International Journal of Plant and Environment
Vol. 3, No. 1 (2017), 65-76

Effect of Wastewater Irrigation on Heavy Metal Accumulation, Growth and Yield of Vegetables

Saima Kausar[1]*, Shahla Faizan[2] and Irfana Haneef[2]

[1]Jhunjhunwala Degree College, Faizabad-224133, INDIA;
[2]Environmental Physiology Laboratory, Department of Botany, Aligarh Muslim University, Aligarh-202002, INDIA

Publication Info

Article history:
Received : 01.10.2016
Accepted : 15.06.2017
DOI : 10.18811/ijpen.v3i.8448

Key words:
Carrot
Fenugreek
Growth
Heavy metal
Radish
Spinach
Wastewater
Yield

Corresponding author:
Dr. Saima Kausar
Tel: +91-9415962818
Email: kausar.saima25@gmail.com

Abstract

Use of wastewater for irrigation is on the rise in India and other developing countries. Wastewater contains plant nutrients that favour crop growth but leave a burden of heavy metals which can enter the food chain and is a cause of great concern. This study was conducted to explore the possibility of using wastewater to grow four vegetables fenugreek (*Trigonella foenum-graecum* L.), spinach (*Spinacia oleracea* L.), radish (*Raphanus sativus* L.) and carrot (*Daucus carota*L.). Two aspects namely (1) effect on plant growth and yield (2) accumulation of cadmium (Cd), chromium (Cr), copper (Cu), nickel (Ni), lead (Pb) and zinc (Zn) in leaves and roots of the plant have been presented in this paper. The physico-chemical analysis of the wastewater showed that it was rich in total suspended and dissolved solids with large amount of BOD and COD. The higher amount of Cl^-, Ca^{++}, Mg^{++} and K^+ were also present in the effluent. The heavy metal (Cd, Cr, Cu, Ni, Pb and Zn) content in wastewater is comparatively more than groundwater (GW). The values of these heavy metals were slightly higher in the soil irrigated with wastewater. The effluent severely affects crop plants and soil properties when used for irrigation. The growth characteristics (plant length, plant fresh and dry weight, leaf number and leaf area) as well as yield characteristics (seed number, 1000 seed weight and seed yield) of all the plants, irrigated with 100% wastewater, were more than that with groundwater. The pattern of increase for the plants was fenugreek> radish> spinach> carrot. Though the wastewater contains low levels of the heavy metals, the soil and plant samples show higher values due to accumulation, but their level was under permissible limits in plants. The trend of metal accumulation in wastewater-irrigated soil is in the order: Pb> Ni> Zn> Cu> Cr> Cd. Of the four plants that are grown, the order of total heavy metal accumulation in roots is Carrot> Radish, while in leaves the order is Spinach> Fenugreek. The enrichment factor (EF) of the heavy metals in contaminated soil is in the sequence of Ni (3.1)>Pb (2.6)> Cd (2.35)> Zn (2.18)> Cu (1.66)> Cr (1.05), while in plants EF varies depending upon the species and plant part. Radish and carrot show a high transfer factor (TF >1) for Cd signifying a high mobility of Cd from soil to plant whereas the TF values for Pb are very low as it is not bioavailable. Thus, it may be concluded that wastewater may be used profitably for the cultivation of these vegetables and could effectively supplement not only the nutrient requirement of the crop but may also act as the source of water.

1. Introduction

Environmental pollution affects the quality of pedosphere, hydrosphere, atmosphere, lithosphere and biosphere (Lone *et al*., 2008). Land and water are precious natural resources on which rely the sustainability of agriculture and the civilization of mankind. Unfortunately, they have been subjected to maximum exploitation and severely polluted due to anthropogenic activities. Water pollution is a problem of worldwide concern and groundwater is extremely polluted due to unplanned disposal of untreated domestic sewage and industrial effluents into watercourses (Mashiatullah *et al*., 2005). Disposal of sewage water and industrial wastes is a great problem. Often it is drained to the agricultural lands where it is used for growing crops including vegetables.

Heavy metals are ubiquitous in the environment, as a result of both natural and anthropogenic activities, and humans are exposed to them through various pathways (Wilson and Pyatt, 2007). Wastewater irrigation is known to contribute significantly to the heavy metal contents of soil (Mapanda *et al*., 2005). Heavy metals in wastewater come from industries and municipal sewage, and they are one of the main causes of water and soil pollution. Long-term use of wastewater on agricultural lands contributes significantly to the build-up of the elevated levels of these metals in soils and plants (Mapanda *et al*., 2005; Sharma *et al*., 2007) which is of serious concern. Although problems occur in waterways when pollutants are leached out of the soil. If the plants die and decay, heavy metals taken into the plants are redistributed, so the soil is enriched with the pollutants. Uptake and accumulation of elements by plants may follow two different paths i.e. through the roots and foliar surface (Sawidis *et al*., 2001). The uptake of metals from the soil depends on different factors such as their soluble content in it, soil pH, plant growth stages, types of species, fertilizers and soil (Ismail *et al*., 2005; Sharma *et al*., 2006).

Plants have a natural propensity to take up metals. Some of them like Cu^{++}, Co^{++}, Fe^{++}, Mo^{++}, Mn^{++} and Zn^{++} are essential plant micronutrients (Baker *et al*., 1991), while few others like Hg^{++}, Cd^{++}, Ni^{++} and Pb^{++} are toxic to plants. However such toxic effects are even varying from genotype to genotype of the same crop (Liu *et al*., 2001). Leafy vegetables are high accumulator of metal ions as compared to root vegetables and legumes (Alexander *et al*., 2006).

This study was therefore; conducted to investigate the soil pollution load, to understand the appropriateness of wastewater-irrigated soils for vegetable cultivation, to study the effect of wastewater irrigation on growth and yield of fenugreek, spinach, radish and carrot, and to determine the concentration of accumulated trace elements by these vegetables irrigated with different concentrations of wastewater.

2. Materials and Methods

2.1. Experimentation

An experiment of randomized block design was conducted in 2009-10 in the naturally illuminated net house of the Department of Botany, Aligarh Muslim University, and each treatment was set simultaneously in triplicate. The statistical design used included three treatments i.e. (a) control (GW-fresh irrigation water or ground water), (b) 50% wastewater (50%WW- the dilution was obtained by mixing the GW and WW in 1:1 ratio), and (c) 100% wastewater. The urban wastewater used in this study includes wastewater from the households and sewage together with wastewater from local industries of locks and electroplating as well, and was collected from the outskirts of Aligarh city nearly 5 km from the town where it is being used by local formers to irrigate crops.

The surface of the seeds was sterilized with 0.5% (v/v) sodium hypochlorite solution followed by repeated washing with double distilled water (DDW) (Sauer and Burroughs, 1986). The surface-sterilized seeds were sown in earthen pots (8" diameter), filled with sandy loam soil mixed with farm-yard manure (Tadesse *et al*., 2013).

2.2. Analytical methods

Before irrigation the water samples were collected and analysed for physico-chemical characteristics adopting the procedures outlined in the standard methods (APHA, 1998). Microbiological analyses were also carried out according to and mean values were obtained from three random samples. The wastewater samples were collected directly from the drain and were also used to irrigate the plants. Collected samples were then transported directly to the laboratory and kept in the refrigerator. The samples were examined within 24 h of collection for the presence of members of the coliform bacteria group by most probable number (MPN) method and total bacterial count by spread plate technique adopting the procedures outlined in the standard methods (APHA, 1998).

