Egypt. J. Bot., **Vol. 60,** No.3 pp. 855-863 (2020)

Egyptian Journal of Botany
http://ejbo.journals.ekb.eg/

Accumulation of Cobalt in Soils and Forages Irrigated with City Effluent

Zafar Iqbal Khan(1), Tasneem Ahmad(2), Hareem Safdar(1), Muhammad Nadeem(3), Kafeel Ahmad(1), Humayun Bashir(1), Mudasra Munir(1), Ilker Ugulu(4),Yunus Dogan(5)#

(1)Department of Botany, University of Sargodha, Sargodha, Pakistan; (2)Paki Thatti Research and Development Farm, Toba Tek Singh, Pakistan; (3)Institute of Food Science and Nutrition, University of Sargodha, Sargodha, Pakistan; (4)Faculty of Education, Usak University, Usak, Turkey; (5)Buca Faculty of Education, Dokuz Eylul University, Izmir, Turkey.

DESPITE of its nutrient content, sewage water includes different contaminants responsible for the pollution of soil and plants. In this study, an experiment was conducted at the University of Sargodha to evaluate the danger of the presence of cobalt in forages irrigated with city effluent. Moreover, the health risks associated with livestock feed on these contaminated forages were evaluated. Cobalt accumulation in different parts of plants was analysed by atomic absorption spectrophotometer (AA-6300 Shimadzu Japan). Some critical indices like pollution load index, bioconcentration factor and health risk index were also determined. The observed cobalt values in water samples used for irrigation were 0.164 mg/L in tap water and 0.191mg/L in sewage water. The highest cobalt concentration was observed in the root of the winter forage *Trifolium alexandrinum* (1.560mg/kg) irrigated with sewage water, while the minimum concentration was recorded in *Sorghum bicolor*, a summer forage irrigated with tap water (0.085mg/kg). The maximum bioconcentration factor value for cobalt was 7.7 in the winter crop of *T. resupinatum*. The maximum pollution load index, daily intake and health risk index values for cobalt were 0.8910,0.026 and 0.6104, respectively. All of these maximum values were determined for the samples irrigated with sewage water. In summary, the cobalt contents in soil and plant samples significantly increased when the samples were irrigated with sewage water as compared to tap water.

Keywords: Accumulation, Cobalt, Health risk, Plant, Trace element.

Introduction

Heavy metals have a great ability to accumulate in the environmental matrices and to induce toxicity in biota, even in small quantities (Dogan et al., 2014a; Unver et al., 2015; Ugulu et al., 2019a). Some microelements are necessary for plants as well as humans, but if their quantity increased beyond a specific limit, these elements like heavy metals can be toxic (Ugulu et al., 2016). The heavy metals released from the natural sources and through human activities are likely to accumulate in soil, thus the soil act as a sink for these toxic metals (Ahmad et al., 2018a, b; Khan et al., 2020). For instance, heavy metals may reach the soil through automobiles, industries, land applications, fertilizers, pesticides, wastewater irrigation, spillage of petrochemicals and also through the atmospheric depositions (Durkan et al., 2011; Ugulu et al., 2012).

Beside its use as an additional irrigation source, wastewater delivers some beneficial nutrients, particularly carbon-based matter, nitrogen and phosphorus that could improve soil productiveness (Khan et al., 2018a, b; Ahmad et al., 2019). However, wastewater also contains some heavy metal that could contaminate the crops and decrease the productivity (Khan et al., 2019a, b, c). Unfortunately, heavy metals cause harmful effects for humans and for the whole ecosystem because of their persistent nature (Baslar et al., 2009; Dogan et al., 2010; Dogan et al., 2014b).

#Corresponding author email: yunus.dogan@deu.edu.tr , 0000-0001-9404-6031

Received 18/12/ 2019 ; Accepted 28/7/ 2020
DOI :10.21608/ejbo.2020.19829.1394
Edited by: Dr. Ahmed M. Saleh, Biology Department, Faculty of Science at Yanbu, Taibah University, KSA.

One of the major pollutants that present in city effluents, used for irrigation in some developing countries, is cobalt (insert citation). Cobalt is not an essential element for cultivated plants (Ugulu, 2015a). Since excessive amounts of cobalt in agricultural areas have toxic effects on cultivated plants, it is undesirable to be found in agricultural soils (Khan et al., 2018c). For this reason, the amount of cobalt in the soil solution is extremely important for plants. The root apices of plants are impassable with heavy metals due to their immature cells and low-density cell walls. The root cells absorb the toxic metals from the contaminated soil and then these elements are transferred to the above-ground parts of the plants (Tung & Temple, 1996).

