended for cultivation in such metal contaminated soil where irrigation is being made with industrial effluent.

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Table 1: Conditions specification for atomic absorption spectrophotometer

Element	Wavelength(nm)	Slit Width(nm)	Lamp Current	Flame Nature
Copper (Cu)	324.8	0.7	10	Air-Acetylene
Cobalt(Co)	240.7	0.2	30	Air-Acetylene
Iron(Fe)	248.8	0.2	30	Air-Acetylene
Lead(Pb)	283.3	0.7	10	Air-Acetylene
Chromium(Cr)	357.9	0.7	25	Air-Acetylene
Manganese (Mn)	279.5	0.2	20	Air-Acetylene
Zinc(Zn)	213.9	0.7	15	Air-Acetylene
Nickel(Ni)	232.0	0.2	25	Air-Acetylene

Figure 1. Sampling map, a. Industrial irrigated field b. Tube well water irrigated field (Control) Hayatabad peshawar

Figure 2. Mean concentration of different heavy metals mixed industrial effluent (water) and soil irrigated with wastewater (soil). Bars labeled with different letters indicate significant differences among means determined by using Duncan's multiple-range test ($P = 0.05$).

Figure 3. Accumulation of heavy metals in different vegetables including onion (a), tomato (b), brinjal (c), okra (d) and Chilli (e), irrigated with industrial effluent mixed wastewater (IE) and tube well water (Control). Bars labeled with different letters indicate significant differences among means determined by using Duncan's multiple-range test ($P < 0.05$).

Figure 4 Transfer factor (root to fruit) of heavy metals from root to fruit in vegetables including onion (a), tomato (b), brinjal (c), okra (d) and Chilli (e), irrigated with industrial effluent mixed wastewater (IE) and tube well water (Control). Bars labeled with different letters indicate significant differences among means determined by using Duncan's multiple-range test ($P < 0.05$).

Figure 5: Bioconcentration of different metals in vegetables including onion (a), tomato (b), brinjal (c), okra (d) and Chilli (e), irrigated with industrial effluent mixed wastewater (IE) and tube well water (Control). Bars labeled with different letters indicate significant differences among means determined by using Duncan's multiple-range test ($P < 0.05$).

