Determination of the Cadmium, Chromium, Nickel, Lead and Cobalt in Selected vegetables and their transfer and translocation factor

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Abstract: The heavy metals pollution is one of the problems that arise because of the increased uses of fertilizers and other chemicals to fulfil the higher demands of food production for human consumption. In the present study different methods, for assessment of the heavy metals concentration in the human body contributed by contaminated vegetables, are discussed. The concentration of Cr, Co, Cd, Pb and Ni in the soils was ranged from 0.11-0.231, 5.151-6.662, 3.2-4.32, 7.19-8.79, 7.16-7.72mg/kg with mean value ($p = 0.05$) of 0.146 ± 0.046, 5.731±0.527, 3.803±0.503, 7.770±0.504, 7.435±0.504mg/kg, respectively. The contamination, sources and bioaccumulation of the heavy metals i.e. Cr, Cu, Pb, Cd and Ni in the vegetables are described.

Key words: Heavy metals, Vegetables, Transfer factor, Translocation factor

1.Introduction: The safety of food is a key global societal concern. Researchers have become more aware of the problems connected with eating contaminated foods, such as pesticides, heavy metals, or toxins in vegetables, as a result of rising demands for food and food safety [1, 2].

Heavy metals contamination is a major problem of our environment and they are also one of the major contaminating agents of our food supply [3, 4]. This problem is receiving more and more attention all over the world, in general and in developing countries in particular. The biological half-lives of these heavy metals are long and have possibility to accumulate in different body organs and thus produce unwanted side effects [5-7]. Lead and Cadmium are the most toxic and the most abundant metals in food.

Excessive accumulation of the heavy metals in human bodies creates the problems like cardiovascular, kidney, nervous and bone diseases [8-10]. It is known that serious systemic problems can develop as a result of increased accumulation of dietary heavy metals such as cadmium and lead in the human body [11]. Heavy metals [12] are exceedingly persistent in the environment, are not biodegradable, and are not thermodegradable. As a result, their buildup quickly reaches dangerous levels.

Heavy metals can impair important biochemical systems, constituting an important threat for the health of plants and animals. The adverse health effects of several chemical elements have been documented throughout history: Greeks and Roman physicians were able to recognize symptoms of acute lead and arsenic poisoning long before toxicology became a science. Currently, the advances of toxicology has improved our knowledge but human exposure to toxic elements and their health effects, such as developmental retardation, several types of cancer, kidney damage, endocrine disruption, immunological disorders (autoimmunity) and then death.

Significant contamination of seeds, plants and plant products with toxic chemical elements due to contaminated soil and water has been observed as result of release of these toxicants into the sea, rivers, lakes and even into irrigation channels. Following that, consuming infected vegetables is a significant way for both animals and people to become exposed.

Vegetable cultivation has a long history in cities and their surrounding areas [13]. Realise that the majority of these agricultural lands are heavily polluted with heavy metals, primarily from vehicle emissions, pesticides and fertilisers, industrial effluents, and other anthropogenic activities. These poisoned soils have caused infected foods to grow [14-16].

Because heavy metals in soil interfere with plants' metabolic activities, the production of vegetables is decreased [17, 18]. According to Singh and Kumar's 2006 report, the heavy metals Cu, Cd, Pb, and Zn significantly pollute the soil, irrigation water, and select vegetables from peri-urban settings [19]. Additionally, it was shown that Cd and Pb posed greater dangers than Cu and Zn.

The acceptable limits of these heavy metals are routinely lowered in these vegetables since they not only damage the nutritional value of vegetables but also human health. Regulatory authorities at the national and international levels are in charge of this regulation [20].

Most of the time, soils have higher heavy metal contents than vegetables cultivated in those soils. This shows that the root serves as a barrier to the transfer of heavy metals within plants, and that only a limited amount of soil's heavy metals are transported to the veggies [21].

The effects of heavy metal contamination varied depending on the use of fertilisers and other human activities at each location. According to research, phosphate fertilisers are the primary cause of soil pollution from heavy metals. The presence of cadmium as an impurity in phosphate rocks is the cause of this pollution. The widespread usage of artificial fertilisers has led to the poisoning of the soil with heavy metals [22].

The majority of heavy metals are naturally occurring components of the earth's crust, where they are absorbed by plants and subsequently moved up the food chain. The levels of these metals differ from one soil to another. Vegetables' metal content is mostly influenced by the texture of the soil or medium in which they are grown, but plant type and nature also have a role [23].

