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Heavy metal and microbial loads in sewage irrigated vegetables of Kabul, Afghanistan

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Abstract

Little is known about the heavy metal and microbial contamination of vegetables produced in Central Asian cities. We therefore measured the concentration of cadmium (Cd), copper (Cu), lead (Pb), and zinc (Zn) and of faecal pathogens (Coliform bacteria, *Salmonella* sp., *Shigella* sp., *Ascaris lubricoides*, *Entamoeba* sp. and pinworms [*Oxyuris vermicularis* syn. *Enterobius vermicularis*]) in soil, irrigation water, and marketed vegetables of Kabul City, Afghanistan. Leaf Pb and Zn concentrations of leafy vegetables were with 1–5 and 33–160 mg kg⁻¹ dry weight (DW) several-fold above respective international thresholds of 0.3 mg Pb kg⁻¹ and 50 mg Zn kg⁻¹. The tissue concentration of Cu was below threshold limits in all samples except for spinach in one farm. Above-threshold loads of microbes and parasites on vegetables were found in five out of six gardens with coliforms ranging from $0.5-2 \times 10^7$ cells $100g^{-1}$ fresh weight (FW), but no Salmonella and Shigella were found. Contamination with 0.2×10^7 eggs $100g^{-1}$ FW of *Ascaris* was detected on produce of three farms and critical concentrations of *Entamoeba* in a single case, while *Oxyuris vermicularis*, and *Enterobius vermicularis* were found on produce of three and four farms, respectively. Irrigation water had *Ascaris*, Coliforms, *Salmonella*, *Shigella*, *Entamoeba*, and *Oxyuris vermicularis* syn. *Enterobius vermicularis* ranging from 0.35×10^7 to 2×10^7 cells 1^{-1} . The heavy metal and microbial loads on fresh UPA vegetables are likely the result of contamination from rising traffic, residues of the past decades of war and lacking treatment of sewage which needs urgent attention.

Keywords: Faecal contamination, Food safety, Urban agriculture, Vegetable quality

1 Introduction

As an important subsistence and income generating activity urban and peri-urban agriculture (UPA) is of particular importance in Kabul city which is rapidly growing as a consequence of the continuing arrival of war-refugees and infrastructural constraints in the countryside where the majority of the country's population live (CSO, 2010). Lacking collection and disposal infrastructure lead to 70% of Kabul's total solid waste (at least 300 t day⁻¹) being accumulated at the road-sides and in backyards, drains, rivers and open places

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where it represents a significant environmental hazard and to which the effluents of the virtually non-existent sewage system have to be added (Afghanistan Online, 2010; UN HABITAT, 2010). At present most sewage is disposed of in domestic drainage pits and shallow open sewage channels along the streets, threatening the largely shallow aquifers with microbial contamination and heavy metals which add to the already existing loads of borate and nitrate (Houben et al., 2009). A recent US geological survey reported 70% of all wells in the Kabul basin being contaminated by faecal bacteria (Akbari et al., 2007). This likely contributes to the high infant mortality as a consequence of water-borne diseases (UNICEF, 2008) even if possible cause-effect relationships between sewage water-related contamination of leafy vegetables (Drechsel et al., 2000; Sonou, 2001) and human health problems seem still to be largely ignored by consumers and policy makers alike. The same is true for heavy metals that are of growing concern in UPA produce of many developing countries (Qadir *et al.*, 2000; Abdu *et al.*, 2011) as their accumulation via the atmosphere-soil-plant chain in the human body can lead to a variety of health disorders, including cancer (Voutsa *et al.*, 1996; Anikwe & Nwobodo, 2002; Türkdogan *et al.*, 2002; Liu *et al.*, 2006; Khan *et al.*, 2008).

Given lacking quantitative data on UPA produce contamination with microbial pathogens and heavy metals, this study aimed at determining the levels of both types of contaminants in irrigation water, UPA soils, and leafy vegetables using five representative gardens in the centre of Kabul city.