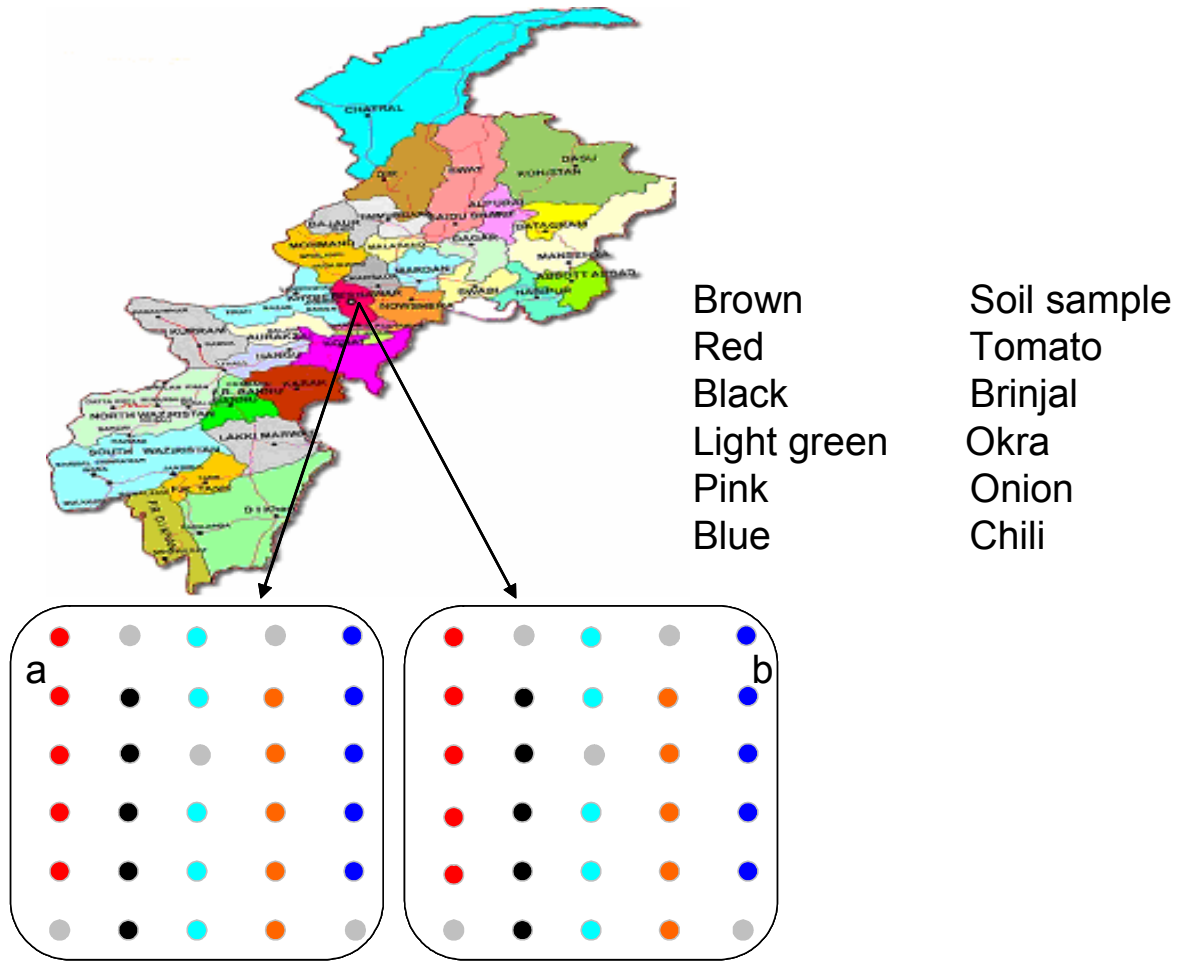


Figure 1. Sampling map , a. industrial irrigated field b. Tube well water irrigated field (Control) Hayatabad peshawar

