ASSESSING METALS TRANSLOCATION INTO DIFFERENT PART OF RADISH (*RAPHANUS SATIVUS L.*) IRRIGATED WITH SEWAGE WATER

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Abstract

A field experiment was performed to assess the accumulation and translocation of Copper, Iron, Manganese and Zinc in Radish. The vegetable was taken from Malir, Sharafi Goth. The land was irrigated with 100% sewage water for unknown period of time. The quantitative analysis of metals was made in different parts of the vegetable, soil and sewage water using Atomic Absorption technique. The pattern of metal accumulation in soil, sewage water and vegetable were in order of soil > sewage Water > vegetable. The metal accumulated in root soil, root, leaf in order of Fe > Mn > Zn > Cu while stem showed Fe > Zn > Mn > Cu. The results showed that irrigation with sewage water can not only accumulate high amount of metals in soil but it can also be transferred into vegetable growing on that soil.

Introduction

Urbanization has increased food demand in the city of Karachi that has forced farmers adopt methods of irrigation through which higher crop yields may be obtained (Liu *et al.*, 2009). Irrigation with sewage water is one of the alternative method being practiced in the metropolitan cities. Sewage contain plant macro and micro nutrient elements; nitrogen, potassium, phosphorus, zinc, copper, iron manganese etc. (Fang and Wong, 1999). This is the reason why growers utilize sewage water for irrigation purposes as it may be an alternative source of fertilizers. Application of sewage water can also increase crop yield (Frost and Ketchum, 2000). However, prolonged use may be detrimental due to accumulate metals in different parts of the plant (Gu *et al.*, 2012) and their high concentration can become potentially toxic (Hall, 2002).

Once plants grown through continuous sewage water may contaminate and incorporate metals in food chain. However, translocation of these metals within plants is also depended on both plant and soil nature (Kim and Kim, 1999). Clays and organic soils hold nutrients and water much better than sandy soils due to higher cation exchange capacity (CEC) than sandy soil. Metal accumulation in plant can also depend on root uptake, root selectivity, ion interactions, rhizosphere processes, leaf uptake from the atmosphere, and plant partitioning. The behavior of metals (such as cadmium, chromium, cobalt, copper, mercury, molybdenum, nickel, lead and zinc in contaminated soils depends on the intrinsic charge, valence and speciation of the contaminant ion, and soil properties; pH, redox status and contents of clay and organic matter (Brummer, 1986)

Zinc (Zn) is the micronutrient elements needed by plant for proper growth as zinc take part in enzymatic reactions and is necessarily required for chlorophyll production and starch formation (Abbas *et al.*, 2013). Iron is absorbed by the plants in its ionic form and plays important role in chlorophyll's biosynthesis; however deficiency may lead to chlorosis (Katyal and Randawa, 1983). Importance of copper in protein synthesis, nitrogen fixation and metabolism of carbohydrate is higher and deficient within plants may cause retardation of fruit and seed however excess may be toxic due to free radical formation (Hiroaki *et al.*, 2008). Manganese is an essential nutrient required for normal growth and development of plants (Mchargue, 1923). It is absorbed by the plants in its ionic form Mn^{+2} and functioning as an enzymes activator influencing photosynthesis, chloroplast formation, carbon dioxide and nitrogen metabolism (Mengel and Kirsby, 1987).

Radish is an edible root vegetable carries several benefits as its diuretic impacts in nature (Kumar *et al.*, 2009). Its justified use may decrease urinary related disorders is powerful detoxifier and purifies the blood. Besides, It is rich in anthocyanins flavonoids which have displayed reduce occurrence of cardiovascular disease (Taylor, 2011)

The health risks depend on the chemical composition of the waste material, type of vegetable cultivated and the consumption rate (Cobb *et al.*, 2000). Insufficient work so far has been carried out on radish completely irrigated with sewage waters in Karachi. Therefore, this study is designed and field experiment was conducted to investigate the pathway of accumulation of metals in radish.

Materials and Methods

The study area was located in catchment of Malir river in east of Karachi metropolitan city, where the vegetable & fruit grown along and within the catchment of the river are irrigated with domestic and industrial

effluent from Korangi and Landhi industrial zones. Agricultural lands in this area have been under wastewater irrigation for an unknown period of time.

Contaminated Soil Analysis: Different samples of soil were taken from the depth of 20-25cm including the channel path and root soil of vegetable. For essential metal extraction, about 1g dried sample of soil was air dried and sieved. Sample was digested in 20.0 mL aquaregia, which was prepared by mixing HNO₃, HCl and HClO₄ in a ratio of (20:20:20). Sample was digested at 80°C until a transparent solution was obtained. This transparent solution was then filtered through whatman filter paper 542 and diluted in 100mL volumetric flask with 0.1M HNO₃. The concentration of essential trace nutrient in the filtrate was determined by using Atomic Absorption Spectrophotometer (Varian 220FS, Australia).