The soil samples were collected before the start of the experiment. These samples were also analysed for standard physico-chemical properties according to Jackson (1973) and Ghosh *et al*. (1988).

Based on type of industries in the local area and on earlier reports, six heavy metals were selected for their estimation in wastewater and soil-Cadmium (Cd), Copper (Cu), Chromium (Cr), Lead (Pb), Nickel (Ni) and Zinc (Zn)-using the atomic absorption spectrophotometer (SENSAA GBC Avanta Var. 2.02). Heavy metal was estimated in wastewater by using the method of Ademoroti (1996) and in soil by the method of Lindsey and Norwell (1978).

The plants were sampled at 30, 60 and 90 days after sowing (DAS) to make various observations. The plants were uprooted gently and washed under running tap water to remove adhering soil. The plant length, plant fresh weight, leaf number and leaf area were assessed. The samples were then dried in an oven at 80°C for 48 h. The dehydrated samples were then weighed to record dry mass. Leaf area was measured using a portable leaf area meter (LA-21, Systronics, India). At harvest, yield attributes including seed number, 1000 seed weight and seed yield per plant were noted. Seed number was taken on per plant basis in spinach and carrot whereas on per pod basis in fenugreek and radish.

The plant samples were analysed for metal accumulation at 90 days after sowing (DAS). Each treated plant sample was washed separately with 1% HCl. They were then followed by 3-4 washings with de-ionized water to remove the foreign materials. These samples were then spread on blotting paper and air-dried, then oven dried at 70°C. After drying, leaf samples were crushed to powdered form in a grinder, and then converted into liquid following wet digestion procedure (Campbell and Plank, 1992). Then the accumulation of heavy metals was analysed in the diluted digested sample with the help of atomic absorption spectrophotometer (SENSAA GBC Avanta Var. 2.02).

After the results obtained from AAS, enrichment factor, transfer factor and pollution load index were calculated from the following procedure:

2.2.1. Enrichment factor

Enrichment factor (Barman *et al*., 2000) has been calculated to determine the degree of soil pollution and heavy metal accumulation in plants growing in soil contaminated (wastewater-irrigated) with the effluent with respect to soil and plants growing in the uncontaminated (control) pots.

$$EF = \frac{\text{Concentration of metals in soil or plant in contaminated pots}}{\text{Concentration of metals in soil or plant in uncontaminated pots}}$$

2.2.2. Transfer factor

The translocation of heavy metal from soil to plant parts [transfer factor (TF); Chamberlin, 1983; Harrison and Chirgawi, 1989] was calculated to determine the relative uptake of heavy metals by the plants with respect to soil.

$$TF = \frac{\text{Concentration of metals in plant body in contaminated pots}}{\text{Concentration of metals in soil in that pots}}$$

2.2.3. Pollution Load Index

The degree of soil pollution for each metal was measured using the pollution load index (PLI) technique depending on soil metal concentrations. The following modified equation was used to assess the PLI level in soils (Liu *et al*., 2005).

$$PLI = \frac{\text{Concentraion of heavy metal in wastewater irrigated or contaminated soil}}{\text{Concentration of heavy metal in uncontaminated soil (control)}}$$

2.3. Statistical Analysis

Results obtained were statistically analysed, following the procedure described by Steel and Torrie (1996).

3. Results and Discussion

3.1. Physico-chemical quality of irrigation waters

Table 1 shows the average physico-chemical characteristics of the two irrigation waters. Overall the load of most of the analyzed parameters was much higher for WW than for the GW. The physical characteristics (pH, EC, TDS, TS and TSS) of wastewater were in agreement with the recommendations of Food and Agriculture Organization, FAO (Ayers and Westcot, 1994). In terms of chemical quality, high values (with high standard deviation) were observed for WW (Pescod, 1992; Ayers and Westcot, 1994). BOD of WW was in average higher than the 25 mg l^{-1} FAO limit (Pescod, 1992). Clogging (due to high organic content) and pH related risks could be considered low for both water sources.

Negative effect of infiltration and hydraulic conductivity could be considered low as well. Lubello *et al*. (2004) reported that in many investigations, a negative effect of high concentrations of ammonia on crop root growth was observed. In the bulk of soil, ammonia goes through nitrification process. Nitrates indeed migrate into the deep soil layers and can be hazardous for shallow groundwater.

3.2. Nutrients supply of irrigation waters

Wastewater nutrient supply was much higher compared to GW which may cover most of the nutrient requirements of the crop. For example, nitrate nitrogen was 116.67% higher in wastewater as compared to groundwater, however, supply of potassium through GW was not negligible (only 51.6% less than WW). Average NO_3-N was above the limit of 10 mg l^{-1} and average ammonium was in the recommended range (Feigin *et al*., 1991). In conclusion, nutrients supplied by fresh water were consistently much lower than that of wastewater making the later more suitable for irrigation (Table 1).

3.3. Biological quality of irrigation waters

The average load in faecal coliforms of GW was confirm to the WHO recommendation (Blumenthal *et al*., 2000; WHO, 2006) on irrigation water quality for crops susceptible to be eaten uncooked (≤ 10^3 faecal coliforms in 100 ml i.e. 3 decimal logarithmic units per 100 ml or log (CFU/100 ml), but the average load of WW was consistently higher than this standard (Fig. 1). Groundwater was found to be free from helminth eggs but treated wastewater showed helminth eggs concentration of 9.5±11.4 eggs/l. Such a concentration in helminth eggs is higher than the recommendations of the revised WHO standards (less than 0.1 egg/l) for crops likely to be eaten uncooked.

Table 1: Average physicochemical characteristics of groundwater (GW), wastewater (WW) used to irrigate plants. All determinations in mg l^{-1} or as specified. [[a]Ayers and Westcot (1994), [b]Pescod (1992) Number of water samples tested=5]

Determinants	Groundwater	Wastewater	Normal Range
Physical characteristics			
Colour		Light Black	
Odour		Slightly Unpleasant	
pH	7.5	8.2	6.5-8.4[a]
Electrical Conductivity (EC) (ds/m^{-1})	0.74	1.32	0.25-3.0[a]
Chemical characteristics			
Total Dissolved solids (TDS)	542	1421	<2000[a]
Total Solids (TS) (g l^{-1})	947	1288	–
Total Suspended Solids (TSS) (g l^{-1})	431	694	–
Biological Oxygen Demand (BOD)	16.75	52.8	<25[b]
Chemical Oxygen Demand (COD)	62.34	145.23	30-160[b]
Calcium (Ca^{++})	19.24	42.24	<400[a]
Magnesium (Mg^{++})	26	132	<61[a]
Carbonate (CO_3^-)	52.13	132.59	–
Bicarbonate (HCO_3^-)	84.27	94.22	<610[a]
Chloride (Cl^-)	74.66	130.7	<350[a]
Potassium (K^+)	5	11	<2.0[a]
Nitrate Nitrogen (NO_3-N)	0.60	1.30	<10.0[a]
Ammonia Nitrogen (NH_4-N)	0.17	4.12	5.0[a]
Phosphorus (PO_4^-)	0.07	0.89	<2.0[a]
Copper (Cu)	0.135	0.207	0.20[a]
Cadmium (Cd)	0.005	0.008	0.01[a]
Nickel (Ni)	0.092	0.203	0.20[a]
Zinc (Zn)	0.521	0.678	2.0[a]
Chromium (Cr)	0.11	0.15	0.1[a]
Lead (Pb)	2.112	3.264	5[a]

Amahmid *et al*. (1999) and Bouhoum and Amahmid (2002) indicated a strong contamination of the vegetables in particularly the cysts of Giardia and eggs of helminths (Ascaris).

