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Assessment of quality of leafy vegetables grown in periphery of Allahabad district

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Abstract

The study entitled "assessment of quality of leafy vegetables grown in periphery of Allahabad district" was conducted in four study sites of Allahabad district namely Phaphamau, Mundera, Naini and Jhunsi area, where sewage water was used for irrigating the leafy vegetables. A control site was also selected where fresh ground water used for irrigating the leafy vegetables. The collected samples were included Spinach (*Spinacia oleracea* L.), Soya leaves/Dill (*Anethum graveolens* L.) and Fenugreek leaves (*Trigonella foenum-graecum* L.). The main objectives of this study were to assess the knowledge, attitude and practices regarding the consumption pattern of leafy vegetables among vegetable growers, to analyze the heavy metals content like Cd, Cr, Cu, Pb, Ni and Zn in sewage water irrigated leafy vegetables, compare the obtained values with fresh ground water irrigated leafy vegetables and safe limits given by various national and international agencies and to calculate the daily metal intake and their health risk index. No vegetable growers had the knowledge regarding heavy metals contamination and their health hazard.

The study showed that the leafy vegetables irrigated with waste water heavy metals content had slightly higher in comparison to control site where fresh ground water was used. Heavy metal concentrations in leafy vegetables were below the permissible levels, except Pb in Fenugreek leaves for Naini area. However the health risk indexes (HRI) for all heavy metals were above 1 that indicates the health concern.

Thus the study showed that leafy vegetables produced in sewage irrigated area of Allahabad district represent a high health risk index for heavy metals intake through vegetables for consumers in comparison to fresh water grown leafy vegetables. Heavy metal contaminated vegetables grown in sewage water-irrigated areas may pose public health hazards which is not safe and may not be sustainable in the long term.

Keywords: leafy vegetables, heavy metals, sewage water irrigation, daily metal intake, health risk index

Introduction

The fast urbanization cities have resulted in an increase in food demand for market gardening productions. The vegetables production offers a significant opportunity by providing employment for the poor people. Vegetables are fully recognized for their benefits towards healthy living, by their protective properties against cancer and other chronic degenerative diseases such as cardiovascular diseases and diabetes.

The daily vegetables intake of 400 g to 600 g is recommended by the World Health Organization, Food and Agriculture Organization of the United Nations, and the World Cancer Research Fund. In the developing countries, the diarrheic diseases of food or hydrous origin kill 2.2 millions people annually.

Using polluted or sewage water in the periphery of big cities for irrigating the leafy vegetables are a common practice, this is occurring due to constraints in availability of fresh water at very high cost for irrigation of agricultural fields while in another hand farmers are mainly interested in more benefits like increased productions at very low costs includes cheap and easily available water sources. Generally farmers are not aware about harmful effects of sewage or waste water which contain large amount of heavy metals and having potential diseases causing microorganisms and transfer these contaminants to leafy vegetables through irrigation. Sewage effluent constitutes mainly industrial and domestic wastes which are very harmful and hazardous to human health if treatment is not done.

Prolong use of this water for irrigation purpose has resulted in accumulation of various heavy metals. The consumption of these contaminated vegetables is one of the most important pathways of toxic metals and its induced diseases. In recent years, attention focused on this type of contamination has increased throughout the world. The excessive accumulation of toxic heavy metals may also cause serious health problems because of their non-biodegradable nature and their potentiality to accumulate in different body organs and its nature to interfere with the enzyme action by replacing metal ions from the metal enzyme and are capable of causing a reduction in the activity of hydrolysis *viz*; α amylase, phosphates, RNAs proteins and interferences with the membrane permeability. Exposures to heavy metals cause reduction in respiration of leaves, stomatal conductance, reduction in photosystem IInd activity, structural changes in chloroplast and rate of photosynthesis, reduction in total lipids, glycolipids and neural lipids.

Objectives

1. To analyze the heavy metals content of leafy vegetables and compare the obtained values of heavy metals with the control samples and safe limits given by various agencies.
2. To determine the microbial load in green leafy vegetables.
3. To assess the estimated daily intake of heavy metals and their health risk index.