Sewage water analysis: Many water samples were collected from the main stream sewage water connected to the irrigated land. Sample was cleared of organic matter by wet ashing method \cdot . Five hundred-mL samples of wastewater were evaporated to 20 mL on a hot plate and 5 mL HNO₃ added; heating continued with HNO₃ being added until the solution cleared. Solution was than filtered and transferred to volumetric flask, the volume was makeup with deionize water. Samples were analyzed with an Atomic Absorption Spectrophotometer (Varian 220FS, Australia) equipped with a thermal atomizer, air-acetylene flame, autosampler and autodilution.

Radish analysis: Radish samples were washed with distilled and de-ionized water to remove soil particles from the surface, dehydrated at 80 °C for 24 h and crushed. Approximately 0.5g of sample was transferred to digestion tube, 20mL of 65% nitric acid and 3.0 mL of perchloric acid were added, the tubes were left in fuming hood for about 30minutes then they were kept into microwave digester. Digested samples were cooled down, filtered through whatman filter paper and transferred to 100mL volumetric flask and volume was made-up by 0.1M HNO₃. Analysis of essential trace elements (Fe, Cu, Mn, Zn) were carried out using flame atomic absorption spectrophotometer (Varian 220FS, Australia).

Results and Discussion

It is well known hypothesis that application of sewage water for irrigation purpose can change some physicochemical properties of the soil which consequently change the uptake of minerals by plant. Findings of the experiment are shown in the following table:

The soil used in the experiment is found to have a basic pH of 8.2 and concentration of all the nutrient especially Fe showed higher accumulation in both soil samples; contaminated soil-01 and contaminated soil -02 (234238.3 \pm 0.54 and 176268.6 \pm 0.701 mg/Kg) respectively. It could be the reason that soils contain aluminosilicate lattice Fe⁺², Fe⁺³ mainly have an affinity to bind with these silicate lattice and hence iron become biologically unavailable, their ability reduce and they tend to accumulate in soil (Maria and Silvia, 2014)

Cu showed least tendency to accumulate in both soils contaminated soil-01 and contaminated soil-02 $(4314.3 \pm 0.71 \text{ and } 2114.4 \pm 0.93 \text{ mg/Kg})$ respectively shown in figure No 1. As reported by (Brar and Sekhon, 1978) that Fe and Cu have an antagonist effect on each other, higher concentration of Fe in the soil has decreased the accumulation of Cu in soil. The pH of the soil was found to be basic 8.2 and according to (Nyle, 1980) Cu, Zn, Mn are those metals which can easily form chelate which reduces their availability to the system and decreases their mobility (Nyle, 1980).

Previous literature reveals that redox condition of soil and its pH influences Mn bioavailability (Marschner 1995; Porter *et al.*2004). When pH of soil becomes basic >8.0 autoxidation of Mn^{+2} takes place manganese can also form many oxidizable form such as MnO_2 , Mn_2O_3 , Mn_3O_4 and even Mn_2O_7 which become less accessible to plant leading to their accumulation in soil as stated by (Nyle, 1980) metals having high oxidation state tend to accumulate in soil.

The translocation of metal in the soil was found in order of Fe > Mn > Zn > Cu. Heavy metal concentration in soil was also reported by (Yousufzai *et al.* 2000) in Karachi which have an order of Fe > Zn > Mn > Cu in soil. Literature reveals that in the past few years accumulation of Fe in soil has been raised upto17th times when compared with this recent research. Low level of Cu in soil among other metals was also reported in literature.

Effluent of industries, hospitals, laboratories and domestic wastes are the main contributor to form sewage. Sewage contain many heavy metals, essential nutrient, some leached pesticides, microorganism etc. Analysis of sewage water showed that it contain high concentration of Fe (Figure 2) for SW-01 and SW-02 (853.5 ± 0.61 and 631.4 ± 0.74 mg/L), respectively as compared to any other metal in the sewage which is an accordance with (Yousufzai *et al.*, 2000) who has reported maximum amount of Fe in sewage water. The metal concentration in the sewage water was in order of Fe > Mn > Zn > Cu and this order is also observed by (Yousufzai *et al.*, 2000) and (Maria and Silvia 2014). However contaminated soil has also followed this order but the concentration of these metals were very high in soil shows long time irrigation with sewage water has accumulated metals in the soil.



Fig. 1. Concentration of metals in contaminated soil-01 and contaminated soil-02.



Fig. 2. Concentration of metals in Sewage water (SW-01) and Sewage water (SW-02).