The main source of human exposure to heavy metals is the soil-to-plant transfer quotient since vegetables are the primary food source for humans. The transfer quotient is a useful tool for calculating the relative differences in metals' bioavailability to plants. Therefore, the transfer quotient for the human Health Risk Index (HRI) should be evaluated [24]. Cd and Cu had a greater transfer quotient than Pb and Fe, two additional metals [25].

The stronger accumulation of the relevant metal by that vegetable is shown by the larger transfer quotient of heavy metal. A transfer quotient of 0.1 means that the element is being excluded from the plant's tissues [26]. Vegetables will be more likely to be contaminated with metal by anthropogenic activity if the transfer coefficient value is higher than 0.50, necessitating environmental monitoring of the area [27].

According to current research investigations, a transfer value of 0.2 suggests that leafy plants are more anthropogenically contaminated than a transfer coefficient of 0.50 [28, 29]. They claimed that Kloke et al. provided a generalised transfer coefficient based on the uptake of metals by plant roots. In comparison to other vegetables, leafy plants collect substantially higher levels of heavy metals. This is due to the fact that leafy vegetables have a higher translocation and transpiration rate than other vegetables, which causes the accumulation of metals to be lower than that of leafy vegetables [30].

The pollution index of soils and sludges has been widely utilised by researchers to identify element contamination that has raised element toxicity overall [31]. Although the outcomes vary from one location and researcher to another, the fundamental idea stays the same. The average ratio of the metal concentration in the sample to the indicated tolerable/permissible values of soil for plant growth is used to construct the pollution index [32].

2. Material and methodology

The detailed methodology used for the study is given in this chapter in subsequent sections:

2.1 Study area

2.2 Sample collection

2.3 Sample processing

2.4 Sample analysis

2.5 Data analysis

2.1 Study area

In the present study soil and plant samples were collected from Haryana (India). The state of Haryana is in the north-west part of India. Though Haryana lies in the sub-tropical belt still climate is Arid to Semiarid.

For the process of soil and crop sampling, area of a field is selected from Hisar district. There is total 268 *villages* in *Hisar district*. The village selected for sampling is not the heavy metal prone area but for study, the heavy metal uptake by crops from a normal soil and the influence of soil parameters will be a significant part of study. Vegetables cultivated in the area in winter and summer seasons includes *Solanum melongena* (Brinjal) and *Lagenaria siceraria* (Bootlegourd/ Lauki). Winter vegetables are sown during months of October-November and harvested at the end of December-January and summer vegetables are sown in the month of June to July and harvested by August-September.

2.2 Sample collection

In order to evaluate the transfer and translocation factor of heavy metals in food crops and corresponding soils were collected in their respective harvesting seasons. The details of the samples collected are given below:

1. *Lagenaria siceraria* (Bootlegourd/ Lokki): Samples were collected during its harvesting season (August-September) for the year (2021). A total of 10 samples were collected each weighing 500 - 1000g. The collected samples were cleaned, sealed and stored as discussed above.

2. *Solanum melongena* (Brinjal): Samples of vegetable from identified sampling stations were collected during harvesting season (Dec-Jan) for the year (2021). A total of 10 samples were collected each weighing 500 - 1000g. After removal of extraneous matter like soil pebbles, stones and other debris the samples were sealed in plastic containers, taken to the laboratory and stored till further analysis.

3. Soil sample: In order to evaluate transfer and translocation factor of heavy metals from soil to above collected different environmental matrices, corresponding sub surface soil samples from rooting zone were also collected. A total of 40 soil samples were collected during different sampling seasons. From a depth of 5-10 cm, four sub-samples of soil from rectangular grid of 0.5 m2 area were collected and then mixed together to obtain a representative sample. The soil samples each weighing 500g were collected using non-metallic spade so as to get rid of the metal contamination. Samples were sealed in clean plastic bags after removing foreign bodies. Soil samples were air dried, grounded and then passed through a 2.0 mm sieve and stored in air tight containers till further analysis.

2.3 Sample processing

Soil Samples Processing

Soil samples were air dried, grounded and stored in air-tight plastic containers for analysis. After removal of extraneous matter like pebbles and plant roots etc. Soil samples were oven-dried at a temperature of 110°C until a constant weight of the sample was achieved. Then the samples were ground in grinder with care to avoid cross contamination. After milling, the whole samples were sieved using 2.0 mm sieve.