Materials and Methods

The present study was conducted using the following methodology related with the nature of problem and objectives.

Selection of study sites

Periphery areas of Allahabad district namely Naini (Madauka, Fulwaria and near BPCL area) Jhunsi (around Saraitaki area, Mori Daraganj) Phaphamau (around Shantipuram Colony) and Mundera (near Ponghatpul) were selected for the present study as they are peri - urban areas, use of water for irrigating the vegetable crops from tanks are part of the city drainage system that drain untreated and partially treated domestic sewage and industrial effluents from a number of small scale units.

Selection of control site

Control site was selected from Madauka village of Naini area where one vegetable grower was using the tube well water for irrigating the leafy vegetables but all the environmental condition was same as other selected study sites.

Collection of samples

Fresh leafy vegetable samples were collected in triplicate from four different sites namely Naini, Jhunsi, Phaphamau, and Mundera. There were many vegetables grown in selected areas but these leafy vegetables includes *Spinacia oleracea* L. (Spinach), *Anethum graveolens* L. (Dill leaves/Soa) and *Trigonella foenum-graecum* L. (Fenugreek leaves) were commonly irrigated with sewage water.

Estimation of heavy metals in selected leafy vegetables

Lead, Cadmium, Chromium, Copper, Zinc and Nickel were estimated by using Atomic Absorption Spectrophotometer (AAS Perkin Almer 400), in the department of Soil Science, College of Agriculture, SHUATS, Allahabad, U.P. (Perkin Elmer, Inc, 2010) [25].

Sample preparation and treatment

Vegetable samples were brought to the laboratory and washed under clean tap water followed by double distilled water to eliminate soil and air-borne pollutants. The moisture and water droplets were removed with the help of blotting papers.100 g of edible portion of all three samples was homogenized, and immediately filtered with Whatman filter paper number 42 with two to three times.

Digestion processes: 1.Took washed and grinded green leafy vegetable with the help of cleaned pastel and mortars 2. Sample filtered with the help of filter paper. 3. Took 10 ml of filtrated sample in a 50 ml beaker 4.Heated it on 100ºC until 1 ml is left. 5. Added 3 ml HNO_3. 6. Heated it again on 100ºC until the volume 1 ml was left and cool it. 7. Added 1 ml perchloric acid. 8. Heated it again until the volume 1ml was left. 9. Added 25 ml distill water with help of filter paper. 10. Finally made the volume 50 ml in a conical flask

AAS Calibration

Switched on the air and leave it for 10 minutes to maintain the pressure required. 2. Switched on the AAS unit and open software.3. Inserted the lamp in the given blocks 1, 2, 3 or 4. 4. Clicked on the lamp option appeared in window. 5. Clicked on setup (energy and wavelength will be appeared). 6. Clicked on the file tab to open the workplace and select SHUATS Went continuous graphics and set the maximum value 1.00 and min. value 0.00. 7. Turned on the flame Set the burner height according to the lamp up to the min. value appeared in the graph. 8. Adopted the recommended method for running the standard 1 and 2, system is ok for analysis.

3.5.4 AAS analysis method

File → New → Method → Selected the element → Gave name to selected methods (element) → went on calibration and set equation liner through zero → Defined blank and standard 1 and 2 → Set the value of standard in PPM according to selected element by checking the recommended conditions → Then went on file menu and selected save as → In save as selected the method option & given name to above method the save the file and continued the analysis.

Results and Discussions

Analysis of heavy metals in leafy vegetables

Environmental pollution is a major problem and its direct or indirect effect is hazardous to human health. Vegetables are a major part of Indian diet and it is very susceptible to heavy metals contamination.

Cadmium, Chromium, Copper, Lead, Nickel and Zinc were estimated by using Atomic Absorption Spectrophotometer (AAS Perkin Almer 400) in selected leafy vegetables.