3.4. Physico-chemical quality of soil

The data for soil analysis (Table 2) revealed that the soil was sandy loam having favourable pH (8.2) for root growth and nutrient uptake. Soils, watered with groundwater as well as wastewater presented a contamination in faecal coliforms and helminth eggs. Although groundwater was helminth egg free, the latter were found in related irrigated soils.

3.5. Heavy metal content in irrigation water and soil

For heavy metal content in each irrigant, a total of 12 samples were analysed to obtain their mean values (Table 1). It is evident from the table that concentrations of heavy metals were more in wastewater than groundwater. Among the heavy metals, lead concentration was highest followed by that of zinc, copper, nickel, chromium and cadmium. But level of all the heavy metals was under permissible limit. It is obvious that the higher heavy metal content in wastewater than groundwater will result in their accumulation in the soil, however, their levels are under permissible limit (Table 2). The heavy metal content in soil was found maximum for lead, followed by nickel, zinc, copper, chromium and cadmium. No correlation was observed between the heavy metal content of the wastewater or groundwater with that of the soil and plant. This has clearly demonstrated that the fate of particular metal in soil might depend upon several factors including the interaction of the metal with plant

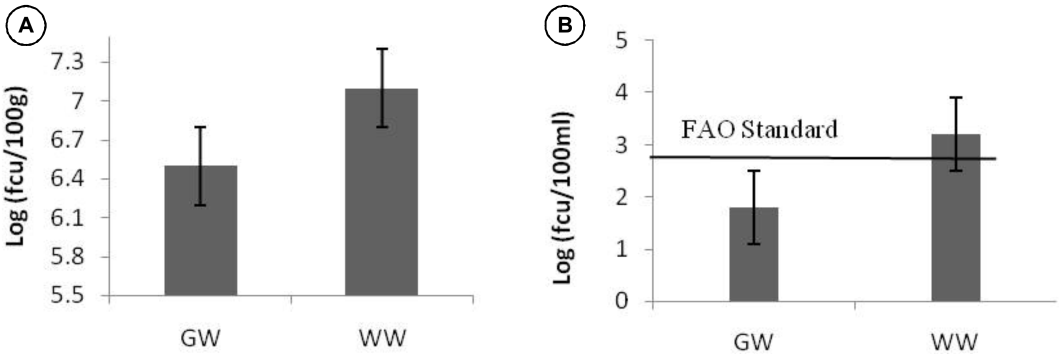

Fig. 1: Average rates of faecal coliforms in (A) irrigated soil cropped with vegetables and (B) groundwater (GW) and wastewater (WW). Error bars denote standard deviation

Table 2: Average physico-chemical characteristics of soil irrigated with groundwater and wastewater. All determinations in mg l^{-1} in 1:5 (soil:water) extract or as specified [Number of soil samples tested=5]

Determinants	GW	WW	Normal Range*
Texture	Sandy loam	Sandy loam	
pH	7.74	8.2	
Cation Exchange Capacity (CEC) (meq 100 gm^{-1} soil)	3.30	3.98	
Electrical Conductivity (EC) (ds/m^{-1})	0.82	0.99	
Total Dissolved solids (TDS)	779.00	835.00	
Calcium (Ca^{++})	29.3	32.06	
Magnesium (Mg^{++})	14.62	23.86	
Carbonate (CO_3^-)	18.08	22.16	
Bicarbonate (HCO_3^-)	185	196	
Chloride (Cl^-)	28.79	39.16	
Potassium (K^+)	9.4	12.50	
Nitrate Nitrogen (NO_3-N) (g kg^{-1}soil)	0.352	0.42	
Phosphorus (PO_4^-) (g kg^{-1}soil)	0.115	0.28	
Cadmium (mg kg^{-1})	2.16	5.07	3-6
Chromium (mg kg^{-1})	27.25	28.55	–
Nickel (mg kg^{-1})	32.50	100.78	75-150
Lead(mg kg^{-1})	50.18	130.64	250-500
Zinc (mg kg^{-1})	45.91	100.06	300-600
Copper (mg kg^{-1})	28.79	47.79	135-270

*Pescod (1992)

(Juste and Mench, 1992) and with organic matter of the soil. However, there was substantial buildup of Cd, Cr, Cu, Ni, Pb and Zn in the wastewater-irrigated soils compared to the groundwater-irrigated soils. On average, the PLI indices for Cd, Cr, Cu, Ni, Pb and Zn were 2.347, 1.05, 1.659, 3.1, 2.6 and 2.18, respectively, using the groundwater irrigated soil concentrations of this study. The enrichment factor of the heavy metals is in the sequence: Ni (3.1)>Pb (2.6)> Cd (2.35)> Zn (2.18)> Cu (1.66)> Cr (1.05). This high enrichment value (>1) indicates higher availability and distribution of metals in soil irrigation with wastewater and thereby increasing the average heavy metal concentration in waste water-irrigated vegetables with respect to their groundwater-irrigated plants. Though the levels of heavy metals in the wastewater are quite low, the irrigated soil shows much higher concentrations of heavy metals.

3.6. Effect on plant characteristics

3.6.1. Growth characteristics

The effect of wastewater on growth characteristics was found significant at the three sampling stages (Fig. 2-3). The increase in wastewater concentration increased the growth characteristics of all the four plants at all sampling stages. The observations recorded at all the sampling stages showed similar pattern of plants to wastewater treatments. The pattern of increase in growth characteristic for the plants was fenugreek> radish> spinach> carrot. Maximum increase in growth characteristics was noted with 100% wastewater followed by 50% wastewater at all the growth stages.

Irrigation with 100% wastewater caused an increase in the plant length of fenugreek by 50, 45 and 48% over their respective controls at three samplings viz. 30, 60 and 90 DAS respectively whereas same irrigation in spinach recorded an increase of 39, 35 and 36.01% over their respective controls at three samplings viz. 30, 60 and 90 DAS respectively. The plant length in root vegetables followed the order radish>carrot due to irrigation with 100% wastewater at all the three sampling stages showing an increase of 44.19, 38.98 and 40% in radish and 35, 33 and 34% in carrot over control. Length of plants increased with the age (Fig. 2 and 5).

100% wastewater increased the plant fresh weight by 45.04, 40 and 41% at 30, 60 and 90 DAS respectively whereas the same irrigation in spinach slightly increased the same parameter by 39.06, 35.18 and 36.57% at 30, 60 and 90 DAS respectively over control. Irrigation with 100% wastewater caused an increase in plant fresh weight by 43, 37.99 and 38.04% in radish and 38.03, 33.76 and 30.09% in carrot at the three sampling stages respectively over groundwater. The fresh weight of the plants increased consistently with ageing (Fig. 2 and 5).

The dry weight in leafy vegetables followed the order fenugreek>spinach due to irrigation with 100% wastewater at all the three sampling stages showing an increase of 42.93, 38.01 and 40% in fenugreek and 39.04, 35.07 and 35.99% in spinach over control. The increase in plant dry weight was 40.26, 36 and 37.68% in radish whereas 37.91, 34.19 and 34.33% in carrot in 100% wastewater irrigation at all the sampling stages, over their respective groundwater. Like fresh weight, dry weight also accumulated upto last stage, therefore maximum recorded at fruiting (Fig. 2 and 5).