For heavy metals analysis, processed samples were stored in air tight plastic containers to have moisture free conditions till further analysis.

Agricultural Sample Processing

Vegetable Samples Processing

- The fresh weight of the samples was recorded just after collection.
- Different parts (roots, shoots, fruits) of vegetables were dried in drying oven at 110°C temperature until a constant weight of the sample was achieved.

- After drying, the dry weight of the sample was recorded and about 100 g dry subsamples were ashed in a muffle furnace at 350-400°C till the ash became white.

- The ashed samples were then stored in moisture free conditions till further analysis

2.4 Sample analysis

Heavy Metals Analysis : following steps are involved in this process.

Sample Digestion:

Total Cd, Ni, Cr, Co and Pb contents of agricultural vegetables and soil samples were determined using Atomic Absorption Spectrophotometer (SenSAAGBC, Australia) and concentrations were finally expressed in mg kg-1 on dry wt. basis. 0.2 g of each agricultural crop ash sample was mixed with 5.0 ml diacid (HNO_3 and $HClO_4$) mixture in the ratio of 9:1 (v/v). Similarly, 0.5g of each soil sample was mixed with 5.0 ml diacid mixture. Sample mixture was then taken in pre-cleaned Teflon vessels, left open overnight at room temperature and digested on hot plate. When digestion was complete, vessels were cooled at room temperature and digests were quantitatively transferred into glass beakers. Then the digests were evaporated to dryness on hot plate at a temperature of 130-150°C and residue was dissolved in double distilled water to make desired volume (50 ml). Extracted solutions were transferred to polypropylene bottles and refrigerated until analysis.

All working standards used for analysis were prepared by diluting 1000 mg/L certified standard solutions. Acetylene gas was used as fuel and air as support in Atomic Absorption Spectrophotometer (AAS). An oxidizing flame was used in all the cases except chromium, where reducing nitrous oxide flame was used for metal quantification. The extracts were analyzed for eight heavy metals, viz. Cd, Ni, Cr, Co and Pb using Atomic Absorption Spectrophotometer (SenSAAGBC, Australia). During processing and analysis of samples, quality assurance and control measures were adopted to ensure the reliability of results. To avoid sample contamination double distilled water was used for rinsing glassware, preparation of standards and dilution of samples. Reagent blanks were analysed and the data were subsequently blank corrected to remove analytical bias. Relative standard deviations (RSDs) of replicate measurements were 0.999. To ensure the reliability of results, standards of respective metals were run after every 10 samples analysed.

2.5 Transfer/ Translocation Factor of Heavy Metals from Soil to Agricultural Vegetables Samples

To evaluate the heavy metals‟ uptake efficiency by edible portions of agricultural samples, Transfer factors (TF) was calculated. It is computed as the ratio of concentration of the heavy metals in edible portion of agricultural samples to their concentration in respective soils. (Singh et al., 2014).

$$\text{TF Edible} = C_{crop} / C_{soil}$$

where, C_{crop} and C_{soil} are the concentration of heavy metal in edible portion of agricultural samples (grain, fodder and vegetables) and rooted soils on dry weight (DW) basis, respectively.

Translocation factor (TF) is defined as the ratio of heavy metals from soil to different parts of the crop.

$$\text{TF} = C_{parts}\,(\text{root, shoot, fruits}) / C_{soil}$$

3. Result and discussion

3.1 Concentration of Cr, Cd, Ni, Pb and Co in plant and soil samples: Results of assessment of Cr, Cd, Ni, Pb and Co in parts of plants are outcome of soil contamination and later, the content of such toxic materials in vegetation including field crops. As per the heavy metal content, high requirements are posed especially on productive parts of plants that are used in human nutrition as plant products, raw-materials of food industry, forage crops, from which heavy metals could be transferred into different products.

The evaluation of contaminating heavy metals is a serious problem and is expanding. According to Food Codex heavy metals as pollutants can translocate into food materials from soil (via roots) and air (mostly via leaves), in some cases also from equipment, etc [33], [34]. Soil acts as a sink of heavy metals, from which they are transferred into plant products and through food chain into animal products. The metal concentration in soil was thus detected to be far more than concentration in plant parts. Similar observations were made by Nwajei G.E et al during their study on tomato fruits and leaves [35].