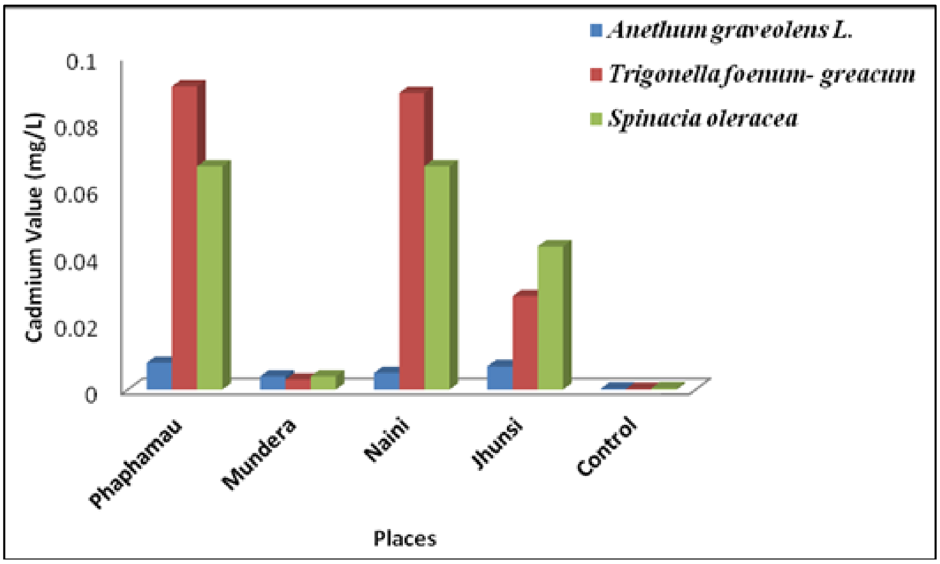

Fig 1: Cadmium content in leafy vegetables (mg/kg)

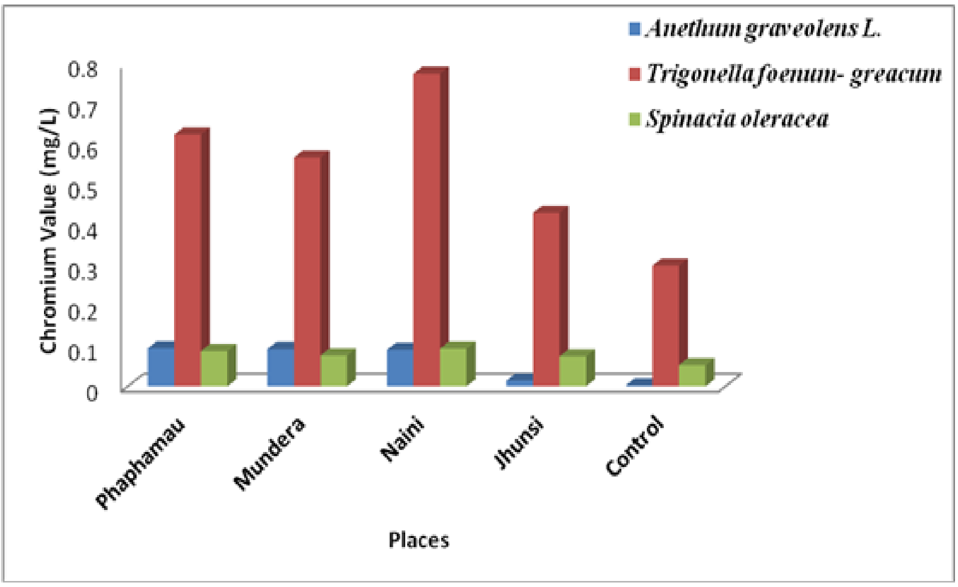

Fig 2: Chromium contents in leafy vegetables (mg/kg)