Cobalt (Co) is not classified as an essential element for plants; however, it is often described as useful. Some plant species have the ability to remove metals (such as Co) from the soil, thereby reducing their harmful effects on the environment (Ugulu, 2015a). Cobalt is known to cause irreversible damage to a number of vital metabolic components, plant cell walls and cell membranes. When the plants became maturity, such as plants such as perennial ryegrass, is likely to a tendency to decrease the content of cobalt (Fleming, 1970; Nadeem et al., 2019). Co-vitamin B_{12} does not occur in plants. While it is stated that normal cobalt concentrations in plants are as low as 0.1-10µg g^{-1} dry weight, its beneficial role as a trace element has been described. Trace elements are essential for normal metabolic functions in plants, but at higher concentrations, these metals are toxic and can seriously interfere with physiological and biochemical functions (Ugulu, 2015b).

Pakistan is an important agricultural country and the most important economic activity of the country is agriculture. However, there is a shortage of clean water in the country and wastewaters such as industrial wastewater and sewage water are widely used in agricultural irrigation. In this direction, the current study was performed to monitor the cobalt accumulation in forages grown with sewage water, to define the transport of cobalt from soil to plants, and to determine the pollution level and health risk originated from cobalt.

Materials and Methods

Study site

This experimental study was conducted by growing winter and summer crops at Department of Botany, University of Sargodha, Pakistan. In this region, summers are extremely hot while winters are cold. Temperature ranges from 0ºC to 50ºC in Sargodha. Jhelum River is located on the north and west side of Sargodha while Chenab River is located on the east side. In Pakistan, where the most important economic resource is agriculture, there is a water shortage, and besides these river resources, wastewater is used for irrigation purposes. Primary crops grown in the region are sugarcane and rice. Due to its large citrus production, the city is well-known in Pakistan.

Plant cultivation

Summer: 70 pots of plants having fertile soil were seeded with summer forage; *Cyamopsis tetragonoloba, Echinochloa colona, Pennisetum typhoideum, Sesbania rostrate, Sorghum bicolor, Sorghum vulgare* and *Zea mays* crop. Thirty-five of the total of 70 pots were supplied with tap water and 35 pots with water from sewage source regularly.

Winter: Six crops were sown; *Brassica campestris, Brassica juncea, Brassica napus, Medicago sativa, Trifolium alexandrinum,* and *Trifolium resupinatum.* Sixty pots having fertile soil were sown with winter crops. Thirty pots were grown with from tap water (control) and 30 pots with water from sewage source.

Sample collection

Plastic bottles were washed with distilled water. Water from tap and sewage source was collected in 100mL plastic containers. The samples were saved against actions of microorganisms by adding 1mL HNO_3 and kept in a refrigerator. Similarly, soil samples irrigated with tap water and sewage were obtained, and sun-dried. After sun drying, these samples were oven-dried. Plants samples were dried in the same manners after washing with clean water. After drying, samples were ground and digested to analyse for cobalt contents.

Metal analysis

Atomic absorption spectrophotometer (AA-6300 Shimadzu Japan) was served to analyse cobalt contents in digested samples. First standards were run to draw standard calibration curves and then samples were estimated.

Statistical analysis

Data obtained from analysing cobalt concentrations of water, soil and plant samples were subjected to analysis of variance technique by standard procedures using Minitab 16 software.

Bioconcentration factor

Transfer of metals from soils to plants is the bioconcentration factor (BCF). Cui et al. (2004) reported the procedure for determining as the rate between the amount of specific elements in the plant and the same elements in the soil as follow:

BCF= Metalcontentsin plant (mg/kg)/ Metal contentsin soil (mg/kg)

The pollution load index

The pollution load index (PLI) of cobalt was calculated using the following equation (Liu et al., 2005) :

PLI = Metal contents in the soil/ Standard value of the metal in the soil

The standard value of cobalt was 9.1 mg/kg (Liu et al., 2005).

Daily intake of metal

Daily intake of metal (DIM) of cobalt was calculated using the following equation (Chary et al., 2008) :

$$DIM= C_{cobalt} \times D_{food\ intake} / B_{average\ weight}$$

where C_{cobalt}= Cobalt contents in plants, $D_{food\ intake}$= Daily intake of plants, $B_{average\ weight}$= Average body weight. The average body weight 550kg was used to calculate the daily intake of metals, and total consumption of plant by an animal per day was 12.5kg per animal (Khan et al., 2019d).

