

Investigation of Metals Accumulation in Some Vegetables Irrigated with Waste Water in Shahre Rey-Iran and Toxicological Implications

¹Mohsen Bigdeli and ²Mohsen Seilsepour

¹Agricultural Extension, Education and Research Organization,

²Department of Soil and Water Research, Varamin Agricultural Research Center, Iran

Abstract: Vegetables grown at environmentally contaminated sites in Sahre Rey could take up and accumulate metals at concentrations that are probably toxic to human health. This study was conducted to analyze the metal contents of some vegetables in Sahre Rey-Iran with emphasis on their toxicological implications. Recently matured leaf and fruit samples of Shahre rey vegetable farms were sampled and analyzed to determine heavy metals. Data showed that metal uptake differences by the vegetables are attributed to plant differences in tolerance to heavy metals and vegetable species. The lead concentration in all vegetable samples was more than maximum permitted concentrations, while Cd pollution was observed in radish, Cress, Dill, spinach and eggplant. Data showed that Zn concentration in Celery, Mint, Dill, Spinach and Green pepper were more than Zn permitted level. There was no evidence about Cu contamination in vegetables. Data also showed that the intake of most of the metals constitutes less than the TMDI (theoretical maximum daily intake) at present and hence health risk is minimal. But with increase in vegetable consumption by the community the situation could worsen in the future. Treatment of industrial effluents and phyto-extraction of excess metals from polluted environments could reduce health risk.

Key words: Metals accumulation • Toxicological Implication • Shahre Rey • Iran

INTRODUCTION

Vegetables constitute an important part of the human diet since they contain carbohydrates, proteins, as well as vitamins, minerals and trace elements [1]. In recent years their consumption is increasing gradually, particularly among the urban community. This is due to increased awareness on the food value of vegetables, as a result of exposure to other cultures and acquiring proper education [2]. However, they contain both essential and toxic elements over a wide range of concentrations. Metal accumulation in vegetables may pose a direct threat to human health [3]. Heavy metals are one of a range of important types of contaminants that can be found on the surface and in the tissue of fresh vegetables. Heavy metals, such as cadmium, copper, lead, chromium and mercury, are important environmental pollutants, particularly in areas under irrigated with waste water. Several investigations of water, soil and vegetables pollution by waste water are available [4-12]. Research findings show that at least 20 million hectares of land in

North and South Africa, South America, Middle East, Southern Europe, South West America, Mexico and a significant part of Central and East Asia is irrigated by raw sewage, mainly for cultivation of vegetables. Consequently, this usage ends to soil contamination and heavy metals accumulation both in soil and crops [13, 14]. Investigations conducted earlier indicate that on an average, more than $6 \text{ m}^3 \cdot \text{s}^{-1}$ water and surface water discharged by the urban conglomerate Tehran, through drains and canals which collect its various urban and industrial wastes, is used for the purpose of irrigating fields and farmland located in the south of Tehran such as Shahre rey [15]. Long-term use of this waste water, which is mainly used for cultivation of leafy and other vegetables, has resulted in the accumulation of heavy metals in the soil and their transfer to the various crops under cultivation, with levels of contamination that exceed permissible limits [15].

Heavy metal accumulation in soils is of concern in agricultural production due to the adverse effects on food quality (safety and marketability), crop growth

(due to phytotoxicity) [16, 17]. Metals such as lead, mercury, cadmium and copper are cumulative poisons. These metals cause environmental hazards and are reported to be exceptionally toxic [18, 19]. Vegetables take up metals by absorbing them from contaminated soils, as well as from deposits on different parts of the vegetables exposed to the air from polluted environments [20]. It has been reported that nearly half of the mean ingestion of lead, cadmium and mercury through food is due to plant origin (fruit, vegetables and cereals). Metal contamination soils may be widespread in urban areas due to past industrial activity and the use of fossil fuels [21-25]. Heavy metals may enter the human body through inhalation of dust, direct ingestion of soil and consumption of food plants grown in metal contaminated soil [26-28]. Potentially toxic metals are also present in commercially produced foodstuffs [29]. Results of Ni *et al.* [30] indicated that the cadmium concentrations in shoots and roots varied both with different Cd levels and type of vegetable. Generally Cd accumulation in various plant parts in vegetable crops was increased with the increasing cadmium concentrations in the growth medium. Root Cd increased more sharply than shoot Cd. Celery contained higher Cd in the edible parts than other vegetable species. Investigation of soil pollution levels in fields and farmlands in the country has revealed that the quantity of Cadmium and Lead in samples collected from contaminated areas in the provinces of Tehran, Gilan, Zanjan, Esfehan and were in the range of 1.9 to 180.5 and 89.4 to 2610.4mg kg soil, respectively [31-33]. The results of investigations conducted by Torabitni and Mahjouii [33] show that the range of contamination levels in the fields and farmland to the south of Tehran, for Cadmium is 0.1 to 7.54 mg kg⁻¹ soil and in the crops cultivated in the area 0.398 to 1.43' mg kg⁻¹ dry weight of the crop, which is higher than the permissible limits for human consumption. Permissible level of consumption of Cadmium, for human is 70 µday-1 [34]. Prolonged use of waste water for irrigation of fields and farmland in Ghazvm has caused concentration of Lead, Copper, Cadmium and Zinc to exceed permissible limits several times over [31].