3.2 Concentration of heavy metals in roots of two plants: The range of heavy metal content in root of bottle gourd collected during August - September, 2021 were: (Co) 2.403 – 3.397 mg kg^{-1} , (Cd) 2.212 - 2.530 mg kg^{-1}, (Ni) 3.157 – 4.14 mg kg^{-1} , (Pb) 2.422 – 3.674 mg kg^{-1} and (Cr) 0.004 - 0.032 mg kg^{-1} with mean value of heavy metal content (mg kg^{-1}) was in the following order: (Ni) 3.583±0.334 > (Pb) 3.345 ± 0.395 > (Co) 2.769 ± 0.366 > (Cd) 2.298 ± 0.117 > (Cr) 0.009 ± 0.019 (Table 1). From the results of the present study, it was observed that content of Nickel was highest and chromium was lowest in the root of the bottle gourd vegetable.

S. No.	Heavy Metals	Mean
1	Ni	3.583±0.344
2	Pb	3.345 ± 0.395
3	Cd	2.298 ± 0.117
4	Cr	0.009 ± 0.019
5	Co	2.769 ± 0.366

Table 1 Concentration of heavy metals (mg kg^{-1}) in root of *Lagenaria siceraria*

The range of heavy metal content in root of brinjal collected during Jan-Feb, 2022 were: (Co) 2.121 – 3.386 mg kg^{-1} , (Cd) 2.231 - 2.533 mg kg^{-1}, (Ni) 4.141 – 4.947 mg kg^{-1} , (Pb) 2.877 – 4.599 mg kg^{-1} and (Cr) 0.005 - 0.033 mg kg^{-1} with mean value of heavy metal content (mg kg^{-1}) was in the following order: (Ni) 4.471±0.334 > (Pb) 3.643 ± 0.430 > (Co) 2.738 ± 0.457 > (Cd) 2.366 ± 0.151 > (Cr) 0.022 ± 0.011 (Table 2). From the results of the present study, it was observed that content of Nickel was highest and chromium was lowest in the root of the brinjal vegetable.

S. No.	Heavy Metals	Mean
1	Ni	4.471±0.334
2	Pb	3.643±0.430
3	Cd	2.366±0.151
4	Cr	0.022±0.011
5	Co	2.738±0.457

Table 2 Concentration of heavy metals (mg kg^{-1}) in roots of *Solanum melongena*

3.3 Concentration of heavy metals in stem of two plants: Heavy metals in the shoot samples of bottle gourd collected during August - September, 2021 was: (Pb) 1.55 – 1.978 mg kg^{-1}, (Ni) 1.321 – 3.149 mg kg^{-1}, (Cd) 1.106 – 1.843 mg kg^{-1}, (Cr) 0.001 - 0.002 mg kg^{-1} and (Co) 1.258 – 1.972 mg kg^{-1} (Table 3). Mean value of heavy metal content (mg kg^{-1}) in shoot of bottle gourd collected in the following order: Ni (1.935 ± 0.634) > Pb (1.827 ± 0.154) > Co (1.555 ± 0.272) > Cd (1.350 ± 0.288) > Cr (0.00 ± 0.001) (Table 4.4). It is clear from the above results that Nickel content was higher and chromium content was lowest in the shoot.

S. No.	Heavy Metals	Mean
1	Ni	1.935 ± 0.634
2	Pb	1.827±0.154
3	Cd	1.350±0.288
4	Cr	0.00±0.001
5	Co	1.555±0.272

Table 3 Concentration of heavy metals in shoot of *Lagenaria siceraria*

Heavy metals in the shoot samples of brinjal collected during Jan-Feb, 2022 was: (Pb) 1.245 – 2.225 mg kg^{-1}, (Ni) 1.32 – 4.349 mg kg^{-1}, (Cd) 1.125 – 1.843 mg kg^{-1}, (Cr) 0.001 - 0.002 mg kg^{-1} and (Co) 2.009 – 2.791 mg kg^{-1}. Mean value of heavy metal content (mg kg^{-1}) in shoot of brinjal collected in the following order: Ni (2.609 ± 1.038) > Co (2.468 ± 0.374) > Pb (1.575 ± 0.263) > Cd (1.386 ± 0.221) > Cr (0.00 ± 0.001) (Table 4). It is clear from the above results that Nickel content was higher and chromium content was lowest in the shoot.