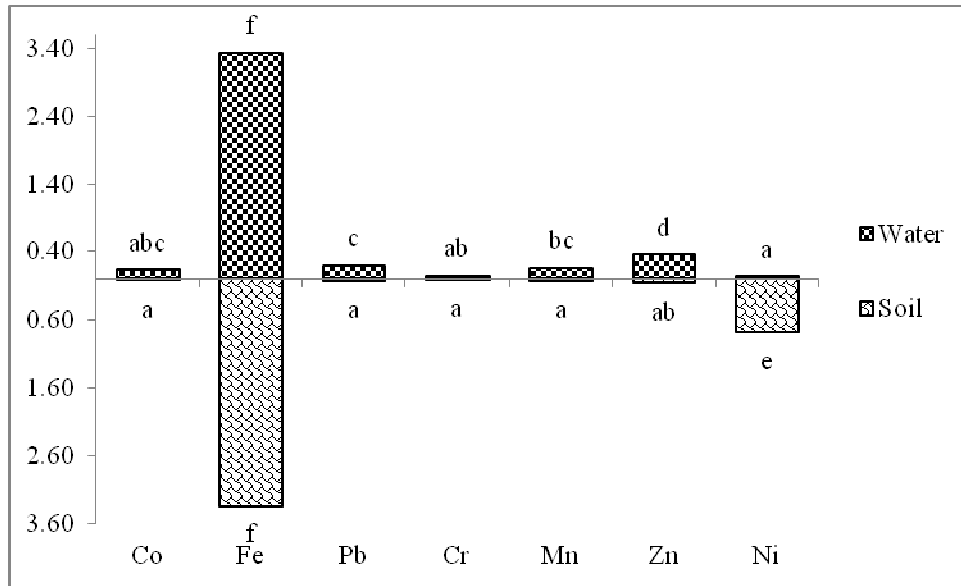


Figure 2