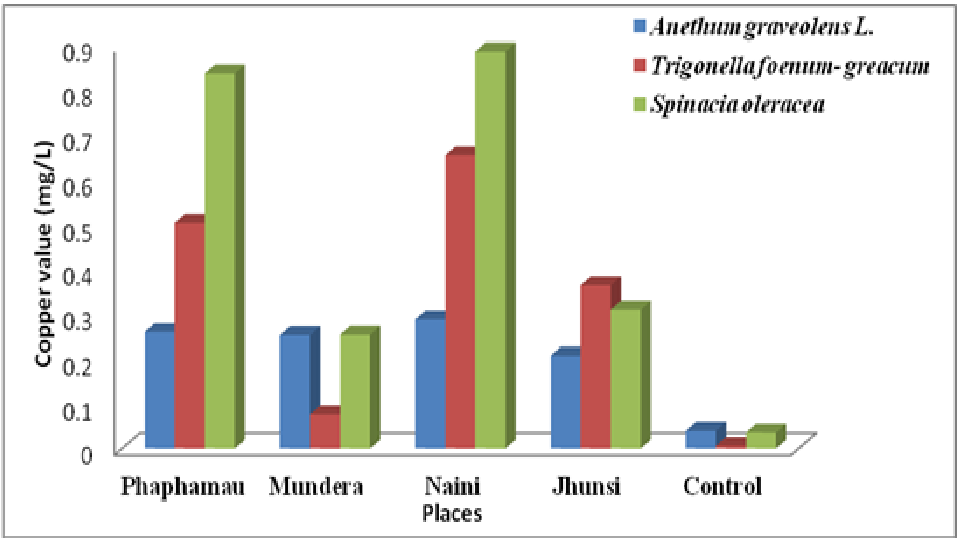

Fig 3: Copper content in leafy vegetables (mg/kg)

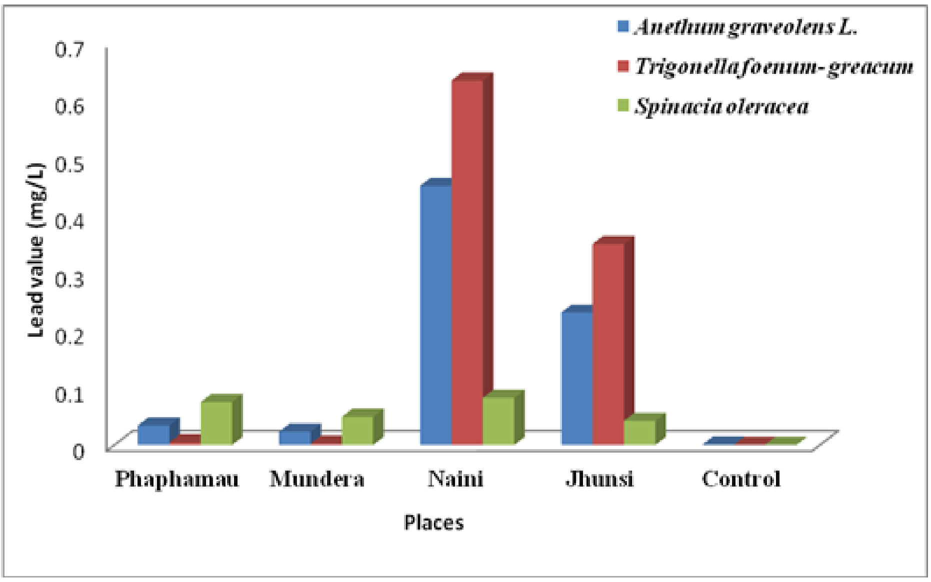

Fig 4: Lead contents in leafy vegetables (mg/kg)