2 Materials and Methods

2.1 The agro-ecological setting and physical structure of the study area

The monitored gardens (N $34^{\circ} 29' 59.76'' \to 69^{\circ}09' 22.06''; 1,765 \text{ m a.s.l.}$) were distributed along a 10 km transect crossing one of the most densely inhabited parts of Kabul city from Bagh-e-Rayees to Hootkhail in an E-W direction (Figure 1). This area has an old irrigation infrastructure with temporary water courses along the Kabul River and sewage channels.

During April 2008 a baseline survey of 100 farms had been conducted for which farms were selected to represent the major agricultural land use systems (Safi *et al.*, 2011). Household selection followed a cluster analysis of production systems and socio-economic status (family composition, household members, on-farm income, off-farm income, education). Based on the survey results five farm households were selected, their garden areas mapped with a handheld GPS and the cropping system recorded (Figure 2). This was followed by a sampling of the irrigation water, soil, and agricultural produce as described below.

2.2 Measurements of heavy metal and microbial contamination

2.2.1 Vegetables

For heavy metal analysis five sub-samples per plot of edible parts of the economically relevant vegetables lettuce (*Lactuca sativa* L.), radish (*Raphanus sativus* L.), coriander (*Coriandrum sativum* L.), mint (*Mentha arvensis* L.), onion (*Allium cepa*), leek (*Allium ampeloprasum* var. *porrum* L.), spinach (*Spinacia oleracea* L.), garden cress (*Lepidium sativum* L.), turnip (*Brassica rapa* var. *rapa* L.), and the forage crop alfalfa (*Medicago sativa* L.) were randomly collected from each

garden over a period of seven months (April-October 2009). The samples were washed with distilled water, sliced, pre-dried on a sheet of paper and subsequently oven-dried to constant weight at 65°C for 48 hours, ground with a ceramic-coated grinder (Liu et al., 2006) and stored in 100 ml PE bottles. For the same species and management system samples were pooled across sampling periods. Heavy metal concentrations (Cd, Pb, Cu, and Zn) were determined according to Liu et al. (2006) and Schumacher et al. (1993) using a microwave assisted digestion procedure. To this end 0.2 \pm 05 g of a homogenized sample was digested under pressure in Teflon vessels with 3 ml of HNO₃ (65%) and 1 ml of H₂O₂ (30%). After completion of the digestion, the solutions were filtered and brought to 50 ml with distilled water. Concentrations of Cd, Pb, Cu, and Zn were determined in duplicates by a GBC 906 atomic absorption spectrophotometer (AAS; GBC Scientific Equipment LLC, Hampshire, IL, USA).

For microbial analysis in 2008 a composite sample of freshly eaten vegetable parts (leaf, stem or storage root) consisting of 5–10 individual plants was harvested and stored at <10°C until parasitological analysis (Anh *et al.*, 2007). Protozoan parasites cysts and eggs in vegetables were counted after washing 10 g plant sample with 100 ml of sterile distilled water. Subsequently, the samples were pulsified (Pulsifier®, Filtaflex, Almonte, ON, Canada), and concentrated by centrifugation for 10 minutes to a final volume of 2 ml. The samples were then processed according to the modified Bailenger method (Ayres & Mara, 1996).

2.2.2 Irrigation water

To determine annual changes in microbial loads of the regularly surface applied sewage irrigation water (flood irrigation system) on two random dates, 15th June and 15th August 2008, 11 composite water samples were collected from five points in each of the five gardens between 8-11 am and immediately transferred to the laboratory for analysis of Coliform bacteria, Salmonella, Shigella, Ascaris lubricoides, Entamoeba, Oxyuris vermicularis syn. Enterobius vermicularis according to the WHO method (Ayres & Mara, 1996). The samples were sedimented for 2 hrs and 90% of the supernatant removed. Subsequently, the sediments were centrifuged for 15 min. Again the supernatant was removed and the sample re-centrifuged before the pellet was suspended in an volume equal to that of the pellet of CH₃C(O)CH₂CO₂H buffer amended by two volumes of CH₃COOCH₂CH₃ and vigorously shaken in a vortex. Thereafter the mixture was centrifuged again for 15 min, re-suspended in five volumes of ZnSO₄ solution and an aliquot transferred to a McMaster counting



Fig. 1: Map of Kabul city indicating the location of the five vegetable gardens monitored in 2008 and 2009.