Shahre Rey is one of the most important areas for vegetables production. This is located in the south of Tehran. Vegetables grown in the Shahre Rey are include leafy vegetables, eggplants, tomato, pepper and etc. These are often grown on the embankments along the major rivers within Shahre Rey town and the neighboring villages of Ghaleno, Firoozabad and Dehkheir. This vegetable farms are among the biggest farms in the capital, where a substantial amount of vegetables is

being produced seasonally. Some parts of these farms are irrigated with the wastewater from Firoozabad River. Before several decades, the water from the rivers in the capital was clean. However, with the increase in the urban population and industrialization, the water has now become contaminated with various pollutants, among which is a heavy metals. Heavy metal intake by human populations through the food chain has been reported in many countries with this problem receiving increasing attention from the public as well as governmental agencies, particularly in developing countries [35-36]. The leafy vegetables under this study were Raddish, Cress, Celery, Dill, Coriander, Spinach and Green pepper, Chili, Tomato and Eggplant. The objective of this study was to analyze the metal concentrations of some vegetables grown in Shahre Rey with special emphasis on their toxicological implications.

MATERIALS AND METHODS

Geographic Setting of the Vegetable Farms: The vegetable farms were located in the central part of the city along the road of Tehran-Varamin. A part of farms extending irrigated with Firoozabad River while more heavily contaminated with heavy metals (Table1). The vegetable samples in this study were taken from the side irrigated with the Firoozabad River.

Plant Sampling, Preparation and Analysis: Twenty five recently matured leaves or fruits of from 25 different plants were sampled at early maturity in the year 2007.

Table 1: Mean Concentration of some trace elements found in soils and Firoozabad River

Elements	Guidelines for irrigation water		Guidelines for soil	
	River (µg.L ⁻¹)	Soil(total mg.kg ⁻¹)	Irrig. Water (µg.L ⁻¹) ^b	Soil (total mg.kg ⁻¹) ^c
As	n.d	n.d	100	20
Cd	80.00	0.67	10	3
Co	n.d	n.d	50	50
Cr	n.d	70.00	550	100
Cu	59.00	54.00	17	100
Fe	332.00	34000.00	500	50000d
Mn	117.00	870.00	200	2000d
Ni	2050.00	32.00	1400	50
Pb	60.00	60.00	65	100
Se	n.d	n.d	20	10
Zn	69.00	190.00	200	300

n.d.=not detected

^bSource: [41] ^cSource: [39] ^dSource: [40]

Table 2: Concentration of some trace elements(mg kg⁻¹) found in some vegetables under waste water irrigation