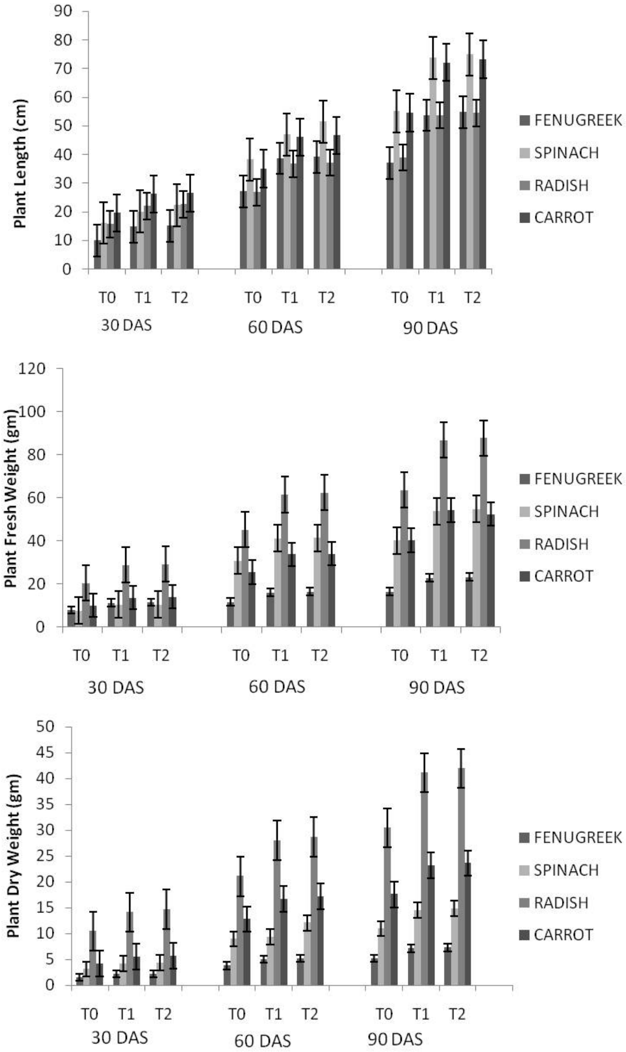

Fig. 2: Effect of GW (T0), 50% WW (T1) and 100% WW (T2) on Plant Length, Fresh Weight and Dry Weight of Vegetables at 30, 60 and 90 DAS [Number of plant samples tested=5]

Leaf number showed a maximum increase of 48.18, 46.87 and 44.63% in fenugreek and 43.94, 40.37 and 40.09% in spinach grown on soil irrigated with 100% wastewater, respectively at the three sampling stages, over their respective groundwater. Leaf number of radish irrigated with 100% wastewater increased by 45.01% at 30 DAS, 43% at 60 DAS and 41% at 90 DAS respectively, over their respective groundwater, whereas in carrot, increase of 40.4% at 30 DAS, 38.51% at 60 DAS and 37.66% at 90 DAS were noted with 100% wastewater, respectively, over their respective groundwater. The leaf number increased from vegetative to flowering and thereafter decreased towards the fruiting stage (Fig. 3 and 6).

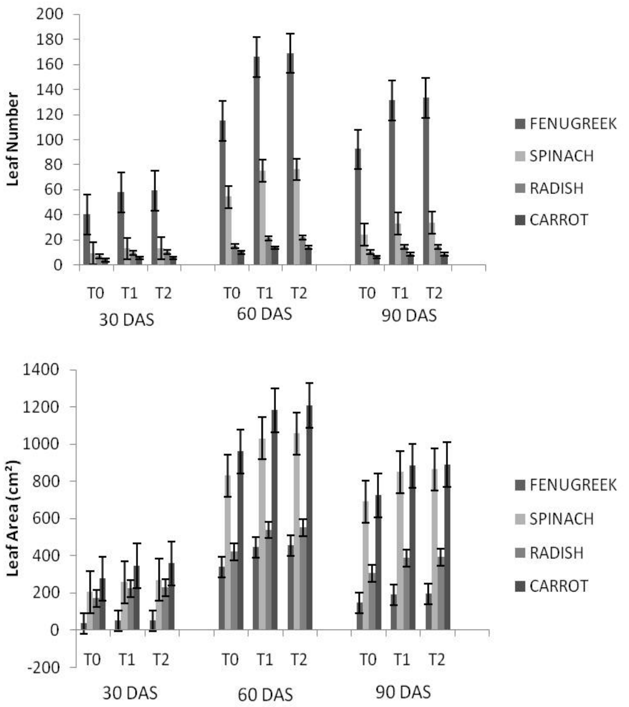

Fig. 3: Effect of GW (T0), 50% WW (T1) and 100% WW (T2) on Leaf number and Leaf area of Vegetables at 30, 60 and 90 DAS [Number of plant samples tested=5]

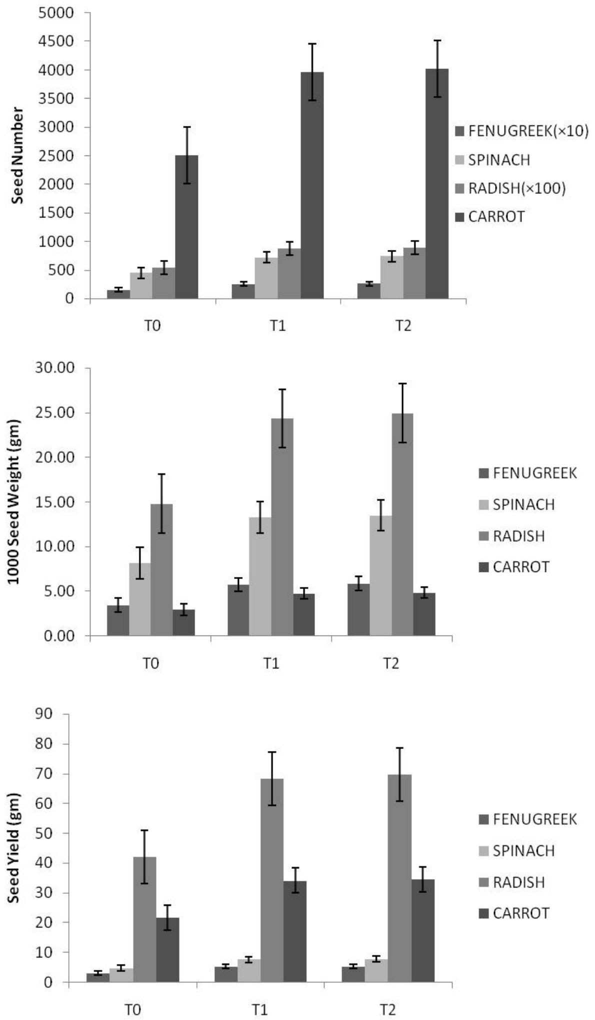

Fig. 4: Effect of GW (T0), 50% WW (T1) and 100% WW (T2) on Seed number and 1000 Seed weight and Seed yield of Vegetables at harvest [Number of plant samples tested=5]

Leaf area of fenugreek irrigated with 100% wastewater increased by 35 % at 30 DAS, 33.78% at 60 DAS and 31.23% at 90 DAS respectively, over their respective groundwater, whereas in spinach, increase of 31.08% at 30 DAS, 27.37% at 60 DAS and 25.39% at 90 DAS were noted with 100% wastewater, respectively, over their respective groundwater. Leaf area showed a maximum increase of 34.82, 31.07 and 28.11% in radish and 29.46, 25.93 and 22.81% in carrot grown on soil irrigated with 100% wastewater, respectively at the three sampling stages, over their respective groundwater. Like leaf number, leaf area also increased from vegetative to flowering and thereafter decreased towards the fruiting stage (Fig. 3 and 6).