Fig. 2: *GIS-based maps showing the plot structure and species grown in four of five studied urban vegetable gardens in Kabul, Afghanistan.*

slide (Chalex Corp., Wallowa, OR, USA). The number of eggs was calculated as:

$$N = \frac{AX}{PV} \tag{1}$$

where N = number of eggs per liter of sample; A = number of eggs; X = volume of final product (ml); P = volume of the McMaster slide (0.3 ml); V = original sample volume (liters)

The number of faecal coliform bacteria was determined using the most probable number (MPN) method based on test tubes with medium A-1 (Hach Co., Loveland, Co, USA; Powell *et al.*, 1979) incubated at 44°C over night. The number and distribution of positive tubes (acidification or gas production or both) were used to obtain the populations of coliform bacteria from MPN tables (Ayres & Mara, 1996; Amoah *et al.*, 2005).

2.2.3 Soil

In each of the five gardens surface soil (0–0.20 m depth) was collected twice per year (Spring and Fall of 2008 and 2009), air dried at room temperature, and passed through a 2 mm nylon sieve mesh in order to remove sand, larger particles and debris. Of each pooled sample per year (spring and fall) 0.3 ± 05 g was digested under pressure in Teflon vessels with 2 ml of HNO₃ (65%) and 6 ml of HCl (37%). After completion of the digestion, the solutions were filtered and brought to 50 ml with distilled water. Concentrations of Cd, Pb, Cu, and Zn were determined on duplicate samples as above.

3 Results

3.1 Heavy metals

Wastewater concentrations of the studied heavy metals were, regardless of the season, in all gardens orders of magnitude below the threshold levels reported by Drechsel et al. (2010, Table 1). Even prolonged use of this irrigation water led to heavy metal concentrations in the surface soil of the five gardens that were uncritical (Table 2). Heavy metal concentrations in the edible plant parts of the harvested vegetables, however, were consistently below the safety threshold only for Cd (Table 3). According to WHO standards all samples were heavily contaminated with Pb, whereby spinach in garden 1 and cress in garden 2 exceeded the WHO safety threshold 11- and 15-fold, respectively and where also above the less strict Indian standards. For Cu only spinach from garden 1 exceeded both the WHO and the Indian standards, but for Zn 50% of all samples across gardens were above the safety thresholds; highest concentrations were found in spinach of garden 1 and cress of garden 2 (Table 3).

Table 1: Concentration of heavy metals $(\mu g l^{-1})$ in irrigation water used for urban vegetable production in Kabul, Afghanistan.

No. of garden	Season	Cd	Pb	Си	Zn
1	Spring *	< 0.15	<2.5	<1.0	<20
1	Fall	< 0.15	<2.5	<1.0	<20
2	Spring	< 0.15	<2.5	<1.0	<20
2	Fall	< 0.15	<2.5	<1.0	<20
3	Spring	< 0.15	<2.5	<1.0	<20
3	Fall	< 0.15	<2.5	11.3	<20
5	Spring	< 0.15	<2.5	<1.0	<20
5	Fall	< 0.15	<2.5	<1.0	<20
Safety Threshol $(\mu g l^{-1})$	ld †	10	5000	200	2000

Seasons of sampling

[†] Source: Drechsel et al. (2010)

3.2 Parasites and microbes

Microbes and parasites in irrigation water were several times higher than the WHO safety thresholds. Coliform loads were four-fold higher in garden 3 and 5 than in garden 2 and 6. Salmonella was detected only in the irrigation water of garden 3 and 5, while Shigella was higher in garden 2 and 6 than in gardens 3 and 5. Ascaris eggs were, except for garden 2, in all gardens above the threshold. There was also considerable difference between irrigation waters for *Entamoeba* and *Oxyuris vermincularis* (Table 4).