Fig. 3. Concentration of metals in root soil, root, stem and leaf of radish.



Fig. 4. Antagonism between Zn and Cu.



Fig. 5. Antagonism between Cu and Fe.

The metals determined in the different part of Radish have been presented in table 1. Data reveals that leaves has accumulated highest concentration of Cu and Mn as compare to other part of the plant 1025.9 ± 0.41 and 2264.8 ± 0.603 mg/Kg respectively. The translocation of Cu was in order of leaves > root soil > root > stem where Mn followed order of leaves > root > root soil > stem having a maximum of 2264.8 mg/Kg and minimum of 1055.36 mg/Kg. Fe has followed root > stem > leaf > root soil pattern having a maximum accumulation of 7426.4 mg/kg and least 4148.4 mg/kg. Zinc showed an order of stem > root > leaf > root soil having a maximum amount of zinc 2369.57 mg/kg and minimum of 1175.5 mg/kg (Figure 3).

Root soil, root and leaf has followed a similar pattern of up taking nutrient Fe > Mn > Zn > Cu while stem has showed different pattern of Fe > Zn > Mn > Cu (Table 2). This pattern of up taking nutrient in stem was also observed by (Kumar and Chopra, 2014).

According to (Arora and Sekhon, 1982; Dangerwala, 2001) zinc behave antagonistically to Cu and we have found the exact relation (Figure 4), where the concentration of Zn was high Cu suppress itself in that part of plant.

Concentration of Cu in all part of plant was found to remain low as compare to other metals. Concentration of Fe can reduce the concentration of Cu (Brar and Sekhon, 1978). It has to be noted that Fe concentration has prominent itself in all part of the plant hence reducing concentration of Cu in plant (Figure 5). (Zarcinas *et al.* 2004) reported that uptake of copper and iron is strongly correlated with organic matters present in sewage water. Low level of Cu may be due to the natural protective response of the plant (Chaoui *et al.* 1997) as high concentration of Cu may cause toxicity to the plant (Hiroaki, *et al.* 2008) due to formation of free radicals.

Table 1. Concentration of Cu, Fe, Mn, and Zn in sewage water (Sw-01), (Sw-02), Contaminated Soil-01, Contaminated soil-02 and in different part of radish.

·	Cu(mg/Kg)	Fe(mg/Kg)	Mn(mg/Kg)	Zn(mg/Kg)
SW-01	83.60 ± 0.15	853.5 ± 0.61	663.90 ± 0.89	276.4 ± 1.10
SW-02	64.20 ± 0.75	631.4 ± 0.74	572.4 ± 0.94	321.5 ± 0.78
Cont. Soil-01	4314.36 ± 0.71	234238.3 ± 0.54	52611.3 ± 0.85	13407.04 ± 0.56
Cont. Soil-02	2114.42 ± 0.93	176268.6 ± 0.71	56294.2 ± 0.87	16453.3 ± 0.57
Root Soil	718.95 ± 0.55	4148.45 ± 0.79	1369.8 ± 0.65	1175.54 ± 0.88
Root	588.79 ± 0.76	7426.41 ± 0.74	2051.21 ± 0.58	1728.33 ± 0.49
Stem	518.28 ± 0.69	6710.44 ± 0.49	1055.36 ± 0.83	2369.57 ± 0.55
Leaf	1025.96 ± 0.41	5729.71 ± 0.61	2264.85 ± 0.60	1413.64 ± 0.73

Table 2. Relative abundance of micronutrients in the different part of radish.

Cu Leaf > Root Soil > Root > Stem

Fe Root > Stem > Leaf > Root Soil

Mn Leaf > Root > Root Soil > Stem

Zn Stem > Root > Leaf > Root Soil

References

Abbas, M., Zahida, T. M., Uddin, R., Sajjid, I., Akhlaq, A., Moheyuddin, K., Salahuddin, J., Mari, A. H. and Panhwar, R. N. (2013). Effect of zinc and boron fertilizers application on some physicochemical attributes of five rice varieties grown in agro-ecosystem of Sindh, Pakistan. *American-Eurasian J. Agric. & Environ. Sci.*, 13(4):433-439.

Arora, C. L. and Sekhon, G. S. (1982). The effect of soil characteristics on the zinc-copper interaction in the nutrition of wheat. *Jour Agric. Sci.* 99(01) pp 185-189

Brar, M. S., and Sekhon, G. S. (1978). Effect of zinc and copper application on the yield and micronutrient content of wheat (*Triticum aestivum L.*). *Jour. Ind. Soc. Soil. Sci.* 26(1):84-86.