The wastewater contained considerable amount of nutrients which are considered essential for maintaining the soil fertility as well as for enhancing the plant growth and productivity. Wastewater in general proved beneficial in increasing the plant growth characteristics and dry matter accumulation was higher in plants receiving it as a source of irrigation water compared to those receiving ground water (GW). 100% WW recorded much more dry matter and growth among the two concentrations of water used. This is primarily due to the presence of certain nutrients of fertilizing value in wastewater (Soumare *et al.*, 2003) that enhances growth of crop plants. Among them nitrogen was present in both ionic forms nitrate and ammonia and as the vegetative growth includes formation of new leaves, stems and roots, the involvement of N through protein metabolism controls them. Another essential nutrient, phosphorus is also important for growth but P applied to the soil is very rapidly changed to less soluble form and therefore regular supply of wastewater could have ensured availability of phosphorus, thus improved the growth. The third important nutrient is potassium which is known to play a significant role in stomata opening and closing (Fischer and Hsiao, 1968) thus contributing to growth.Many research studies and projects have been accomplished on the usage and effects of wastewater as irrigation source on different

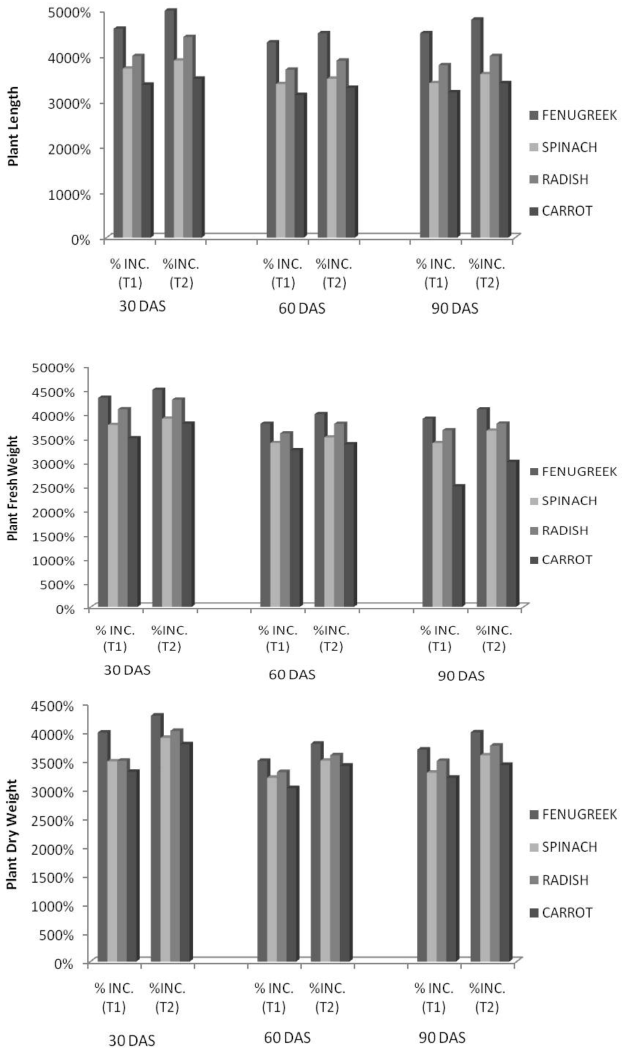

Fig. 5: Percent increase of Plant Length, Fresh Weight and Dry Weight of Vegetables at 30, 60 and 90 DAS due to 50% WW (T1) and 100% WW (T2) over groundwater

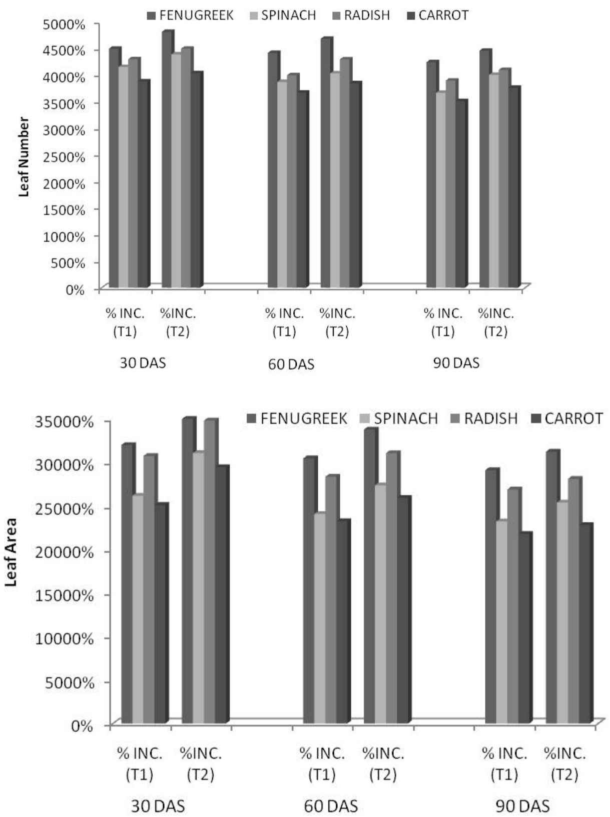

Fig. 6: Percent increase of Leaf Number and Leaf Area of Vegetables at 30, 60 and 90 DAS due to 50%WW (T1) and 100%WW (T2) over groundwater(*In leaf area % increase was multiplied by 10).

crops (Akhtar *et al.*, 2012; Iqbal *et al.*, 2012, 2016; Parveen *et al.*, 2013; Sahay *et al.*, 2013, 2015; Tak *et al.*, 2013; Faizan *et al.*, 2014).

3.6.2. Yield characteristics

The increasing concentration of wastewater significantly increased the yield characteristics of all the four plants (Fig. 4). Maximum increase in yield characteristics was noted with 100% wastewater. The fenugreek and radish exhibited greatest increase in yield characters whereas spinach showed lowest increase followed by carrot. The order of performance for yield characteristics of plants was fenugreek> radish> spinach> carrot.

In fenugreek, the increase in seed number was 68.7%, 1000 seed weight was 69.07% and seed yield was 70.48% due to 100% wastewater respectively, over their respective ground water. On the other hand, spinach showed increase of 63.64, 65.76 and 62.74%in seed number, 1000 seed weight and seed yield respectively (Fig. 4 and 7).

In root vegetables radish showed greater increase of 65.6, 68.31 and 62.74% in seed number, 1000 seed weight and seed yield respectively. In contrast, carrot showed lesser increase in seed number which was 60.43%, 1000 seed weight by 63.92% and seed yield by 59.12% with 100% wastewater respectively (Fig. 4 and 7).