In all gardens coliforms from the irrigation water were transferred to the agricultural produce. Irrigation water induced produce contamination also occurred for *Enterobius vermicularis* syn *Oxyuris vermicularis* in four gardens, for *Ascaris lubricoides* in three gardens and for *Entamoeba* in one garden, while *Salmonella* sp. and *Shigella* sp. were not transferred to the crops (Table 5).

4 Discussion

4.1 Heavy metal loads

The low heavy metal concentrations in irrigation water were surprising and may in view of the still important contamination of agricultural produce reflect the effects of recent (post-war) reductions in water-related contamination sources as well as the contribution of unquantified amounts of dust to the overall loads. The generally low levels of Cd in the vegetable samples may indicate the effect of the high pH of Kabul's garden soils which reduces the availability of Cd for crops (Kuo *et al.*, 1985; He & Singh, 1994). Another reason may be the

No. of garden	Texture	pH^*	$C_{\rm org}$ $(g \ kg^{-1})$	Cd	<i>Pb</i> (<i>m</i>	$\frac{Cu}{g \ kg^{-1}}$	Zn
1	silt loam	8.01	30	0.19	26	66	184
2	sandy loam	8.03	18	0.18	25	52	154
3	silt loam	7.96	22	0.18	33	62	140
4	sandy loam	8.26	20	0.20	30	50	113
5	sandy loam	8.02	18	0.15	26	53	129
Thresholds							
Indian [†]				3–6	250-500	135-270	300
EU [‡]				3	300	140	300-600
UK ‡				3	300	80-200	200-300
USA ‡				20	150	170	1400
				-			

Table 2: Physical properties, pH, organic carbon (C_{org}) and heavy metal concentrations in the surface soil (0–0.20 m depth) of five urban vegetable gardens in Kabul, Afghanistan.

* Measured in a 1:2.5 soil:water suspension

Sources: Awashthi $(2000)^{\dagger}$ and Canadian Council of Ministers of the Environment (CCME) $(2001)^{\dagger}$

Table 3: Concentration of the heavy metals cadmium (Cd), lead (Pb), copper (Cu) and zinc (Zn; all in mg kg⁻¹ dry weight) in vegetables grown in waste-water irrigated gardens of Kabul, Afghanistan.

No. of garden	Crop name	Botanical name	Cd	Pb	Си	Zn
1	Coriander	Coriandrum sativum	n.d.*	1.4	8.8	45.5
1	Radish	Raphanus sativus L.	n.d.	1.5	5.2	55.1
1	Spinach	Spinacia oleracea L.	0.14	3.4	53.7	86.9
1	Leek	Allium porrum L.	n.d.	1.7	10.6	43.1
2	Spinach	Spinacia oleracea L.	n.d.	1.3	8.5	75.4
2	Lettuce	Lactuca sativa L.	n.d.	1.5	8.2	45.0
2	Onion	Allium cepa	n.d.	1.3	5.7	62.5
2	Radish	Raphanus sativus L.	n.d.	1.9	5.9	62.9
2	Garden cress	Lepidium sativum	0.14	4.6	7.0	160.1
2	Mint	Mentha arvensis L.	0.05	2.5	13.7	38.5
3	Turnip	Brassica rapa var. rapa	n.d.	2.0	3.0	35.3
3	Spinach	Spinacia oleracea L.	n.d.	2.1	10.2	64.9
3	Onion	Allium cepa	n.d.	0.9	7.1	39.3
4	Alfalfa	Medicago sativa L.	n.d.	0.8	6.6	34.2
4	Mint	Mentha arvensis L.	n.d.	2.0	9.6	72.1
5	Spinach	Spinacia oleracea L.	n.d.	1.6	6.2	42.9
5	Radish	Raphanus sativus L.	n.d.	0.9	4.5	53.6
5	Mint	Mentha arvensis L.	n.d.	0.8	10.0	32.7
Safety Thresholds						
WHO †			0.2	0.3	40	50
India [‡]			1.5	2.5	30	50