	Cd	Pb	Mn	Fe	B	Zn	Cu	Ni	Co
Radish-root	0.59	0.75	130.50	1666.00	35.75	63.15	12.52	1.73	0.18
Radish-leaf	12.86	1.58	284.75	4272.50	41.20	93.90	5.78	20.52	3.04
Cress	1.25	0.74	164.30	1678.00	27.89	100.00	4.54	3.26	0.33
celery	0.04	3.83	91.77	257.30	49.44	188.00	18.96	0.00	0.01
Mint	0.09	3.07	131.90	374.70	43.95	202.40	24.43	0.00	0.01
Dill	0.23	2.42	37.51	426.03	26.89	263.42	23.18	0.00	0.01
spinach	0.20	2.57	61.05	509.40	21.48	297.40	22.74	0.00	0.01
Coriander	0.13	1.35	43.95	439.60	21.99	61.36	26.92	0.00	0.00
green Pepper	0.00	1.56	89.95	317.80	19.22	1132.00	21.80	0.00	0.01
Red Pepper	0.03	1.41	28.58	413.70	14.67	39.02	24.10	0.23	0.01
eggplant	0.17	1.43	40.55	176.50	20.25	116.50	27.53	0.00	0.00
tomato	0.01	1.94	14.93	179.80	15.32	46.20	39.99	0.03	0.01
MAX1	12.86	3.83	284.75	4272.50	49.44	1132.00	39.99	20.52	3.04
MIN2	0.00	0.74	14.93	176.50	14.67	39.02	4.54	0.00	0.00
AVG	1.30	1.89	93.31	892.61	28.17	216.95	21.04	2.15	0.30
St-DV	3.66	0.93	76.45	1185.55	11.69	300.69	9.75	5.87	0.87
WHO-ML	0.10	0.30	500.00	425.00	-	100.00	73.00	67.00	50.00

The samples were then brought in plastic bags to laboratory and they were cleaned with deionized water repeatedly. These were later dried in an oven at 65°C for about 2 days and were ground there using a Cross beater Grinding mill. After then, 0.5 g of ground plant sample is digested with 5 ml of nitric acid and 3 ml of hydrogen peroxide. The digestion temperature was about 160°C. The samples were then analyzed with the highly sensitive ICP-MS (Inductively coupled plasma mass spectrophotometer). Guidelines for maximum limit (ML) of metals in vegetables was adopted from FAO-WHO [37-38] (Table 2).

Water Sampling, Handling and Analysis: Surface composite samples of water were taken where the rivers are diverted to vegetable farms. About 5ml of HNO₃ acid was added to clean 250ml polyethylene bottles, before adding about 100 of river water. The HNO₃ acid was added for the purpose of acidifying and preserving the water samples. The chemical composition of the water was determined at the laboratory of the Soil and Water Research Institute. Guidelines for maximum limits of metals in irrigation water were summarized (Table 1).

Soil Sampling, Processing and Analysis: Composite surface soil (0-20cm) samples (from a bulk soil made up of 20 different soil samples per farm) of the vegetables farms were collected separately and properly labeled. The soil samples were then air-dried and crushed to pass a 2-mm mesh sieve. The guidelines used in this text for maximum

levels (ML) of metals in soils was adopted from the reference by Ewers *et al.* [39] and for Fe and Mn according Pendias *et al.*[40] and Ayers *et al.*[41] (Table 1).

RESULTS

Concentration of some trace elements found in soils and Firoozabad River were summarized in Table1.

Water analyses revealed that Firoozabad River is contaminated with Ni, Cd, Cu and Pb (Table 1). Data showed that the major industries from which effluents enter Firoozabad River are: Sahre Rey Tannery, Painting Factory, Soap Factory, Melting Industry and etc. The daily domestic and industrial wastewater discharge from the above industries such as wastes from garages, gas stations, hospitals, etc. are discharged into these river this is consistent with plant content of the same elements in the leafy vegetables grown in farms irrigated with these rivers. Farms which irrigated by this water were contaminated, because more industrial effluents from various industrial sources enter into River. Additionally, little or no treatment is applied to the industrial discharges to detoxify the wastewater draining into rivers. Unfortunately, metals emitted in such manner are easily transferred to all of the food chain, thereby affecting human and animal health. Comparing the metal concentration in soil with Guidelines for soils showed that all metal concentration is below the Guidelines for soils but for Cd, Pb, Cu and Zn there were a tent to reach. Concentration of some trace elements(mg kg⁻¹) found

in some vegetables under waste water irrigation that summarized in Table 2 also showed that concentration of Cd,Pb,Zn and Fe in some vegetables is grater than maximum permitted level that extended by WHO.

DISCUSSION

1. Lead: The main cause for concern in terms of contamination of vegetables in Shahre Rey by heavy metals relates to Lead (Pb). Although a maximum Pb limit for human health has been established for edible parts of crops in China is 0.2 mg kg^{-1} [42] but this limit by WHO standards is 0.3 mg kg^{-1} [37]. Data showed that in all vegetables, lead concentration is more than permitted level, so they are not suitable for consumption. Lead is a toxic element that can be harmful to plants, although plants usually show ability to accumulate large amounts of lead without visible changes in their appearance or yield. In many plants, Pb accumulation can exceed several hundred times the threshold of maximum level permissible for human [43]. The introduction of Pb into the food chain may affect human health and thus, studies concerning Pb accumulation in vegetables have increasing importance [44]. On the whole, all vegetables that were studied in this study, were contaminated by lead and they were toxic to consumer.