Health risk index

To determine the exposure of metals to animals through digestion of contaminated plants health risk index (HRI) was determined. It is calculated by dividing daily ingestion of metals in foods by the oral reference dose (R_fD) (USEPA, 2010).

$$HRI = DIM/R_fD$$

R_fD values for cobalt was 0.043mg/kg per day (USEPA, 2010).

Results and Discussion

Metal accumulation in water samples

Significant effect of treatments ($P< 0.05$) was observed on the cobalt contents in the tap water and sewage used for plants. The observed cobalt value was 0.164mg/L in tap water and 0.191mg/L in sewage water (Table 1). Permissible maximum limit of cobalt accumulation in water was reported as 0.05mg/L by WHO, FAO, Standard Guidelines in Europe (Chiroma et al., 2014) and USEPA (2010). The cobalt value in the present study was higher than this limit. According to these results, it can be concluded that there is cobalt pollution in the study area. On the other hand, the cobalt value was higher than 0.1mg/L established by Tunisian Standard (1989). Also, both cobalt amounts of water were higher than observed by Khaskhoussy et al. (2013) (0.0003mg/L) in freshwater and treated wastewater (0.056mg/L). The high cobalt accumulation in water samples might be due to sources such as paints/pigments, supplements, ointments, medicines and food products reported as primarily domestic sources of cobalt pollution in waters (Khan et al., 2018d).

TABLE 1. The concentration of cobalt in water samples (mg/L).

Irrigation water		Mean square
Tap	Sewage	
0.164±0.0214	0.191±0.0238	0.002**
Df	1	
Error	9	
Permissible limit[a]	0.1mg/L	

**:Significant at 0.01 level,Source.

[a]Tunisian Standards (1989)

Metal accumulation in soil samples

According to the plant grown in the soil samples, significant ($P< 0.05$) effect of treatments was noticed on the cobalt contents in soil planted with *B. campestris, B. napus, C. tetragonoloba, M. sativa, P. typhoideum, S. bicolor, S. rostrata, S. vulgare, T. alexandrinum* and *Z. mays,* while non-significant effect was seen in *E. colona, T. resupinatum, B. juncea*. The cobalt contents in plants treated with water from tap were in sequence of: *Z. mays> B. napus> P. typhoideum> B. campestris> M. sativa> T. resupinatum> C. tetragonoloba> E. colona> S. rostrata> B. juncea> S. bicolor> T. alexandrinum> S. vulgare* while the cobalt contents in in plants grown on water of sewage were in sequence of: *T. resupinatum> Z. mays> B. campestris> P. typhooideum> B. napus> B. juncea> E. colona> T. alexandrinum> S. vulgare> C. tetragonoloba> M. sativa> S. rostrate> S. bicolor.* While the maximum mean cobalt content in the soil was 8.109mg/kg seen in *T. resupinatum* grown in winter and the minimum cobalt content was 0.074mg/kg seen in *S. bicolor*in summer crop (Table 2). Permissible maximum limit of cobalt accumulation in soil was reported as 65mg/kg by USEPA (2010). The cobalt value in the present study was lower than this limit. However, cobalt contents found by Khan et al., (2015) (12.12 to 15.75mg/kg) were higher than the present study. Khaskhoussy et al. (2013) found a higher range for cobalt in soil watered with wastewater as 8.00–27.6mg/kg. Ahmad et al. (2014) examined the heavy metal accumulation in the soil samples irrigated with wastewater and tap water in their study in Khushab, Pakistan, and found that the cobalt accumulation in the soil irrigated with sewage water (20.2mg/kg) was more than irrigated with tap water (13.5mg/kg). As mentioned in this study, it was concluded that heavy metal accumulation was higher as a result of irrigation with sewage water. Metals in the ground may be accrued directly by wastewater composition or by enhancing the solubility of metals in soil due to the activity of acidification or chelation of used water (Ugulu et al., 2009; Khan et al., 2019d).

TABLE 2. The concentration of cobalt in soil samples (mg/kg).