The beneficial effects of wastewater application may be due to the greater capacity of wastewater to supply nutrients to plant and to improve soil properties (Heckman *et al.*, 1986). The obtained results also support the suggestion of Staniforth and Smith (1991) who attributed the increased crop yield as a result of increasing rate of sewage application to the available nutrients supplied particularly nitrogen, which is

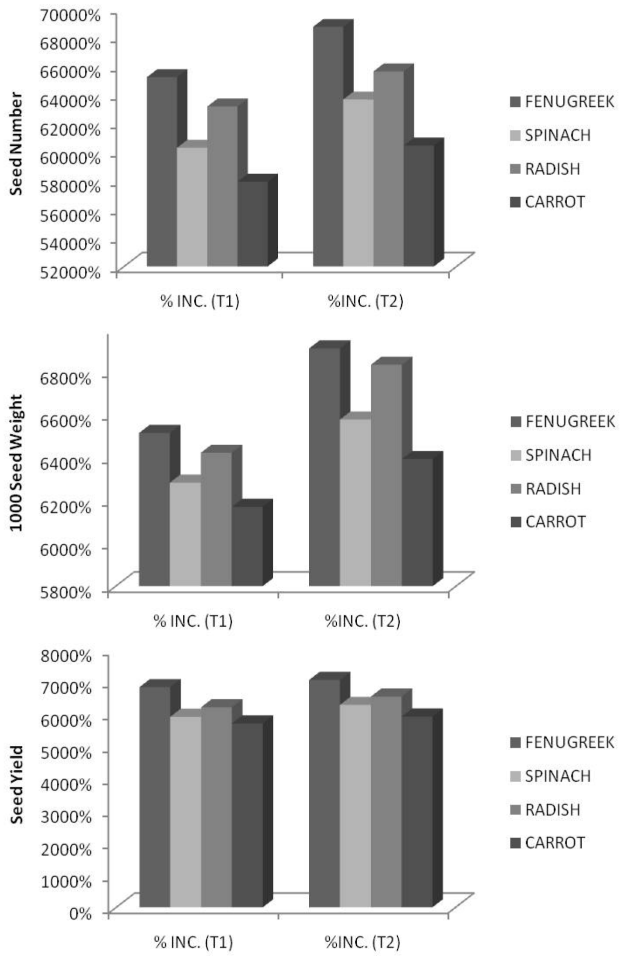

Fig. 7: Percent increase of Seed Number, 1000 Seed Weight and Seed Yield of Vegetables due to 50%WW (T1) and 100%WW (T2) over groundwater(*In seed number % increase was multiplied by 10)

reflected in the increased total nitrogen content of the crop. Tamoutsidis *et al.* (2009) reported that urban treated wastewater effectively increased the yield of cultivated forage crop species, probably due to the nutritive value of the wastewater. Segura *et al.* (2004) advocated the re-use of wastewater in arid and semiarid region of the world and reported significantly higher yield of melon and tomato obtained due to irrigation with effluents.

3.6.3. Heavy metal accumulation in plants

The results of the heavy metal content in plant material of the wastewater-irrigation and groundwater-irrigation pots are presented in Table 3. It is well known that the biomass uptakes trace metals naturally available to them through soil and stores them in their tissues. Absorption and accumulation of heavy metals in plant tissue depend upon many factors, which include temperature, moisture, pH, and nutrient availability (Sharma *et al.*, 2007). The accumulation of heavy metals is greater in case of wastewater-irrigated vegetables than groundwater-irrigated agreeing with many workers (Sinha *et al.*, 2006; Gupta *et al.*, 2007; Sharma *et al.*, 2007). The enrichment of the heavy metals in plants irrigated with contaminated water is as follows:

Fenugreek (leaf) Cr (187) > Zn (65.23) >Pb (30.89) > Cd (24.04) > Ni (4.64) > Cu (3.51)

Spinach (leaf) Zn (61.23) > Cr (34.88) >Pb (30.82) > Cd (6.63) > Ni (5.12) > Cu (3.8)

Radish (root) Zn (102.89) > Cr (65.36) >Pb (33.42) > Ni (10.06) > Cd (8.74) > Cu (3.39)

Carrot (root) Zn (82.62) > Cr (32.63) >Pb (30.06) > Ni (9.24) > Cd (7.64)> Cu (3.037)

It is seen that in most cases Cr has the second highest enrichment in plant bodies followed by Pb with respect to groundwater-irrigated specimens indicating that these metals are mostly accumulated due to irrigation with waste water.

Table 3: Heavy metal content (mg kg^{-1}) in different plants irrigated with groundwater, 50% wastewater and 100% wastewater [Number of plant samples tested=5]

Plant Name	Part Analyzed	Cd			Cu			Ni			Cr			Zn			Pb		
		T_0	T_1	T_2	T_0	T_1	T_2	T_0	T_1	T_2	T_0	T_1	T_2	T_0	T_1	T_2	T_0	T_1	T_2
Fenugreek	Leaf	0.052	0.73	1.25	0.83	1.75	2.91	7.04	24.17	32.69	0.11	14.43	20.57	1.05	53.17	68.49	0.056	0.53	1.73
Spinach	Leaf	0.38	1.04	2.52	1.36	2.83	5.17	8	30.22	40.93	0.72	18.06	25.11	1.24	61.49	75.93	0.098	1.62	3.02
Radish	Root	0.96	3.47	8.39	3.65	8.22	12.36	8.96	58.85	90.14	0.45	20.36	29.41	1.43	138.26	147.13	0.12	2.15	4.01
Carrot	Root	1.37	4.21	10.47	5.36	11.92	16.28	10.11	65.03	93.38	0.94	21.19	30.67	1.94	141.19	160.29	0.18	2.97	4.41
Range of excessive toxicant levels in plants (mg kg^{-1} DW)			5-30[a]			20-100[a]			10-100[a]			5-30[a]			100-400[a]			5[b]	

[a]Kabata-Pendias and Pendias (1984), [b]FAO/WHO standard (1992)

Table 4: Enrichment factor and transfer factor for individual heavy metals caused by the consumption of different selected vegetables grown in wastewater-irrigated soils (T_3)

Plants		Cd	Cu	Ni	Cr	Zn	Pb
Fenugreek	EF	24.04	3.51	4.64	187	65.23	30.89
	TF	0.247	0.061	0.32	0.72	0.684	0.013
Spinach	EF	6.63	3.8	5.12	34.88	61.23	30.82
	TF	0.497	0.108	0.406	0.879	0.759	0.023
Radish	EF	8.74	3.39	10.06	65.36	102.89	33.42
	TF	1.65	0.259	0.894	1.03	1.47	0.031
Carrot	EF	7.64	3.037	9.24	32.63	82.62	30.06
	TF	2.065	0.341	0.927	1.07	1.602	0.041

3.6.4. Heavy metal transfer from soil to plant

The translocation of heavy metal from soil to plant parts (transfer factor) was calculated to determine the relative uptake of heavy metal by the plants with respect to soil (Table 4). The ratio of metals between soil and plant parts (TF) is an important criterion for the contamination assessment of soils with high level of heavy metals. The ratio >1 means higher accumulation of metals in plant parts than soil (Barman *et al*., 2000).