S. No.	Heavy Metals	Mean
1	Ni	2.609 ± 1.038
2	Pb	1.575 ± 0.263
3	Cd	1.386 ± 0.221
4	Cr	0.00 ± 0.001
5	Co	2.468 ± 0.374

Table 4. Concentration of heavy metals (mg kg^{-1}) in shoot of *Solanum melongena*

3.4 Concentration of heavy metals in leaf of two plants: Due to their dietary significance, leaves are given utmost attention. In comparison with roots, the leaves of all plant species exhibited lesser concentration. Thus the absorption pattern did not change at elevated metal concentration in soil.

Bottle gourd vegetable samples from ten sampling stations from the study area were collected during harvesting season. All the collected samples were analysed for Pb, Ni, Cd, Cr and Co content. Heavy metals in the samples of brinjal collected during August - September, 2021 was: (Pb) 0.122 - 0.187 mg kg^{-1}, (Ni) 0.014 - 0.04 mg kg^{-1}, (Cd) 0.014 - 0.03 mg kg^{-1}, (Co) 0.016 - 0.054 mg kg-1 and Cr was not measurable (Table 5). Mean value of heavy metal content (mg kg^{-1}) in bottle gourd samples collected was in the following order: Pb (0.146 ± 0.023) > Co (0.036 ± 0.014) > Ni (0.024 ± 0.009) > Cd (0.019 ± 0.004) > Cr (0.00) (Table 4.5). It is clear from the results that the content of Pb was highest and content of Cr was lowest in the above studied vegetable samples.

S. No.	Heavy Metals	Mean
1	Ni	0.024 ± 0.009
2	Pb	0.146 ± 0.023
3	Cd	0.019 ± 0.004
4	Cr	0.00±0.000
5	Co	0.036 ± 0.014

Table 5 Concentration of heavy metals (mg kg^{-1}) in fruit of *Lagenaria siceraria*

Brinjal vegetable samples from ten sampling stations from the study area were collected during brinjal harvesting season. All the collected samples were analysed for Pb, Ni, Cd, Cr and Co content. Heavy metals in the samples of brinjal collected during Jan-Feb, 2022 was: (Pb) 0.125 - 0.345 mg kg^{-1}, (Ni) 0.038 - 0.389 mg kg^{-1}, (Cd) 0.013 - 0.03 mg kg^{-1}, (Co) 0.01 - 0.079) mg kg^{-1} and Cr was not measurable (Table 6). Mean value of heavy metal content (mg kg^{-1}) in brinjal samples collected was in the following order: Pb (0.193 ± 0.071) > Ni (0.184 ± 0.126) > Co (0.044 ± 0.023) > Cd (0.022 ± 0.006) > Cr (0.00). It is clear from the results that the content of Pb was highest and content of Cr was nil in the studied brinjal vegetable samples.

S. No.	Heavy Metals	Mean

1	Ni	0.184 ± 0.126
2	Pb	0.193 ± 0.071
3	Cd	0.022 ± 0.006
4	Cr	0.00±0.000
5	Co	0.044 ± 0.023

Table 6 Concentration of heavy metals (mg kg^{-1}) in fruit of *Solanum melongena*

3.5 Transfer Factor (TF): Metals from soil are absorbed by plant roots and then distributed in various plant tissues. The Transfer Factor (TF), an index used to measure this metal transfer from soil to plant tissues, is used in the study. It is determined as a ratio between the amounts of a particular metal present in plant tissue and soil, both quantities being expressed in the same units [36]. Higher TF values (1) suggest that the plant will absorb more metal from the soil and that it will be more suitable for phytoextraction and phytoremediation [37]. On the contrary, lower values indicate poor response of plants towards metal absorption and the plant can be used for consumption.

Transfer factor of heavy metal from soil to fruit of vegetable samples collected in August - September, 2021 was: Cr (0.00 - 0.00), Pb (8.488 - 9.604), Cd (0.003 - 0.006), Ni (0.001 - 0.010) and Co (0.002 - 0.009) with mean value was as follows: Pb (0.015 ± 0.002) > Co (0.005 ± 0.002) > Cd (0.004 ± 0.001) > Ni (0.003 ± 0.002) > Cr (0.00 ± 0.00) (Table 4.6 and Fig. 4.1-4.5). It is clear from the results that highest transfer factor of Lead and lowest with nil value of Chromium was observed in all the studied samples of bottle gourd vegetable.