2. Zinc: Maximum Zn tolerance for human health has been established for edible parts of crops is 20 mg kg^{-1} by Chinese Department of Preventive Medicine [45]. WHO [37] maximum permitted level for Zinc in vegetables is 100 mg kg^{-1} . By this way, the concentration of Zn in vegetables was as fallow:

Green Pepper>Spinach>Dill>Mint>Celery>Eggplant>Cress. Data showed that there were not any pollution in Radish, Coriander and tomato in compare to WHO standard level. Knowledge of Zn toxicity in humans is minimal. The most important information reported is its interference with Cu metabolism [46-47]. The symptoms that an acute oral Zn dose may include: tachycardia, vascular shock, dyspeptic nausea, vomiting, diarrhea, pancreatic is and damage of hepatic parenchyma [48]. Vegetables that growing on heavy metal contaminated soils can accumulate high concentrations of Zinc to cause serious health risk to consumers. Research results about the effects of excess Zinc on plant growth of three selected vegetables cabbage, celery and Spinach showed that excess Zn in growth media caused toxicity to all three vegetable crops [49].

3. Cadmium: Data in Table 2 showed that Cd concentration mostly were appeared in leafy vegetables and were in fallow order: Radish leaves>Cress>Radish roots>Dill>Spinach>eggplant. WHO standards for Cd in vegetables are 0.1 mg kg^{-1} [37-38].

Similar results are in line with those obtained by Doyle [14], Torabian and Mahjouri [33], Giordano and Mays [50] and Fazeli [51]. Giordano and Mays [50] showed that among different vegetables, the highest amounts of Cadmium accumulation is in lettuce, spinach and radish. Shariati and Farshi [52] also showed that the levels of Cadmium accumulation are in a descending order in lettuce, radish and spinach, respectively. Torabitni and Mahjouii [33] show that Cadmium accumulation in plants irrigated with wastewaters in South Tehran is in the following order of ranking. Spinach<Radish<Coriander<Mint

The amount of Cadmium accumulation in the aerial parts of a plant is higher than in the parts below the ground (root). This finding is comparable with the findings of other earlier studies [14, 51, 53]. They has been reported that cadmium is a highly mobile metal, easily absorbed by the plants through root surface and moves to wood tissue and transfers to upper parts of plants. Gardiner *et al.* [54] and Ramos *et al.* [55] showed that there is a direct relation between the levels of presence of Cadmium in the root zone end its absorption by plant.

4. Copper: The copper levels found in vegetables were within safe limits in all samples. Yang *et al.* [56] studied the response of three vegetables to Cu toxicity and found that Cu levels in both root and shoot increased, but root Cu concentration increased more sharply than shoot with increasing Cu levels in growth media. Cu mainly accumulated in roots while a small fraction (10%) of absorbed Cu was transported to shoot. Xiong and Wang [57] found that Cu concentration in the shoots was significantly influenced by Cu concentration in soil and increased markedly with an increase in the soil Cu concentration.

Conclusions and Recommendations: Heavy metal depositions are associated with a wide range of sources such as small scale industries (including battery production, metal products, metal smelting & Cable coating industries); brick kilns; vehicular emissions; re-suspended road dust and diesel generator sets. These can all be important contributors to the contamination found in vegetables. Additional potential sources of heavy metals in field locations in urban and per urban areas include irrigation water contaminated by industrial

effluent leading to contaminated soils and vegetables. To avoid entrance of metals into the food chain, municipal or industrial waste should not be drained into rivers and farmlands without prior treatment. Apart from treating the discharge that enters into the farms, it is also imperative to utilize alternative measures of cleaning up the already contaminated substrates. Although there is a general tolerable level of metals in vegetables from Sahre Rey at the moment, there are exceptional cases of metal build up such as Cadmium and Lead in feature. The daily intake of these metals at present is much less than concentrations that affect health, the situation could however change in the future depending on the dietary pattern of the community and the volume of contaminants added to the ecosystems. Data showed that genotypic differences in tolerance and co tolerance to heavy metals are well known in some species and ecotypes of natural vegetation. Leafy vegetables have been shown to accumulate relatively high concentrations of heavy metals in compare to fruit vegetables. Perhaps the most important conclusion that may be drawn from the findings of the study, is that since vegetables tend to absorb and accumulate Cadmium in the stem and leaves, the must consumed parts of the plants and in view of their important role in the food chain, it is recommended that these type of plants should not be cultivated in farms and fields irrigated by urban and industrial waste water or water contaminated by heavy metals.