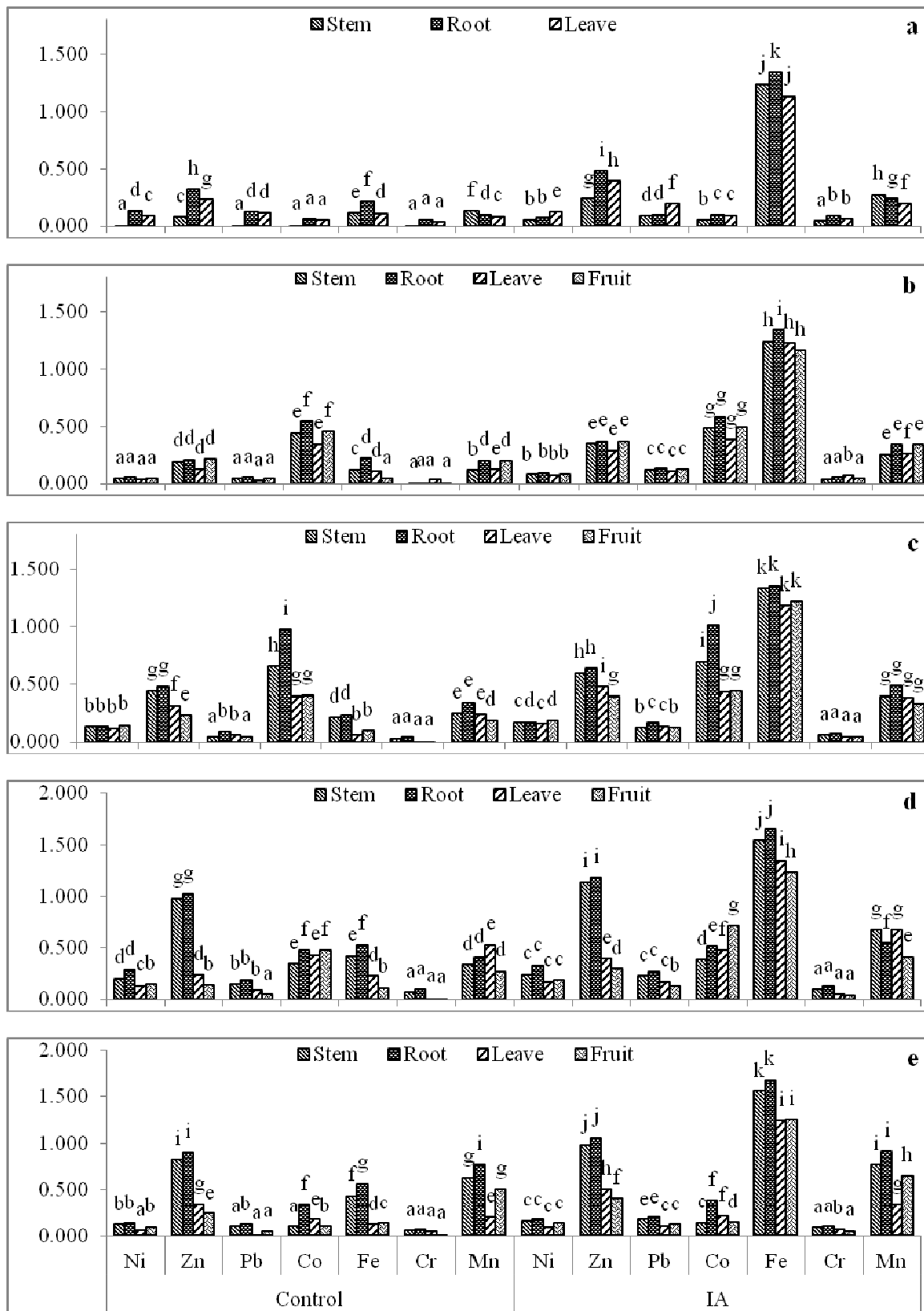


Figure 3

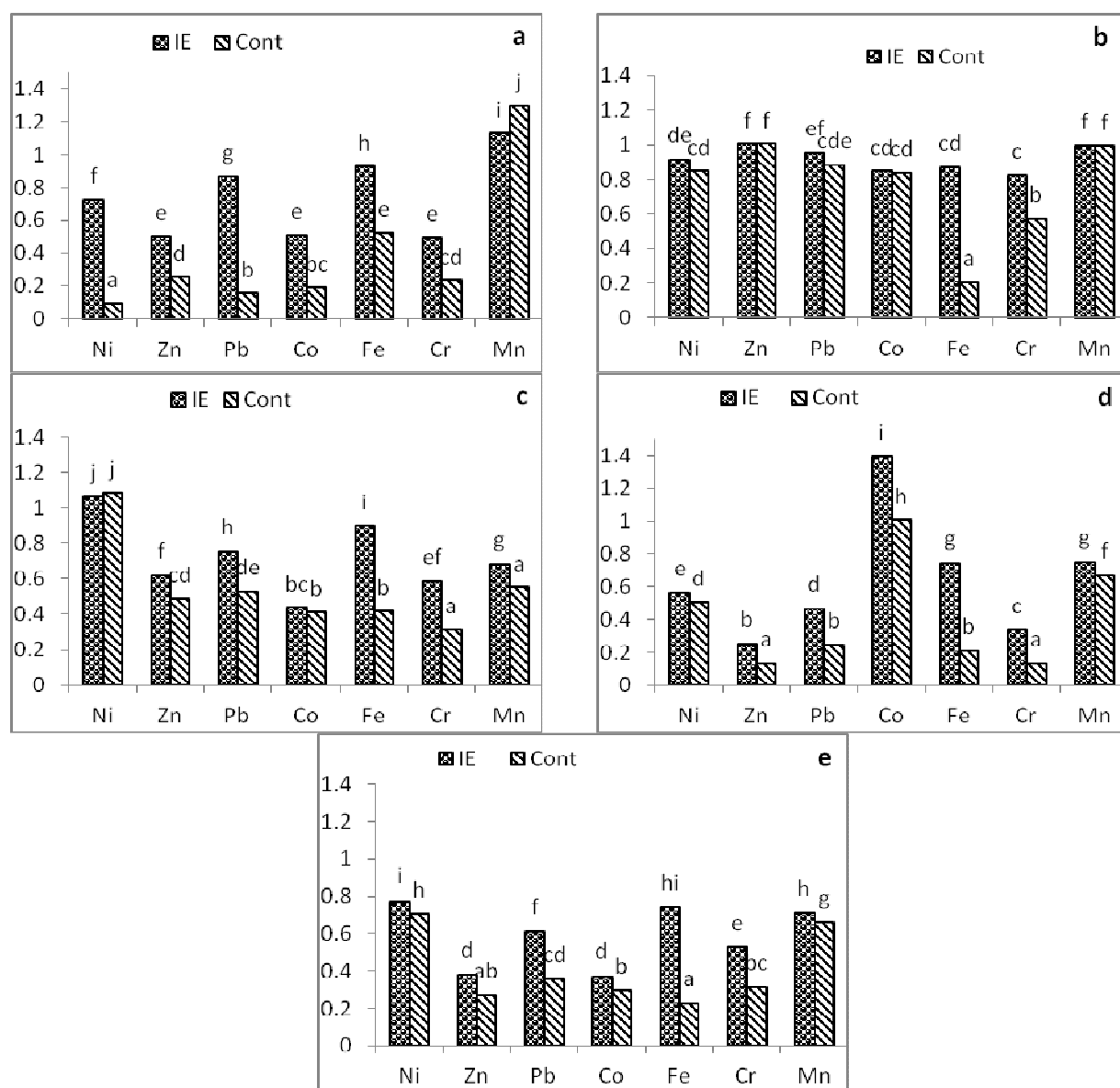