S. No.	**Heavy Metals**	**Mean**
1	Ni	0.003±0.002
2	Pb	0.015 ± 0.002
3	Cd	0.004±0.001
4	Cr	00
5	Co	0.005 ± 0.002

S. No.	Heavy Metals	Mean
1	Ni	0.024±0.017
2	Pb	0.243 ± 0.008
3	Cd	0.005±0.002
4	Cr	0
5	Co	0.007±0.004

Transfer factor of heavy metal from soil to fruit of brinjal vegetable samples collected in Jan-Feb, 2022 was: Cr (0.00 - 0.00), Pb (0.014 - 0.039), Cd (0.004 - 0.01), Ni (0.010 - 0.053) and Co (0.001 - 0.014) with mean value was as follows: Pb (0.243 ± 0.008) > Ni (0.024 ± 0.017) > Co (0.007 ± 0.004) > Cd (0.005 ± 0.002) > Cr (0.00 ± 0.00) (Table 7). It is clear from the results that highest transfer factor of Lead and lowest of Chromium was observed in all the studied samples of brinjal vegetable.

Table 7 Transfer factor for metals in *Lagenaria siceraria*

Table 8 Transfer factor for metals in *Solanum melongena*

3.6 Translocation Factor (TrF): The gradual movement of solute (metal ions) from soil to plant roots and from roots via stem to plant leaves is referred as translocation of solute materials. It is determined as the ratio of metal concentration in shoot to its concentration in the root tissues [38]. Translocation Factors (TrF) of the two plant species Bottle gourd, Brinjal were evaluated for metals, Cd, Ni, Cr, Pb and Co and values are mentioned in table 9 & 10. TrF values nearer to zero, indicate increased retention of metals in plant roots with very less movement to above soil plant parts [39].

Table 9 Translocation factor (shoot to fruit) for metals in *Lagenaria siceraria*

S. No.	Heavy Metals	Mean
1	Ni	0.091±0.100
2	Pb	0.120±0.033
3	Cd	0.015±0.004
4	Cr	0
5	Co	0.017±0.009

Table 10. Translocation factor (shoot to fruit) for metals in *Solanum melongena*

S. No.	Heavy Metals	Mean
1	Ni	0.013±0.006
2	Pb	0.072±0.025
3	Cd	0.014±0.005
4	Cr	0
5	Co	0.023±0.009

4. Conclusion: The nickel, chromium, cadmium, lead and cobalt concentration of the two nutritionally important plant varieties namely Bottle gourd (*Lagenaria siceraria*) and Brinjal (*Solanum melongena*) with corresponding soil concentration were determined in soil. Study was significant because of the dietary importance of the the target vegetables. Subsequently, the transfer factors (TF) and translocation factors (TrF) were determined in order to draw conclusions. It was seen that all studied metals were absorbed readily except chromium by two plant varieties. However, TF values did not approach 1. More specifically the values were found closer to 0. This proved that all these plants are not hyper-accumulator species. However, root to shoot transfer was moderate.The comparison with international as well as Indian permissible standards shows that soil metal concentration remains in correlation with plant metal concentration. At higher levels of soil metal concentration, plant metal concentration may exceed permissible limits causing toxicity in living organisms.

Refrences:

1. Khan, S., Farooq, R., Shahbaz, S., Khan, M. A., & Sadique, M. (2009). Health risk assessment of heavy metals for population via consumption of vegetables. *World Appl Sci J*, *6*(12), 1602-1606.
2. Kormoker, T., Proshad, R., Islam, M. S., Shamsuzzoha, M., Akter, A., & Tusher, T. R. (2021). Concentrations, source apportionment and potential health risk of toxic metals in foodstuffs of Bangladesh. *Toxin reviews*, *40*(4), 1447-1460.
3. Sardar, K., Ali, S., Hameed, S., Afzal, S., Fatima, S., Shakoor, M. B., ... & Tauqeer, H. M. (2013). Heavy metals contamination and what are the impacts on living organisms. *Greener Journal of Environmental management and public safety*, *2*(4), 172-179.
4. Jamal, Q., Durani, P., Khan, K., Munir, S., Hussain, S., Munir, K., & Anees, M. (2013). Heavy metals accumulation and their toxic effects. *Journal of Bio-Molecular Sciences (JBMS)*, *1*(1), 27-36.
5. Sathawara, N. G., Parikh, D. J., & Agarwal, Y. K. (2004). Essential heavy metals in environmental samples from Western India. *Bulletin of Environmental Contamination & Toxicology*, *73*(4).
6. Singh, S. P., & Singh, M. K. (2020). Soil pollution and human health. *Plant Responses to Soil Pollution*, 205-220.
7. Mudgal, V., Madaan, N., Mudgal, A., Singh, R. B., & Mishra, S. (2010). Effect of toxic metals on human health. *The open Nutraceuticals journal*, *3*(1).
8. Baby, J., Raj, J. S., Biby, E. T., Sankarganesh, P., Jeevitha, M. V., Ajisha, S. U., & Rajan, S. S. (2010). Toxic effect of heavy metals on aquatic environment. *International Journal of Biological and Chemical Sciences*, *4*(4).