It is seen that in radish and carrot the TF of Cd is greater than 1 indicating its high mobility from soil to plants. Thus, the plants accumulate higher levels of Cd from the soil, though Cd concentration in soil is very low in comparison to the other five heavy metals, its relative accumulation is greater in plants. The high values for Cd transfer factor may be explained by the fact that Cd is easily absorbed by plants (Sheng *et al*., 2001). Cd is one of most moveable metals; it can be moved within organisms by active ion pumps that normally transport Ca^{++} (Berg *et al*., 1995). Following Cd are Zn and Cr. In all cases, the TF values for Cu, Ni and Pb are less than 1 indicating the low mobility of the plants. Pb is one of the least available metals to plants (Berg *et al*., 1995). Hence, though present in much higher concentrations in soil, Pb is of lesser concern as it is less bioavailable than Cd which is more mobile (Gupta *et al*., 2010; Ghosh *et al*., 2012; Xue *et al*., 2012).

4. Conclusion

Acute shortage of drinking water is on the rise and consequently allocation of freshwater for irrigation is steadily declining. Reuse of wastewater is becoming lucrative option especially because of its high plant nutrient content. Based on the findings we can conclude that the wastewater proved beneficial for the crop growth and productivity. Build-up of heavy metals (Cd, Cu, Cr, Zn, Ni and Pb) has been observed in soils irrigated with wastewater as compared to GW irrigated soils, however mobility/ bioavailability of heavy metals was dependent on soil properties. The pollution load index values indicated that the wastewater-irrigated soils were moderately enriched with Cr, Cu, Ni, Pb, Zn and Cd. Difference in metal accumulative behaviour among vegetables has been observed. The increase in metal content of vegetables grown with wastewater was well within the permissible limits of human consumption proposed by Joint FAO/WHO Expert Committee on Food Additives (1992) and Kabata-Pendias and Pendias (1984). Thus, there is no urgent food safety issue concerning use of wastewater for irrigation of vegetable crops.

Acknowledgements

The authors thank to Chairman, Department of Botany, Aligarh Muslim University, Aligarh for providing all the necessary help required at different stages of the study.

References

Ademoroti, C.M.A. 1996. *Standard method for Water and Effluents Analysis.* Foludex press Ltd, Ibadan pp. 22-23, 44-54, 111-112.

Akhtar, N., Inam, A., Inam, A. and Khan, N.A. 2012. Effects of city wastewater on the characteristics of wheat with varying doses of nitrogen, phosphorus and potassium. *Recent Research in Science and Technology* **4**(5):18-29.

Alexander, P.D., Alloway, B.J. and Dourado, A.M. 2006. Genotypic variations in the accumulation of Cd, Cu, Pb and Zn exhibited by six commonly grown vegetables. *Environmental Pollution* **144**(3):736-745.

Amahmid, O., Asmama, S. and Bouhoum, K. 1999. The effect of wastewater reuse in irrigation on the contamination level of food crops by *Giardia* cysts and *Ascaris* eggs. *International Journal of Food Microbiology* **49**:19-26.

APHA 1998. *Standard Methods for the Examination of Water and Wastewater*. 20th Ed., American Public Health Association, Washington, DC.

Ayers, R.S. and Westcot, D.W. 1994. Water quality for agriculture. In: *FAO Irrigation and Drainage Paper* 29 Rev. **1**:1-130.

Baker, A.J.M., Reeves, R.D. and McGrath, S.P. 1991. *In situ* Decontamination of Heavy Metal Polluted Soils Using Crops of Metal-Accumulating Plants- A Feasibility Study. In: R.E. Hinchee and R.F. Olfenbuttel (Eds.). *In Situ Bioreclamation*. Butterworth-einemaan Publishers, Stoneham, MA, pp. 539-544.

Barman, S.C., Sahu, R.K.,Bhargava, S.K. and Chatterjee, C. 2000. Distribution of heavy metals in wheat, mustard and weed grown in fields irrigated with industrial effluents. *Bulletin of Environmental Contamination and Toxicology* **64**:489-496.

Berg, H., Kiibus, M. and Kautsky, N. 1995. Heavy metals in tropical kariba, Zimbabwe. *Water, Air and Soil Pollution* **83**:237-252.

Blumenthal, U.J., Mara, D.D., Peasey, A., Ruiz-Palacios, G. and Stott, R. 2000. Guidelines for the microbiological quality of treated wastewater used in agriculture: Recommendations for revising WHO guidelines. *Bulletin of the World Health Organization* **78**:1104-1116.

Bouhoum, K. and Amahmid, O. 2002. Municipal wastewater reuse for irrigation: Productivity and contamination level of irrigated crops by pathogens. In: *Proceedings of the International Symposium on Environmental Pollution Control and Waste Management*, January 7-10, 2002, Tunis.

Campbell, C.R. and Plank, C.O. 1992. Sample Preperation. In: Dr. C.O. Plank (Ed.) *Plant Analysis Reference Procedures for the Southern Region of the United States*. Southern Cooperative Series Bulletin #368.

Chamberlin, A.C. 1983. Fallout of lead and uptake by crops. *Atmospheric Environment* **17**:693-706.

Faizan, S., Kausar, S. and Akhtar, N. 2014. Influence of wastewater application and fertilizer use on growth, photosynthesis, nutrient homeostatis, yield and heavy metal accumulation in Okra (*Abelmoschus esculentus* L. Moench). *Pakistan Journal of Biological Sciences* **17**(5):630-640.

FAO/WHO Standard. 1992. In: *Wastewater Treatment and use in agriculture*. FAO Irrigation and Drainage Papers p. 47.

Feigin, A., Ravina, I. and Shalhevet, J. 1991. *Irrigation with treated sewage effluent: management for environmental protection*. Springer-Verlag, Berlin, pp. 224.

Fischer, R.A. and Hsiao, T.C. 1968. Stomatal opening in isolated epidermal strips of *Viciafaba*. II. Responses to KCl concentration and the role of potassium adsorption. *Journal of Plant Physiology* **43**:1953-1958.

Ghosh, A.B., Bajaj, J.C., Hassan, R. and Singh, D. 1988. *Soil and water testing methods*. A Laboratory Manual, New Delhi, IARI.

Ghosh, A.K., Bhatt, M.A. and Agrawal, H.P. 2012. Effect of long-term application of treated sewage water on heavy metal accumulation in vegetables grown in Northern India. *Environmental Monitoring and Assessment* **184**(2):1025-1036.

Gupta, S., Nayek, S.,Saha, R.N. and Satpati, S. 2007. Assessment of heavy metal accumulation in macrophyte, agricultural soil, and crop plants adjacent to discharge zone of sponge iron factory. *Environmental Geology* **55**:731-739.

Gupta, S., Satpati, S., Nayek, S. and Garai, D. 2010. Effect of wastewater irrigation on vegetables in relation to bioaccumulation of heavy metals and biochemical changes. *Environmental Monitoring and Assessment* **165**:169-177.

Harrison, R. and Chirgawi, M.B. 1989. The assessment of air and soil as contributors of some trace metals to vegetable plants. 1. Use of a filtered air growth cabinet. *Science of the Total Environment* **83**:13-34.

Heckman, J.R., Angle, J.S. and Chancey, R.L. 1986. Soybean nodulation and nitrogen fixation in soil previously amended with sewage sludge. *Biology and Fertilityof Soils* **2**:181-185.

Iqbal, S., Inam, A.,Inam, A., Sahay, S. and Chalkoo, S. 2012. Balance use of inorganic fertilizers on chilli (*Capsicum annuum* L.) irrigated with wastewater. *Biosciences International* **1**(3):82-89.