ACKNOWLEDGEMENT

The Tehran Jehade Agriculture Organization is acknowledged for the financial support during research. Thanks are extended to plant production deputy office of ministry of Agriculture. Khak Azma Negin soil test laboratory are acknowledged for the soil, plant and water analysis.

REFERENCES

1. Abdola, M. and J. Chmelnicka, 1990. New Aspects on the Distribution and Metabolism of Essential Trace Elements after Dietary Exposure to Toxic Metals. *Biol. Trace Element Res.*, 23: 25-53.
2. Thompson, HC. and WC. Kelly, 2003. *Vegetable Crops*. 5th Edn. New Delhi: McGraw Hill Publishing Company Ltd; 199067. Türkdogan MK, Kilicel F, Kara K, Tuncer I, Uygan I. Heavy metals in soil, vegetables and fruit in the endemic upper gastrointestinal cancer region of Turkey. *Environ Toxicol Pharmacol.*, 13(3): 175-179.
3. Damek-Poprawa, M. and K. Sawicka-Kapusta, 2003. Damage to liver, kidney and teats with reference to burden of heavy metals in yellow-necked mice from areas around steelworks and zinc smelters in Poland. *Toxicol.*, 186(1-2): 147-158.
4. Koenig, W., J. Leistner and C. Prange, 1990. Soil examination in vegetable gardens on hazardous-waste sites. *Umwelt*, 20: 105-106.
5. Mench, M., J. Vangronsveld, V. Didier and H. Clijsters, 1994. Evaluation of metal mobility, plant availability and immobilization by chemical agents in a limed-silty soil. *Environ. Pollut.*, 86: 279-286. [ISI][Medline]
6. Manz, M., 1995. Environmental pollution by arsenic and heavy metals in soils, dumps, plants and slags of former mining regions of the central and southern Schwarzwald. (In German.) *Karlsruher Geochem. Hefte*, 7: 235-245.
7. Wegelin, T., B. Jenka and R. Etter, 1995. Heavy metals in soils and plants of allotment gardens in the city of Zurich (Switzerland). *Soil Environ.*, 5(1): 269-270.
8. Gimmler, H., B. Degenhardt, S. Lang and C. Track, 1998. Uptake of sodium, boron and heavy metals from an alkaline top layer and translocation to leaves and lower roots of *Vitis vinifera* during a long time duplex study. *J. Appl. Bot.*, 72: 191-202. [ISI].
9. McLaughlin, M.J., N.A. Maier, R.L. Corell, M.K. Smart and C.D. Grant, 1998. In-situ immobilization techniques to remediate cadmium-contaminated agricultural soils. pp: 253-460. *In Proc. 6th Int. Conf. on Contaminated Soils*, Edinburgh, UK. 17-21 May 1998. Telford, London.
10. Meuser, H., U. Schleuss, H. Taubner and Q. Wu. 1998. Soil properties of coal, iron and steel industrial sites in Essen, Germany. *Z. Pflanzenernaehr. Bodenkd.* 161: 197-203.
11. Trautmannsheimer, M., P. Schramel, R. Winkler and K. Bunzl, 1998. Chemical fractionation of some natural radionuclides in a soil contaminated by slags. *Environ. Sci. Technol.*, 32: 238-243. [ISI]
12. Tossavainen, M. and E. Frossberg, 1999. The potential leachability from natural road construction materials. *Sci. Total Environ.*, 239: 31-47. [ISI]
13. Carr, R., 2005. WHO guidelines for safe waste water use: More than just numbers. *J. Iris. Drain*, 54: 103-111.
14. Doyle, P.J., 1998. Survey of literature and experience on the disposal of sewage on land. Available from: <http://www.ecobody.com/reports/sludje/dole-report-vptoc.htm>.