9. Adekanmi, A. T. (2021). Health Hazards of Toxic and Essential Heavy Metals from the Poultry Waste on Human and Aquatic Organisms. *Animal Feed Science and Nutrition-Production, Health and Environment*.
10. HUSSA ARA BEGUM, M. H., ZAMAN, K., SHINWARI, Z. K., & HUSSAIN, A. (2017). Heavy metal analysis in frequently consumable medicinal plants of Khyber Paktunkhwa, Pakistan. *Pak. J. Bot*, *49*(3), 1155-1160.
11. Khanna, P. (2011). Assessment of heavy metal contamination in different vegetables grown in and around urban areas. *Research journal of environmental toxicology*, *5*(3), 162.
12. Chopra, A. K., Pathak, C., & Prasad, G. (2009). Scenario of heavy metal contamination in agricultural soil and its management. *Journal of Applied and Natural Science*, *1*(1), 99-108.
13. Chen, J., Franklin, J. F., & Spies, T. A. (1995). Growing- season microclimatic gradients from clearcut edges into old- growth Douglas- fir forests. *Ecological applications*, *5*(1), 74-86.
14. Luo, C., Liu, C., Wang, Y., Liu, X., Li, F., Zhang, G., & Li, X. (2011). Heavy metal contamination in soils and vegetables near an e-waste processing site, south China. *Journal of hazardous materials*, *186*(1), 481-490.
15. Luo, C., Liu, C., Wang, Y., Liu, X., Li, F., Zhang, G., & Li, X. (2011). Heavy metal contamination in soils and vegetables near an e-waste processing site, south China. *Journal of hazardous materials*, *186*(1), 481-490.
16. Sandeep, G., Vijayalatha, K. R., & Anitha, T. (2019). Heavy metals and its impact in vegetable crops. *Int J Chem Stud*, *7*(1), 1612-1621.
17. Aderinola, O. J., & Kusemiju, V. (2012). Heavy metals concentration in Garden lettuce (Lactuca sativa L.) grown along Badagry expressway, Lagos, Nigeria. *Transnational Journal of Science and Technology*, *2*(7), 115-130.
18. Noor, I., Sohail, H., Sun, J., Nawaz, M. A., Li, G., Hasanuzzaman, M., & Liu, J. (2022). Heavy metal and metalloid toxicity in horticultural plants: Tolerance mechanism and remediation strategies. *Chemosphere*, 135196.
19. Kumar, M., Antony, J., Singh, R. K., Tiwari, M. K., & Perry, D. (2006). Implementing the Lean Sigma framework in an Indian SME: a case study. *Production Planning and Control*, *17*(4), 407-423.