- Brummer, G. W. (1986). The Importance of Chemical "Speciation" in Environmental Processes. Vol. 33;pp 169-192.
- Chaoui, A., Mazhoudi, S., Ghorbal, MH., and Ferjani, E. (1997) Cadmium and zinc induction of lipid peroxidation and effect on antioxidant enzyme activities in bean (*Phaseolus vulgaris L.*). *Plant Sci* 127:139–147.
- Cobb, G. P., Kristin, S., Melissa, W., bobby, G. W. and Elaine, D. K. (2000). Accumulation of heavy metals by vegetables grown in mine wastes. *Envi. Toxi. Chem.* 19(3) pp. 600–607.

- Dangerwala, R. T. (2001). Need for sustaining balanced supply of micronutrient in soil rather than their correction. *Jour. Ind. Soc. Soil. Sci.* 49(4):647-652.
- Fang, M., and Wong, J. W. C. (1999). "Effects of lime amendment on availability of heavy metals and maturation in sewage sludge composition. *Environ Pollut*. 106:83-89.
- Frost, H.L., and Ketchum, L.H. (2000). "Trace metal concentration in durum wheat from application of sewage sludge and commercial fertilizer. *Adv Environ Res* 4:347–355
- Gu, C., Bai, Y., Tao, T., Chen, G., Shan, Y. (2012). "Effect of sewage sludge amendment on heavy metal uptake and yeild of Ryegrass seedling in a Mudflat Soil. *Jour Environ Qual* 42(2):421-428.
- Hall, L. (2002). "Cellular mechanisms for heavy metal detoxification and tolerance". Jour Exp Biol 53:1-11
- Hiroaki, Y., Marinus, P. and Toshiharu S., (2008). How do plants respond to copper deficiency? *Plant Signaling & Behavior* 3:4, 231-232.

Katyal, J. C., Randhwa, N. S. 1983. Micronutrients. F.A.O, Fertilizer Bulletin No. 7, Rome (Italy).

- Kim, K. H., and Kim, S. H. (1999). "Heavy metal pollution of agricultural soils in central regions of Korea". *Water Air Soil Pollut*. 111:109–122.
- Kumar, R., Mehrotra, N. K., Nautiyal, B. D., Kumar, Praveen., and Singh, P. K. (2009). Effect of copper on growth, yield and concentration of Fe, Mn, Zn and Cu in wheat plants (*Triticum aestivum L.*). Journal of Environmental Biology. 30(4): 485-488.
- Kumar, V. and Chopra, A. (2014). Accumulation and translocation of metals in soil and different parts of French bean (*Phaseolus vulgaris L.*) amended with sewage sludge. *Bulletin of environmental contamination* and toxicology. 92(1): p. 103-108.
- Liu, W. X., Liu, J. W. (2009). Accumulation and translocation of toxic heavy metals in winter wheat (Triticum aestivum L.) growing in agricultural soil of Zhengzhou, China. *Bulletin of environmental contamination* and toxicology. 82(3): 343-347.
- Maria., D. P. M., Silvia G. A. (2014). The Transfer of Cu, Zn, Mn and Fe between Soils and Allium Plants (Garlic and Onion), and Tomato in the Southwest of the Buenos Aires Province, Argentina. A. J. of Plant Sci. 5(4):480-487
- Marschner, H. (1995). Mineral Nutrition of Higher Plants (2nd ed.). San Diego C: Academic Press. 889
- Mchargue J. S. (1923). Effect of different concentrations of manganese sulphate on the growth of plants in acid and neutral soils and the necessity of manganese as a plant nutrient. *Journal of Agricultural Research*. Washington, D. C. June; 24(9): 793
- Mengel, K., and Kirkby, E. A., (1987). Principles of Plant Nutrition, 5th Ed. Kluwer Dordrecht. 849.
- Nyle C. Brady. (1980). Nature and Properties of Soil. Macmillan Publishing Co. Inc. New York. 565.
- Porter, G., Bajita-Locke, J., Hue, N., & Strand, S. (2004). Manganese solubility and phytotoxicity affected by soil moisture, oxygen levels, and green manure additions. *Communications in Soil Science and Plant Analysis*. 35, 99–116.
- Taylor., C. W. (2011). Anthocyanins in Cardiovascular Disease. Adv Nutr. 2(1): 1-7.
- Yousufzai A. H. K., Durdana R H., Ishaq, Q., Farooq, A. and Ishretullah, S. (2000). Determination of heavy metals in vegetables and soils at sewerage farm in Sindh industrial trading estate (SITE), Karachi. *Jour. Chem Soc Pak.* 23(1):7-15.
- Zarcinas, B. A., Pongsakul, P., Mclaughlin, M. J. and Cozens, G. (2004) Heavy metal in soils and crops in Southeast Asia. 2. Thailand. *Environ Geochem Health*. 26:359–371