15. Nrgholi, B., 2007. Investigation of the Firozabad wastewater quality-quantity variation for agricultural use. Final report. Iranian Agril. Eng. Res. Institute.
16. Msaky, J.J. and R. Calvert, 1990. Adsorption behavior of copper and zinc in soils: influence of pH on adsorption characteristics. *Soil Sci.*, 150(2): 513-522.
17. Fergusson, J.E., 1990. *The Heavy Elements: Chemistry, Environmental Impact and Health Effects*. Oxford Press, pp: 382-399.
18. Ellen, G., J.W. Loon and K. Tolsma, 1990. Heavy metals in vegetables grown in the Netherlands and in domestic and imported fruits. *Z Lebensm Unters Forsch*, 190(1): 34-39. doi: 10.1007/BF01188261. [PubMed]
19. Yargholi, B. and A.A. Azimi, 2008. Investigation of Cadmium absorption and accumulation in different parts of some vegetables. *American Eurasian J. Agric and Environ. Sci.*, 3(3): 357-364.
20. Zurera-Cosano, G., R. Moreno-Rojas, J. Salmeron-Egea and R. Pozo Lora, 1984. Heavy metal uptake from greenhouse border soils for edible vegetables. *J. Sci. Food Agric.*, 49(3): 307-314.
21. Chronopoulos, J., C. Haidouti, A. Chronopoulou-Sereli and I. Massas, 1997. Variations in plant and soil lead and cadmium content in urban parks in Athens, Greece. *Sci. Total Environ.*, 196(1): 91-98. doi: 10.1016/S0048-9697(96)05415-0.
22. Sánchez-Camazano, M., M.J. Sánchez-Martín and L.F. Lorenzo, 1994. Lead and cadmium in soils and vegetables from urban gardens of Salamanca (Spain). *Sci. Total Environ.*, 146/147: 163-168.
23. Sterrett, S.B., R.L. Chaney and C.H. Gifford, 1996. Influence of fertilizer and sewage sludge compost on yield of heavy metal accumulation by lettuce grown in urban soils. *Environ Geochem Health*, 18(4): 135-142. doi: 10.1007/BF01771236.
24. Van Lune, P., 1987. Cadmium and lead in soils and crops from allotment gardens in the Netherlands. *Neth. J. Agric. Sci.*, 35: 207-210.
25. Wong, J.W.C., 1996. Heavy metal contents in vegetables and market garden soils in Hong Kong. *Environ Technol.*, 17(4): 407-414.
26. Cambra, K., T. Martínez, A. Urzelai and E. Alonso, 1999. Risk analysis of a farm area near a lead- and cadmium-contaminated industrial site. *J. Soil Contam.*, 8(5): 527-540.
27. Dudka, S. and W.P. Miller, 1999. Permissible concentrations of arsenic and lead in soils based on risk assessment. *Water Air Soil Poll.*, 113(1/4): 127-132.
28. Hawley, J.K., 1985. Assessment of health risk from exposure to contaminated soil. *Risk Anal.*, 5(4): 289-302.
29. DEFRA (Department of Environment, Food and Rural Affairs). 1999. *Total Diet Study-Aluminium, Arsenic, Cadmium, Chromium, Copper, Lead, Mercury, Nickel, Selenium, Tin and Zinc*. London: The Stationery Office.
30. Ni, W.Z. and XX. Long, 2002. Studies on the criteria of cadmium pollution in growth media of vegetable crops based on the hygienic limit of cadmium in food. *J. Plant*.
31. Mostachari, M., 2002. Investigation of Qazvin soils and plants pollution with heavy metals during irrigation with waste water. *Proceeding of 75 Water and Soil Conference*, (In Persian).
32. Jafarzadeh, N., 1997. Assessment the wastewater use effects in Shiraz on heavy metals concentration on soil and plants. *Proceeding of 6th Water and Soil Conference* (In Persian).
33. Torabian, A. and M. Mahjouri, 2002. Heavy metals uptake by vegetable crops irrigated with waste water in south Tehran *Journal of Environmental Study*. Volume 16, Number 2 (In Persian).
34. Chaney, R.I., 1980. Health risks associated with toxic metals in municipal sludge. 14th Health risks of land application. G.L. Damron, pp: 59-84. *Ann. Arbor. Science Publication. Inc.*
35. Nriagu, J.O., 1989. A global assessment of natural sources of atmospheric trace metals. *Nature*, 338(6210): 47-49.
36. Bilos, C., J.C. Colombo, C.N. Skorupka and M.J. Rodriguez, 2001. Source, distribution and variability of airborne trace metals in La Plata City area, Argentina. *Environ. Pollut.*, 111(1): 124-132.
37. Codex Alimentarius Commission (FAO/WHO). *Food additives and contaminants. Joint FAO/WHO Food Standards Program 2001; ALINORM 01/12A:1-289*.
38. CODEX ALIMENTARIUS COMMISSION: CX/FAC 95/19. Nov. 1994. *Position Paper on Cadmium*. 27. March 1995.
39. Ewers, U., 1991 *Standards, guidelines and legislative regulations concerning metals and their compounds*. In: Merian E, ed. *Metals and Their Compounds in the Environment: Occurrence, Analysis and Biological Relevance*. Weinheim: VCH, pp: 458-468.
40. Pendias, A.K. and H. Pendias, 1992. *Elements of Group VIII*. In: *Trace Elements in Soils and Plants*. Boca Raton: CRC Press, pp: 271-276.