20. Jordana, J., Levi-Faur, D., & i Marín, X. F. (2011). The global diffusion of regulatory agencies: Channels of transfer and stages of diffusion. *Comparative political studies*, *44*(10), 1343-1369.
21. Shahid, M., Dumat, C., Khalid, S., Schreck, E., Xiong, T., & Niazi, N. K. (2017). Foliar heavy metal uptake, toxicity and detoxification in plants: A comparison of foliar and root metal uptake. *Journal of hazardous materials*, *325*, 36-58.
22. Atafar, Z., Mesdaghinia, A., Nouri, J., Homaee, M., Yunesian, M., Ahmadimoghaddam, M., & Mahvi, A. H. (2010). Effect of fertilizer application on soil heavy metal concentration. *Environmental monitoring and assessment*, *160*, 83-89.
23. Gebrekidan, A., Weldegebriel, Y., Hadera, A., & Van der Bruggen, B. (2013). Toxicological assessment of heavy metals accumulated in vegetables and fruits grown in Ginfel river near Sheba Tannery, Tigray, Northern Ethiopia. *Ecotoxicology and environmental safety*, *95*, 171-178.
24. Rashid, H., Arslan, C., & Khan, S. N. (2018). Wastewater irrigation, its impact on environment and health risk assessment in peri urban areas of Punjab Pakistan-a review. *Environ Contam Rev*, *1*, 30-5.
25. Garg, V. K., Yadav, P., Mor, S., Singh, B., & Pulhani, V. (2014). Heavy metals bioconcentration from soil to vegetables and assessment of health risk caused by their ingestion. *Biological trace element research*, *157*, 256-265.
26. Boularbah, A., Schwartz, C., Bitton, G., Aboudrar, W., Ouhammou, A., & Morel, J. L. (2006). Heavy metal contamination from mining sites in South Morocco: 2. Assessment of metal accumulation and toxicity in plants. *Chemosphere*, *63*(5), 811-817.
27. Khanna, P. (2011). Assessment of heavy metal contamination in different vegetables grown in and around urban areas. *Research journal of environmental toxicology*, *5*(3), 162.
28. Kloke, A., Sauerbeck, D. R., & Vetter, H. (1984). The contamination of plants and soils with heavy metals and the transport of metals in terrestrial food chains. In *Changing Metal Cycles and Human Health: Report of the Dahlem Workshop on Changing Metal Cycles and Human Health, Berlin 1983, March 20–25* (pp. 113-141). Springer Berlin Heidelberg.

29. Kachenko, A. G., & Singh, B. (2006). Heavy metals contamination in vegetables grown in urban and metal smelter contaminated sites in Australia. *Water, air, and soil pollution*, *169*, 101-123.
30. Abbas, M., Parveen, Z., Iqbal, M., Riazuddin, M., Iqbal, S., Ahmed, M., & Bhutto, R. (2010). Monitoring of toxic metals (cadmium, lead, arsenic and mercury) in vegetables of Sindh, Pakistan. *Kathmandu University Journal of Science, Engineering and Technology*, *6*(2), 60-65.
31. Chary, N. S., Kamala, C. T., & Raj, D. S. S. (2008). Assessing risk of heavy metals from consuming food grown on sewage irrigated soils and food chain transfer. *Ecotoxicology and environmental safety*, *69*(3), 513-524.
32. Chen, H., Teng, Y., Lu, S., Wang, Y., & Wang, J. (2015). Contamination features and health risk of soil heavy metals in China. *Science of the total environment*, *512*, 143-153.
33. Tomas J., Arvay J. and Toth T., Heavy metals in productive parts of agricultural plants, Journal of Microbiology, Biotechnology and Food Science, 1, 819 – 817 (2012)
34. Food Codex, SR no. 981/1996-100, part 2, chapter 3, appendix 2 (1996)
35. Nwajei G. E, Okwagi P., Nwajei R.I. and Obi-Iyeke G.E, Analytical Assessment of Trace Elements in Soils, Tomato Leaves and Fruits in the Vicinity of Paint Industry, Nigeria, Research Journal of Recent Sciences, 1(4), 22-26, (2012)
36. Rangnekar S.S., Sahu S.K., Pandit G.G. and Gaikwad V.B., Study of uptake of Pb and Cd by three nutritionally important Indian vegetables grown in artificially contaminated soils of Mumbai, India, International Research Journal of Environment Science, 2(9), 1-5, (2013)
37. Blaylock M., Salt D., Dushenkov S., Zakharova O.,Gussman C., Kapulnik Y., Ensley B. and Raskin I., Enhanced accumulation of Pb in Indian Mustard by soil applied chelating agents, Environ. Sci. Technol., 31, 860- 865 (1997)
38. Marchiol L., Assolari S., Sacco P. and Zerbi G., Phytoextraction of heavy metals by canola and radish grown on multi-contaminated soil, Environ. Pollution. 132, 21-27 (2004)
39. Mellem J.J., Baijnath H. and Odhav B., Bioaccumulation of Cr, Hg, As, Pb, Cu and Ni with the ability for hyperaccumulation by Amaranthusdubius, African Journal of Agricultural Research, 7(4), 591-596 (2012)