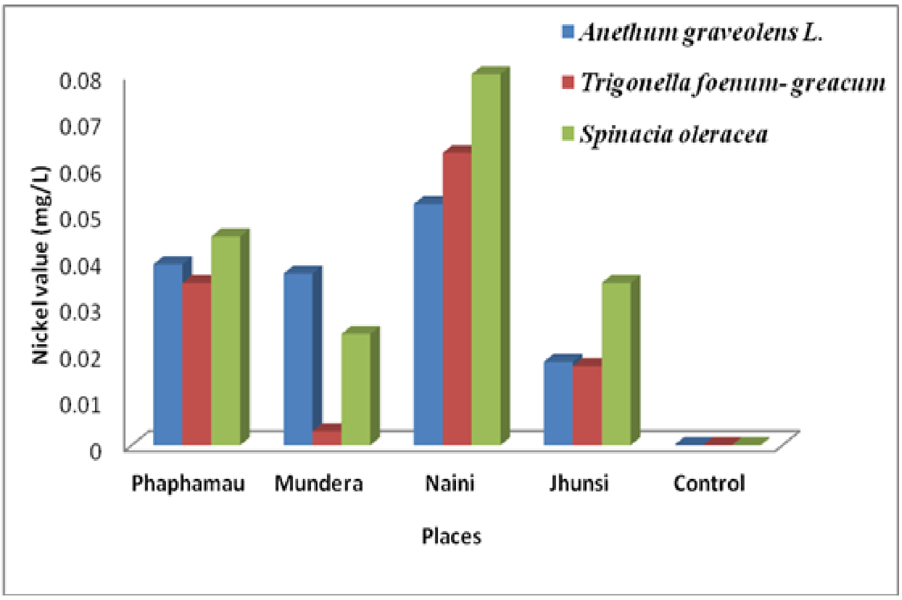

Fig 5: Nickel content in leafy vegetables (mg/kg)

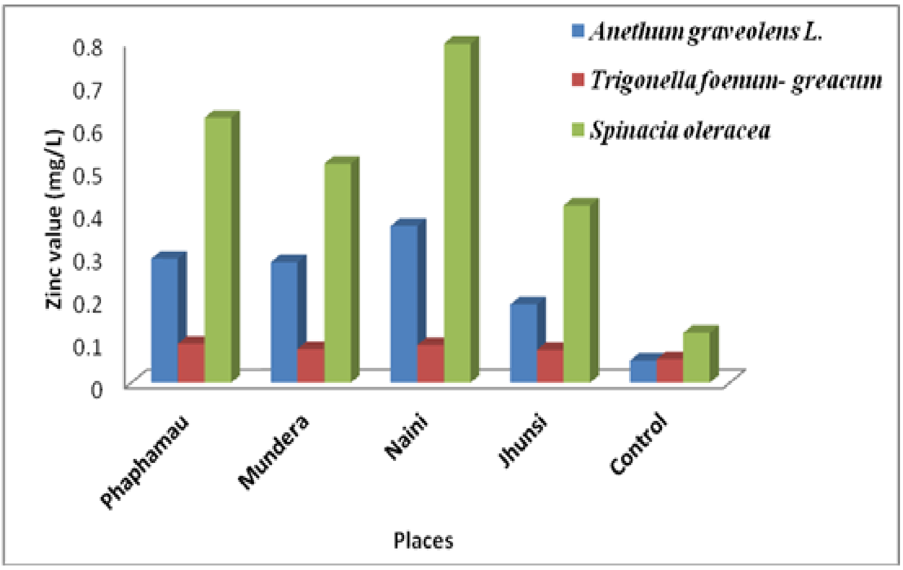

Fig 6: Zinc content in leafy vegetables (mg/kg)

The above figure 1, showed that the highest Cd content was found in *Trigonella foenum-graecum* L. for Naini area while lowest was also in *Trigonella foenum-graecum* L. leaves for Mundera area. The averages Cd content trends in all the leafy vegetables for all places was *Trigonella foenum-graecum* L. (0.0527) > *Spinacia oleracea* L. (0.0452) > *Anethum graveolens* L. (0.006). Cd content was not detected in control samples.

ANOVA concluded significant differences between vegetables while no significant difference was observed between places.