Soils	**Irrigation water**		**Mean square**
	Tap	**Sewage**	
	Summer		
Z. mays	2.063±0.0293	2.075±0.0164	0.001**
P. typhoideum	1.742±0.0138	1.869±0.0396	0.013*
C. tetragonoloba	0.111±0.0078	0.152±0.0172	0.004**
S. rostrata	0.087±0.0053	0.146±0.0120	0.009**
E. colona	0.090±0.0157	0.354±0.0253	0.174^{ns}
S. bicolor	0.074±0.0098	0.113±0.0176	0.004**
S. vulgare	0.052±0.0093	0.162±0.0116	0.031*
	Winter		
B. campestris	1.292±0.0021	2.072±0.0164	1.523^{ns}
B. napus	1.773±0.0395	1.859±0.0050	0.018*
T. resupinatum	0.148±0.0016	8.109±0.0134	15.484^{ns}
B. juncea	0.097±0.0181	0.358±0.0245	0.170^{ns}
M. sativa	0.113±0.0078	0.158±0.0174	0.005**
T. alexandrinum	0.053±0.0082	0.170±0.014	0.034*
df	1	Error	9
Permissible limit^{a}		65mg/kg	

*,**: Significant at 0.05 and 0.01 levels, ns: Non–significant, Source:^{a}USEPA (1997)

Metal accumulation in root samples

All proceedings havea significant positive effect (P< 0.05) on cobalt amounts in root samples according to variance analysis in forages. The cobalt content in roots of forages grown with supply of tap water was in sequence: *T. alexandrinum*> *T. resupinatum*> *S. rostrata*> *S. vulgare*> *B. campestris*> *C. tetragonoloba*> *P. typhoideum*> *Z. mays*> *M. sativa*> *E. colona*> *B. juncea*> *S. bicolor*> *B. napus*. The cobalt content in roots of forages grown with the supply of water from sewage source was in sequence of *T. alexandrinum*> *B. napus*> *T. resupinatum*> *P. typhoideum*> *C. tetragonoloba*> *M. sativa*> *S. rostrata*> *B. campestris*> *B. juncea*> *Z. mays*> *E. colona*> *S. vulgare*> *S. bicolor*. The highest cobalt concentration in the root of *T. alexandrinum* (1.560mg/kg) in winter forage and minimum concentration in *S. bicolor* (0.085mg/kg) a summer forage (Table 3). Permissible maximum limit of the cobalt accumulation in plants was reported as 50mg/kg by WHO (1996), FAO/WHO (2001), Standard Guidelines in Europe (Chiroma et al., 2014). Cobalt values in the present study were lower than this limit in plant samples. Ahmad et al. (2014) observed a higher range of cobalt in the root samples (1.07–1.26mg/kg) of the plants irrigated with sewage water. As mentioned in this study, it was concluded that heavy metal accumulation was higher as a result of irrigation with sewage water. In addition to the irrigation factor, it has been reported that there were different factors such as plant genotype, the structure of plant root system, soil and climatic conditions and the response of plant species to elements related to the seasonal cycles can affect the bioavailability of elements to plants (Khan et al., 2020).

Metal accumulation in leaf samples

Significant (P< 0.05) effect of treatments on cobalt contentswas noticed for *Z. mays, P. typhoideum, C. tetragonoloba, S. rostrata, S. bicolor, S. vulgare, B. campestris, B. napus, M. sativa, T. alexandrinum, E. colona, B. juncea* while the non-significant effect was seen in *T. resupinatum*. The cobalt content in leaves of forages grown with supply of water from tap was in sequence of *T. resupinatum*> *B. campestris*> *P. typhoideum*> *C. tetragonoloba*> *E. colona*> *M. sativa*> *Z. mays*> *B. napus*> *S. vulgare*> *S. bicolor*> *S. rostrata*> *T. alexandrinum*> *B. juncea*. The cobalt content in leaves of forages grown with the supply of water from sewage source was in sequence of *T. resupinatum*> *P. typhoideum*> *B. napus*> *M. sativa*> *Z. mays*> *T. resupinatum*> *B. juncea*> *T. alexandrinum*> *B. campestris*> *S. vulgare*> *C. tetragonoloba*> *E. colona*> *S. rostrata*. The highest average concentration of cobalt in the leaves of the plants was seen in *T. resupinatum* (1.155mg/kg), and the lowest was seen in *B. juncea* (0.043mg/kg) in the winter forage (Table 4). Permissible maximum limit of the cobalt accumulation in plants was reported as 50mg/kg by WHO (1996), FAO/WHO (2001), Standard Guidelines in Europe (Chiroma et al., 2014). Cobalt values in the present study were lower than this limit in plant samples. Khan et al. (2009) found higher cobalt concentration (0.023–0.030mg/kg) in forages samples. Ahmad et al. (2014) examined the heavy metal accumulation in the leaves of *Brassica napa* samples irrigated with wastewater and tap water in their study in Khushab, Pakistan, and found that the cobalt accumulation in the leaves samples irrigated with sewage water (0.77mg/kg) was more than irrigated with tap water (0.49mg/kg). Alloway & Ayres (1997) analysed heavy metal uptake by forages and explained that metal accumulation depends upon their age, edaphic factors and the climatic factors.