Iqbal, S., Inam, A., Inam, A., Ashfaque, F. and Sahay, S. 2016. Potassium and wastewater interaction in the regulation of photosynthetic capacity, ascorbic acid and capsaicin in chilli (*Capsicum annuum* L.) plant. *Agricultural Water Management* (Accepted).

Ismail, B.S., Farihah, K. and Khairiah, J. 2005. Bioaccumulation of heavy metals in vegetables from selected agricultural areas. *Bulletin of Environmental Contamination and Toxicology* **74**:320-327.

Jackson, M.L. 1973. *Soil chemical analysis*. Prentice Hall of India, New Delhi.

Juste, C. and Mench, M. 1992. Long Term Application of Sewage Sludge and its Effects on Metal Uptake by Crops. In: Adriano, D.C. (Ed.) *Biogeochemistry of Trace Metals.* Lewis Publishers, Boca Raton, FL., USA, pp. 159-193.

Kabata-Pendias, A. and Pendias, H. 1984. *Trace elements in Soils and Plants*.CRC Press, Boca Raton, FL.

Lindsay, W.L. and Norvell, W.A. 1978. Development of DTPA soil test for zinc, iron, manganese and copper. *Soil Science Society of America Journal* **42**:421-428.

Liu, D.H., Jiang, W.S. and Hou, W.Q. 2001. Uptake and accumulation of copper by roots and shoots of maize (*Zea mays* L.).*Journal of Environmental Sciences* **13**:228-232.

Liu, W.H., Zhao, J.Z., Ouyang, Z.Y., Soderlund, L. and Liu, G.H. 2005.Impacts of sewage irrigation on heavy metals distribution and contamination in Beijing, China. *Environment International* **31**:805-812.

Lone, M.I., He, Z., Stoffella, P.J. and Yang, X. 2008. Phytoremediation of heavy metal polluted soils and water: Progresses and perspectives. *Journal of Zhejiang University Science B* **9**(3):210-220.

Lubello, C., Gori, R., Nicese, F.P. and Ferrini, F. 2004. Municipal-treated wastewater reuse for plant nurseries irrigation. *Water Research* **38**:2939-2947.

Mapanda, F., Mangwayana, E.N., Nyamangara, J. and Giller, K.E. 2005. The effect of long-term irrigation using wastewater on heavy metal contents of soils under vegetables in Harare, Zimbabwe. *Agriculture, Ecosystems and Environment* **107**:151-165.

Mashiatullah, Riffat, A., Qureshi, M., Niaz, A., Javed, T. and Nisar, A. 2005.Biological quality of groundwater in Rawalpindi/Islamabad. *The Environmental Monitoring* **5**:13-18.

Parveen, T., Inam, A. and Mehrotra, I. 2013. Treated municipal wastewater for irrigation: effect on turnip (*Brassica rapa*). *Desalination and Water Treatment* **51**:5430-5443.

Pescod, M.B. 1992. Wastewater treatment and use in agriculture. In: *FAO Irrigation and Drainage Paper No. 47*, FAO, Rome, p. 125.

Sahay, S., Inam, A. and Iqbal, S. 2013. Effect of wastewater irrigation on soil, metal tolerance and its remediation by four oil yielding *Brassica* cultivars. *International Journal of Environmental Sciences* **4**(2):158-170.

Sahay, S., Inam, A., Inam, A. and Iqbal, S. 2015. Modulation in growth, photosynthesis and yield attributes of black mustard (*B. nigra*cv IC247) by interactive effect of wastewater and fly ash under different NPK levels. *Cogent Food and Agriculture* **1**:1087632

Sauer, D.B. and Burroughs, R. 1986. Disinfection of seed surfaces with sodium hypochlorite. *Phytopathology* **76**:745-749.

Sawidis, T., Chettri, M.K., Papaionnou, A., Zachariadis, G. and Stratis, J. 2001. A study of metal distribution from lignite fuels using trees as biological monitors. *Ecotoxicology and Environmental Safety* **48**:27-35.

Segura, M.L., Grandos, M.R., Moreno, J. and Urrestarazu, M. 2004. Response of greenhouse melon and tomato crops to wastewater irrigation. *ActaHorticulturae* **633**:391-396.

Sharma, R.K., Agrawal, M. and Marshall, F. 2007. Heavy metal contamination of soil and vegetables in suburban areas of Varanasi, India. *Ecotoxicology and Environmental Safety* **66**(2):258-266.

Sharma, R.K., Agrawal, M. and Marshall, F. 2006. Heavy metals contamination in vegetables grown in wastewater irrigated areas of Varanasi, India. *Bulletin of Environmental Contamination and Toxicology* **77**:312-318.

Sheng, P.H., Zhou, A.X. and Li, P.J. 2001.*Pollution Ecology*. Science Press, Beijing.

Sinha, S., Gupta, A.K., Bhatt, K., Pandey, K., Rai, U.N. and Singh, K.P. 2006. Distribution of metals in the edible plants grown at Jajmau, Kanpur (India) receiving wastewater irrigated tannery wastewater: Relation with physic-chemical properties of the soil. *Environmental Monitoring and Assessment* **115**:1-22.

Soumare, M., Tack, F.M.G. and Verloo, M.G. 2003.Effect of a municipal solid waste compost and mineral fertilization on plant growth in two tropical agricultural soils of Mali. *Bioresource Technology* **86**:15-20.

Staniforth, A.R. and Smith, S.R. 1991. The use of sewage sludge-straw based compost as a fertilizer for agriculture. *Agricultural Progress* **66**:23-35.

Steel, R.G.D. and Torrie, V.J.H. 1996. *Principles and Procedures of Statistics*. McGraw Hill, New York.

Tadesse, T., Dechassa, N.,Bayu, W. and Gebeyehu, S. 2013. Effect of farmyard manure and inorganic fertilizer application on soil physic-chemical properties and nutrient balance in rain-fed lowland rice ecosystem.*American Journal of Plant Sciences* **4**:309-316.

Tak, H.I., Babalola, O.O.,Huyser, M.H. and Inam, A. 2013. Urban wastewater irrigation and its effect on growth, photosynthesis and yield of chickpea under different doses of potassium. *Soil Science and Plant Nutrition* **1**:1-12.

Tamoutsidis, E., Lazaridou, M., Papadopoulos, I., Spanos, T., Papathanasiou, F., Tamoutsidou, M., Mitlianga, P. and Vasiliou, G. 2009. The effect of treated urban wastewater on soil properties, plant tissue composition and biomass productivity in berseem clover and corn. *Journal of Food, Agriculture and Environment* **7**:782-786.

WHO 2006. Guidelines for the Safe Use of Wastewater, Excreta and Greywater. In: *Wastewater Use in Agriculture*, Volume 2, WHO (Ed.). World Health Organization, Geneva, Switzerland.

Wilson, B. and Pyatt, F.B. 2007. Heavy metal dispersion, persistence, and bioaccumulation around an ancient copper mine situated in Anglesey. UK. *Ecotoxicology and Environmental Safety* **66**:224-231.

Xue, Z.-J., Liu, S.-Q., Liu, Y.-L. and Yan, Y.-L. 2012. Health risk assessment of heavy metals for edible parts of vegetables grown in sewage-irrigated soils in suburbs of Baoding city, China. *Environmental Monitoring and Assessment* **184**:3503-3513.