 $* < 10 \,\mu g \, kg^{-1}$ or not detectable

[†] FAO/WHO (2001), Joint Codex Alimentarius Commission. [‡]Indian limit, Awashthi (2000)

No. of garden	Water source	Coliforms	Salmonella	Shigella	Ascaris lubricoides	Entamoeba	Enterobius vermicularis*
1	Well+sewage [†]	_	_	_	1.0×10^{7}	_	_
2	Sewage+river	0.5×10^{7}	_	2.5×10^{7}	_	_	0.5×10^{7}
3	Sewage+river	2.0×10^{7}	2.0×10^{7}	1.0×10^{7}	2.0×10^{7}	0.5×10^{7}	1.0×10^{7}
4	Sewage+river	2.0×10^{7}	1.0×10^{7}	1.5×10^{7}	1.75×10^{7}	0.9×10^{7}	1.25×10^{7}
6	Sewage+river	0.5×10^{7}	—	2.5×10^{7}	1.0×10^{7}	1.5×10^{7}	0.5×10^{7}
Safety limit [‡]		1000	1000	1000	1	1	1

Table 4: Most probable number data of faecal pathogens in sewage water used for irrigation of urban vegetables in Kabul,

 Afghanistan.

* syn. Oxyuris vermicularis

[†] Well water was dominated by sewage

[‡] Source WHO (1989)

Table 5: Most probable numbers of faecal pathogens in wastewater irrigated urban vegetables in Kabul, Afghanistan.

No. of garden	Crop	Coli form bacteria	Salmonella	Shigella	Ascaris lubricoides	Entamoeba	Enterobius vermicularis *
1	Garden cress	1.0×10^{7}	_	-	_	_	_
2	Lettuce	0.5×10^{7}	_	_	_	_	0.4×10^{7}
3	Coriander+radish	1.0×10^{7}	_	_	0.2×10^{7}	_	0.7×10^{7}
5	Radish+coriander	2.0×10^{7}	-	_	0.2×10^{7}	0.5×10^{7}	0.4×10^{7}
6^{\dagger}	Onion+radish	1.5×10^{7}	_	_	0.2×10^{7}	_	0.4×10^{7}

* syn. Oxyuris vermicularis

[†] Note: Due to a sudden crop change garden 4 was replaced by garden 6

widespread absence of Cd sources. The soil and produce contamination with Pb may be the result of fuel combustion from the rapid increase in traffic and city waste (Maleki & Zarasvand, 2008). Nabulo *et al.* (2006) reported atmospheric deposition to be the dominant pathway for Pb to leafy vegetables. The overall low levels of Cu may again be the results of lacking Cu-using industries in the Kabul area. The high Zn loads in the studied vegetables may partly come from war-related junk that for long caused some rivers, streams, and ditches to be filled with broken tanks, military vehicles and bullet cartridges.

4.2 Contamination with parasites and microbes

The high levels of bio-contaminants in the irrigation water sources may be due to septic wastes from the city's hospitals, and sewage from schools and residential areas. An elevated contamination of vegetables with parasite eggs excreted by human and animals was also reported by Uga *et al.* (2009) from Hanoi, Vietnam and

by Abougrain *et al.* (2010) from Tripoli, Libya. Significant contamination of vegetables with *Amoeba* sp., *Ascaris lubricoides* and *Enterobius vermicularis* were also found in neighbouring Iran by Gharavi *et al.* (2002), but their values were lower than ours. In Kabul many farmers accept the direct unloading of sewage tanks into their gardens given the free input of plant nutrients contained therein.

5 Conclusions

Though the data of this study only indicate excessive Pb and Zn loads as well as pathogen contaminations of UPA produce to exceed international thresholds levels, the high incidence of intestinal diseases and diarrhoea in Kabul's population calls for further surveys to confirm our results. Improvements in the Kabul's sewage infrastructure are nevertheless urgently needed to eliminate potential health risks and decrease the widespread odour nuisance.

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