Figure 4

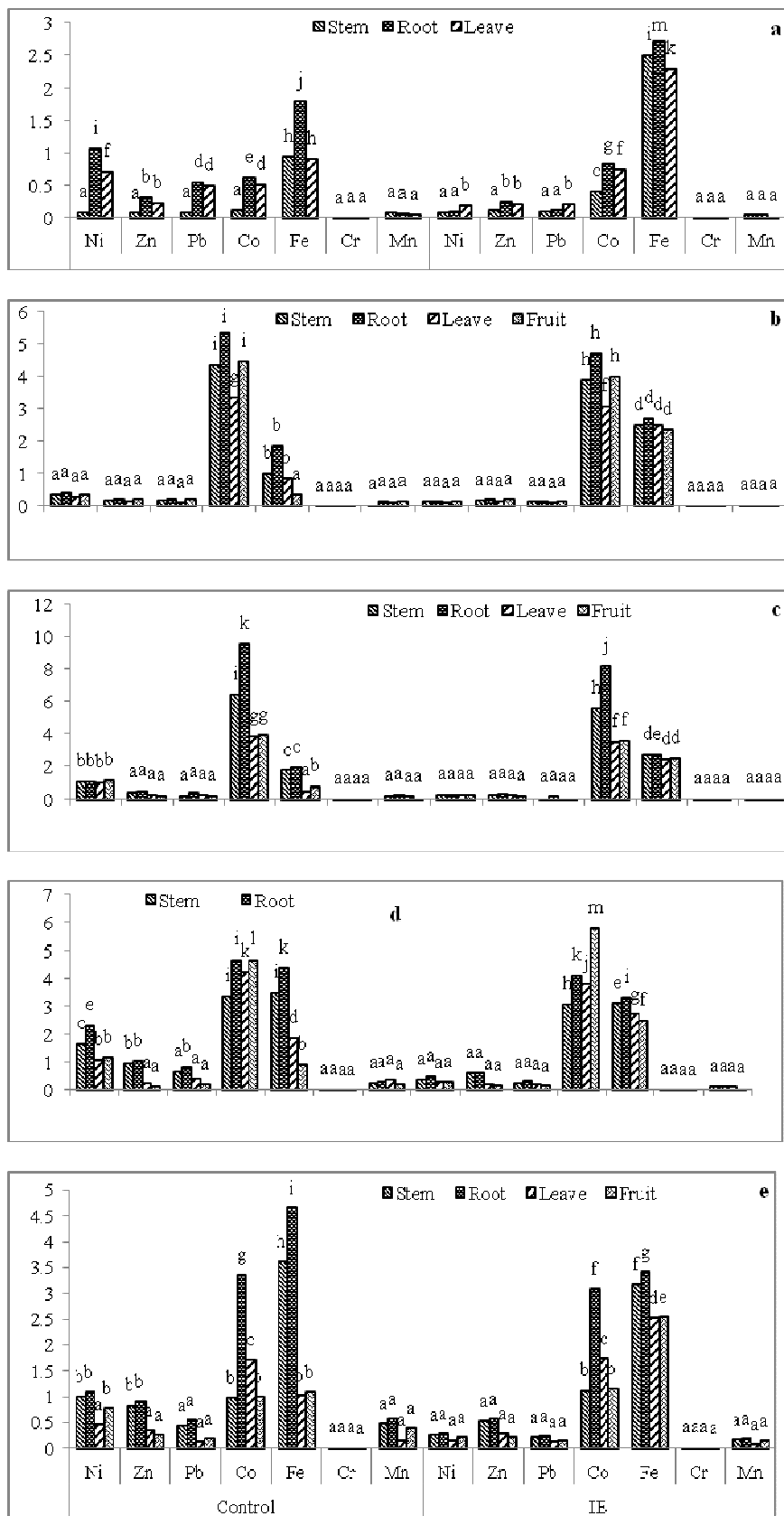


Figure 5