Figure 2, revealed that the highest Cr content was found in *Trigonella foenum-graecum* L. for Naini area, while lowest was found in *Anethum graveolens* L. for Jhunsi area. The average Cr content trends in all the leafy vegetables for all places were trend as follows: *Trigonella foenum-graecum* L. (0.499) > *Spinacia oleracea* L. (0.085) > *Anethum graveolens* L. (0.0266). In control samples the Cr content were lower than waste water irrigated leafy vegetables, for *Anethum graveolens* L., *Trigonella foenum-graecum* L. and *Spinacia oleracea* L. Cr content were 0.0266±0.001, 0.442±0.0044 and 0.064±0.0011, respectively.

ANOVA concluded significant difference between vegetables and places.

Figure 3, reported that the highest Cu content was found in *Spinacia oleracea* L. for Naini area, while lowest was also in *Anethum graveolens* L. for Jhunsi area. The average Cu content trends in all the leafy vegetables for all places were as follows: *Spinacia oleracea* L. (0.5727) > *Trigonella foenum-graecum* L. (0.400) > *Anethum graveolens* L. (0.252). In control samples the content was lower than waste water irrigated grown leafy vegetables for *Anethum graveolens* L., *Trigonella foenum-graecum* L. and *Spinacia oleracea* L. was 0.004±0.009, 0.006±.0063 and 0.036±0.0009.

As regards to copper ANOVA showed significant difference for vegetables and places also. The above figure 4, it was showed highest content of lead was found in *Trigonella foenum-graecum* L. for Naini area while lowest was found in *Anethum graveolens* L. for Mundera area. The averages lead content in leafy vegetables trends in all the leafy vegetables for all places as follows: *Trigonella foenum-graecum* L. (0.247) > *Spinacia oleracea* L. (0.0619) > *Anethum graveolens* L. (0.031). In control samples the content was lower than the sewage water irrigated grown leafy vegetables, for *Anethum graveolens* L., *Trigonella foenum-graecum* L. and *Spinacia oleracea* L. contents were 0.0006±0.0002, 0.0001±0.0272 and 0.0003±0.0001, respectively.

According to calculated ANOVA significant effect was found between vegetables as well as leafy vegetables.

In this study figure 5, revealed that the Ni content range for *Anethum graveolens* L**.** for all places were from 0.018±0.0026 to 0.052±0.0092, in *Trigonella foenum-graecum* L. from 0.003±0.0038 to 0.063±0.0069 and in *Spinacia oleracea* L. from 0.08±0.0083 to 0.45±0.005. Highest Ni content was found in *Trigonella foenum-graecum* L. for Naini area while lowest was also in *Trigonella foenum-graecum* L. for Mundera area. Ni content trends in all the leafy vegetables for all places were trends as follows: *Trigonella foenum-graecum* L. (0.295) > *Spinacia oleracea* L. (0.0365) > *Anethum graveolens* L. (0.046). In control samples of leafy vegetables the Ni content for *Anethum graveolens* L., *Trigonella foenum-graecum* L., *Spinacia oleracea* L. were 0.014±0.0023, 0.001±0.002 and 0.002±0.0004, respectively.

ANOVA revealed significant effect among places but not for between vegetables.

In the figure 6, the obtained results reported that the highest Zn content was found in *Spinacia oleracea* L. for Naini area while lowest was in *Trigonella foenum-graecum* L. for Jhunsi area. Zn content trends in all the leafy vegetables for all places were as follows: *Spinacia oleracea* L. (0.4877) > *Anethum graveolens* L. (0.223) > *Trigonella foenum-graecum* L. (0.0656). In fresh water irrigated grown leafy vegetables the content was lower than waste water irrigated leafy vegetables for *Anethum graveolens* L., *Trigonella foenum-graecum* L. and *Spinacia oleracea* L. was 0.0283±0.004, 0.0792±0.0021 and 0.0514±0.0006, respectively.

ANOVA concluded significant difference between vegetables but not among the selected places.