TABLE 3. The concentrations of cobalt in root samples (mg/kg).

Roots	Irrigation water		Mean square
	Tap	Sewage	
	Summer		
Z. mays	0.114±0.0017	0.162±0.0035	0.006**
P. typhoideum	0.118±0.0016	0.219±0.0017	0.025*
C. tetragonoloba	0.117±0.0018	0.217±0.0018	0.025*
S. rostrata	0.185±0.0016	0.212±0.0015	0.002**
E. colona	0.098±0.0017	0.150±0.0150	0.007*
S. bicolor	0.085±0.0016	0.103±0.0018	0.001**
S. vulgare	0.136±0.0023	0.142±0.0021	0.001**
	Winter		
B. campestris	0.135±0.0017	0.185±0.0026	0.006**
B. napus	0.075±0.016	0.235±0.0028	0.064*
T. resupinatum	0.217±0.0018	0.220±0.0192	0.001**
B. juncea	0.092±0.0015	0.175±0.0176	0.017*
M. sativa	0.090±0.0017	0.204±0.0023	0.032*
T. alexandrinum	1.375±0.019	1.560±0.0231	0.086*
df	1	Error	9
Permissible limit[a]		0.1 mg/kg	

*, **: Significant at 0.05 and 0.01 levels, ns: Non–significant, Source: [a]McDowell (1997).

TABLE 4. The concentration of cobalt in leaf samples (mg/kg) of different forages.

Leaves	Irrigation water		Mean square
	Tap	Sewage	
	Summer		
Z. mays	0.139±0.0012	0.158±0.0016	0.001**
P. typhoideum	0.135±0.0017	0.195±0.0013	0.009**
C. tetragonoloba	0.093±0.0018	0.162 ±0.0018	0.012*
S. rostrata	0.105±0.0018	0.097 ±0.0016	0.001**
E. colona	0.058± 0.0058	0.140 ± 0.0015	0.017*
S. bicolor	0.113±0.0017	0.126 ±0.0066	0.001**
S. vulgare	0.118 ±0.0016	0.136±0.0017	0.001**
	Winter		
B. campestris	0.137±0.0021	0.166±0.0034	0.002**
B. napus	0.127 ±0.0017	0.183±0.0017	0.008*
T. resupinatum	0.780±0.0215	1.155±0.0215	0.352[ns]
B. juncea	0.043±0.0017	0.148±0.0017	0.028*
M. sativa	0.140±0.0099	0.183±0.0996	0.005**
T. alexandrnium	0.087±0.0021	0.141±0.0012	0.007**
df	1	Error	9
Permissible limit[a]		0.1 mg/kg	

*, **: Significant at 0.05 and 0.01 levels, ns: Non–significant, Source: [a]McDowell (1997)

Bioconcentration factor

Bioconcentration factor for cobalt content in of forages grown with supply of water from tap was in sequence of: *M. sativa*> *S. bicolor*> *S. vulgare*> *T. alexandrinum*> *S. rostrata*> *C. tetragonoloba*> *B. juncea*> *E. colona*> *B. campestris*> *P. typhoideum*> *T. resupinatum*> *B. napus*> *Z. mays*. The order of cobalt contents of forages grown with the supply of water from the tap was in sequence of *T. resupinatum*> *E. colona*> *S. vulgare*> *T. alexandrinum*> *S. bicolor*> *C. tetragonoloba*> *M. sativa*> *B. napus*> *S. rostrata*> *B. juncea*> *B. campestris*> *P. typhoideum*> *Z. mays*. The highest value was seen for cobalt BCF in *T. resupinatum* (7.7), and the lowest value wasseen in *Z. mays* (0.068) (Table 5). The mean values for BCF reported by Ahmad et al. (2014) were (0.036, 0.038) in *Brassica rapa* grown at tap water and sewage water irrigated sites, respectively. Although it was close to each other, it was observed that BCF value in sewage water irrigation area is higher like as in the present study. Some characteristics of the soil like its pH could impact the mobility of soil metals. For instance, the mobility of metals from soil to plants was limited by the high value of pH, unlike in case of low pH (Siddique et al., 2019). It is possible that the pH of analysed soil affects the bioconcentration factor of metal (Ugulu et al., 2019 b).