41. Ayers, R.S. and D.W. Westcott, 1985. Water quality for agriculture. FAO Irrigation and Drainage Paper, 29(1): 1-120.
42. Chinese Department of Preventive Medicine, 1994 *Threshold for Food Hygiene*. Beijing: China Standard Press (In Chinese).
43. Wierzbicka, M., 1995. How lead loses its toxicity to plants. *Acta Soc. Bot. Pol.*, 64: 81-90.
44. Coutate, T.P., 1992. Food, the Chemistry of Its Component. 2nd Edn. Cambridge: Royal Society of Chemistry, pp: 265.
45. Chinese Department of Preventive Medicine, 1995. *Threshold for Food Hygiene*. Beijing: China Standard Press (In Chinese).
46. Barone, A., O. Ebesh and R.G. Harper, 1998. Placental copper transport in rats: Effects of elevated dietary zinc on fetal copper, iron and metallothionien. *J. Nutr.*, 128(6): 1037-1041.
47. Gyorffy, E.J. and H. Chan, 1992. Copper deficiency and mycrocytic anemia resulting from prolonged ingestion of over-the-counter zinc. *Am. J. Gastroenterol.*, 87: 1054-1055. [PubMed].
48. Salgueiro, M., J. Zubillaga, M. Lysionek, A. Sarabia, M.I. Caro and R. Paoli, 2000. J. Zinc as an essential micronutrient: A review. *Nutr. Res.*, 20(5): 737-755.
49. Long, X.X., X.E. Yang, W.Z. Ni, Z.Q. Ye, Z.L. He, D.V. Calvert and J.P. Stoffella, 2003. Assessing zinc thresholds for phytotoxicity and potential dietary toxicity in selected vegetable crops. *Common. Soil Sci. Plant Anal.*, 34(9 and 10): 1421-1434.
50. Giordano, P.M. and D.A. Mays, 1977. Yield and heavy metal content of several vegetable species grown in soil amended with sewage sludge hi lbiokical Implications of Heavy Metals in the Environment. ERDA Rep. Conf, Oak. Ridge, Tennessee.
51. Fazeli, M.S., 1998. Enrichment of heavy metal in paddy crops irrigated by paper mill effluents near Nanjangud, Mysore District. Karnatuke, India *Environmental. Geology*, 34: 42-54.
52. Shariati, M. and S. Farshi, 1997, Heavy metal accumulation in south Tehran vegetable crops. *J. Soil and Water*, 5: 3 (In Persian).
53. Sanita di Toppi, L. and R. Gabbrielli, 1999. Response to cadmium in higher plants.review. *J. Env. and Exp. Bot.*, 41: 105-130.
54. Gardiner, D.T., R.W. Miller, B. Badamchian, AS. Azan and D.R. Sisson, 1995. Effects of repeated savage sludge applications on plant accumulation of heavy metals *J. Agri. Ecosyst. Env.*, 55: 1-6.
55. Rarnos, I., E. Esteban, J.J. Lucena and A. Garate, 2002. Cadmium uptake and subellular distribution in plants of lactuca ap. Ca-Mn interaction. *J. Plant Sci.*, 162: 761-767.
56. Yang, X.E., XX. Long and W.Z. Ni, 2002. Assessing copper thresholds for phytotoxicity and potential dietary toxicity in selected vegetables crops. *J. Environ. Sci. Health*, B37(6): 625-635.
57. Xiong, Z.T. and H. Wang, 2005. Copper toxicity and bioaccumulation in Chinese cabbage (*Brassica pekinensis* Rupr.). *Environ. Toxicol.*, 20(2): 188-194. *Nutr.*, 25(5): 957-968.