Estimation of daily intake of heavy metals and their health risk index

The degree of toxicity of heavy metals to human beings depends upon their daily intake and concentration of heavy metals (FAO/WHO, 2007). Numerous reports indicate that vegetables have been heavily polluted by Lead (Pb), Arsenic (As), Copper (Cu), Chromium (Cr), Zinc (Zn) and Cadmium (Cd) near the peri-urban areas. Pb, As, Cu, Cr and Cd are important toxic heavy metals and have been identified as health risks (ECFAC, Joint FAO/WHO Expert Committee on Food Additives, 1993).

Amount of vegetables consumed (estimates) were based on expert knowledge and previous studies of the consumption behavior of the peri-urban population of Allahabad district (Amoah *et al.*, 2007) [4]. Formal interviews were conducted and estimated the average consumptions of *Anethum graveolens* L**.**, *Trigonella foenum-graecum* L. and *Spinacia oleracea* L. in g/person/day. It showed that:

a. ***Anethum graveolens* L.:** Generally people were consuming with mix "*sag"* and "*Paratha"* combined with *Trigonella foenum-graecum* L. and *Spinacia oleracea* L. The average weekly consumption was 100 g/capita/week. Thus the daily average intake was 100 /7= 14.2 g/capita/day.
b. ***Trigonella foenum-graecum* L.:** "*Paratha"* mix "*sag"* adding with other vegetables for the taste and flavor instead of "*Methidana"*, were the main form of consumption**.** The average weekly consumption was 300 g/capita/week. Thus the average daily intake was 300/7=42.87 g/capita/day.
c. ***Spinacia oleracea* L.:** Generally people were consuming in the form of "*sag"*, *"paratha"* and "*juice"* form**,** people were taking twice to thrice in a week. The average consumption was 600 g/capita/week. Thus the average daily intake was 600/7= 85.71 g/capita/day.

Table 1: Daily intake of metals (DIM) and health risk index (HRI) of leafy vegetables (µg kg^{-1} $person^{-1}day^{-1}$

H.M \ Veg.		*Anethum graveolens* L.	*Trigonella foenum-graecum* L.	*Spinacia oleracea* L.	*PTDI (µg/kg)
Cd	DIM	0.0001	0.003	0.0054	60
	HRI	0.1	3.00	5.4	
Cr	DIM	0.001	0.030	0.010	10
	HRI	0.0006	0.02	0.006	
Cu	DIM	0.005	0.024	0.0695	300
	HRI	0.125	0.6	1.73	
Pb	DIM	0.0006	0.015	0.0075	214
	HRI	0.15	3.75	1.87	
Ni	DIM	0.0007	0.0179	0.005	1
	HRI	0.035	0.895	0.25	
Zn	DIM	0.0045	0.0039	0.05921	60
	HRI	0.035	0.13	1.97	

*PTDI- Provisional tolerable daily intake, Sources; WHO/FAO –JECFA (1999), IOM (2004) also reported by Singh *et al.* (2010)

The above table 1 showed that for adults, the average EDI values of Cd, Cr, Cu, Pb, Ni and Zn for *Anethum graveolens* L. were in ascending order trends as Cd < Pb < Ni < Cr < Cu < Zn, for *Trigonella foenum-graecum* L. trends as Cd < Zn < Pb < Ni < Cu < Cr while the EDI for *Spinacia oleracea* L. were trends as Ni < Cd < Pb < Cr < Zn < Cu.

EDI for all the studied heavy metals in comparison with provisional tolerable daily intake (PTDI) was found to be safe for human health. (WHO/FAO –JECFA, 1999) and IOM (2004) also reported by Singh *et al.* (2010). While comparing with the HRI, (US-EPA, 2006) the value of all the selected individual leafy vegetables for each metal was less than 1, except Cd and Pb for *Trigonella foenum-graecum* L. and *Spinacia oleracea* L., Cu and Zn only for *Spinacia oleracea* L. The HRI values below 1 indicating that the intake of a single metal through consumption of vegetables does not pose a considerable potential health hazard. However, total metal HRI value (sum of individual metal HRI) for individual vegetable was higher than 1 and it can be suggested that the consumption of average amounts of these contaminated green leafy vegetables might pose a health risk for the consumers, therefore of some concern. It assumes that the magnitude of the adverse effect will be proportional to the sum of multiple metal exposures. It also assumes similar working mechanisms that linearly affect the target organ.

Radwan and Salama (2006) [27] and Khan *et al.* (2008) had also observed no risk due to consumption of vegetables grown under waste water irrigated areas. Singh (2010); Sharma (2010) and Zheng *et al.* (2007) [35] (except for Cd), Khan *et al.* (2008) and Guerra *et al*. (2012) [19] also found lower values than tolerable daily intake limits. On the other hand, Chary *et al.* (2008) [10] and Chauhan (2014) [11] recorded higher DIM values for heavy metals than tolerable daily intake limits. While the lowest DIM of Cd was estimated to 0.000415 mg/kg/day which represent approximately 41.5% of RfDo value of 0.001g/kg/day for a 0.345Kg for adults. However The DIM of Cd was lower than tolerable daily intake (Biego *et al.*, 1998) [9]. This lower DIM of Cd was lower than that reported in literature, which ranged between 0.008 mg/kg and 0.052 mg/kg/day by Santos *et al.* (2004) [29, 30] and Tripathi *et al.* (1997) [31, 32].

Nevertheless, consumption of foodstuff with elevated levels of heavy metals for long term may lead to high level of accumulation in the body causing related health disorders. It is therefore suggested that regular monitoring of heavy metals in vegetables is essential in order to prevent excessive build-up of these metals in the human food chain. Besides, the metal contaminated areas should be discouraged for commercial farming of such leafy vegetables. Instead, those areas may be replaced by some other non-metal accumulating plants.

Conclusion

It was concluded that The average consumption of *Anethum graveolens* L. (Dill/Soa leaves), *Trigonella foenum-graecum* L. (Fenugreek leaves) and *Spinacia oleracea* L. (Spinach leaves) were 100 g, 300 g, and 600 g/capita/week, respectively Majority of the respondents were just consuming the leafy vegetables as a matter of their habits. The main form of consumption was "*sag*", "*paratha*" and "*soup"*. Practices regarding washing of the vegetables were inappropriate. No respondents had the knowledge regarding consumption effects of heavy metals and cross- contamination.

In terms of heavy metals, obtained data revealed that the highest cadmium, chromium, lead and nickel contents were found in *Trigonella foenum-graecum* L. while zinc and copper contents were found to be highest in *Spinacia oleracea* L. In control samples the cadmium content was not detected whereas remaining all analyzed heavy metals content found to be much lower than waste water irrigated leafy vegetables.

The average daily intake for *Anethum graveolens* L., *Trigonella foenum-graecum* L. and *Spinacia oleracea* L. was 14.2 g, 42.87 g and 85.71 g/capita/day, respectively. EDI values of Cd, Cr, Cu, Pb, Ni and Zn for *Anethum graveolens* L. were in ascending order trends as Cd < Pb < Ni < Cr < Cu < Zn, for *Trigonella foenum-graecum* L. trends as Cd < Zn < Pb < Ni < Cu < Cr while the EDI for *Spinacia oleracea* L. were trends as Ni < Cd < Pb < Cr < Zn < Cu. EDI for all selected heavy metals in comparison with provisional tolerable daily intake (PTDI) was found to be safe for human health. While comparing with the HRI, (US-EPA, 2006) the value of all the selected individual leafy vegetables for each metal was less than 1 (does not pose a considerable potential health hazard) except Cd and Pb for *Trigonella foenum-graecum* L. and *Spinacia oleracea* L., Cu and Zn only for *Spinacia oleracea* L.

Recommendations

Further study is needed in the respect such as metal uptake studies at cellular level including efflux and influx of different metal ions by organelles and membranes. Most of the heavy metals discussed have toxic potential but the detrimental impact become apparent only after decades of exposures. It is therefore suggested that regular monitoring of heavy metals in plant tissue is essential in order to prevent excessive buildup of this metals in the human chain.

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