TABLE 5. Bioconcentration factor for forage/soil.

Forage	BCF	
	Irrigation Water	
	Tap	Sewage
Summer		
Z. mays	0.068	0.076
P. typhoideum	0.076	0.106
C. tetragonoloba	0.609	1.464
S. rostrata	0.668	1.214
E. colona	0.162	2.435
S. bicolor	1.120	1.519
S. vulgare	0.833	2.305
Winter		
B. campestris	0.079	0.106
B. napus	0.068	0.103
T. resupinatum	0.096	7.778
B. juncea	0.412	0.438
M. sativa	1.155	1.233
T. alexandrinum	0.827	1.626

Pollution load index

The series of PLI value was *Z. mays*> *B. napus*> *P. typhoideum*> *B. campestris*> *M. sativa*> *B. juncea*> *C. tetragonoloba*> *E. colona*> *S. rostrata*> *S. bicolor*> *T. alexandrinum*> *S. vulgare* in the forage samples when irrigated with freshwater. The series of PLI value according to the forage samples irrigated with sewagewater was: *T. resupinatum*> *P. typhoideum*> *Z. mays*> *B. campestris*> *B. napus*> *B. juncea*> *E. colona*> *T. alexandrinum*> *S. vulgare*> *M. sativa*> *C. tetragonoloba*> *S. rostrata*> *S. bicolor*. The maximum PLI value was seen in *T. resupinatum* (0.8910), and the lowest in *S. vulgare* (0.0056) (Table 6). It was observed obviously that the PLI values of the sewagewater irrigated samples were higher than the tap water irrigated samples. However, the measured values were lower than the values found by Khan et al. (2015). Higher PLI value (1.493) was noticed by Ahmad et al. (2014) when sewage source was used to irrigate the soil instead of tap water.

TABLE 6. Pollution load indexfor cobalt in soil samples.

Forage	PLI	
	Irrigation water	
	Tap	Sewage
Summer		
Z. mays	0.227	0.228
P. typhoideum	0.102	0.294
C. tetragonoloba	0.012	0.017
S. rostrata	0.0095	0.016
E. colona	0.0099	0.039
S. bicolor	0.0081	0.012
S. vulgare	0.0056	0.018
Winter		
B. campestris	0.142	0.228
B. napus	0.195	0.204
T. resupinatum	0.016	0.891
B. juncea	0.011	0.039
M. sativa	0.0124	0.017
T. alexandrinum	0.0058	0.019

Daily intake of metal and health risk index

The series of DIM value was: *T. resupinatum*> *M. sativa*> *Z. mays*> *B. campestris*> *P. typhoideum*> *B. napus*> *S. vulgare*> *S. bicolor*> *S. rostrata*> *C. tetragonoloba*> *T. alexandrinum*> *E. colona*> *B. juncea* when tap water was used to irrigate the soil. The series of DIM value was: *T. resupinatum*> *M. sativa*> *P. typhoideum*> *B. napus*> *B. campestris*> *C. tetragonoloba*> *Z. mays*> *B. juncea*> *T. alexandrinum*> *E. colona*> *S. vulgare*> *S. bicolor*> *S. rostrata* when water from sewage source was used to irrigate the soil. The highest DIM value was seen in *T. resupinatum* (0.026), and the lowest value was seen in *B. juncea* (0.00096). In the current results, the DIM values were lower than 1, and it suggests no risk of health (Ugulu et al., 2019c). Health risk index for cobalt was calculated in forages showed maximum value in *T. resupinatum* (0.6104) and minimum value in *E. colona* (0.0303) (Table 7). Khan et al. (2015) found lower HRI value (1.33–1.73) in wastewater irrigated sites. Although PLI and HRI values did not indicate a health risk, it is noteworthy that sewage water irrigation values were higher in both index values. According to Sajjad et al. (2009), the HRI depends on the chemical composition and the physical characteristics of soil, type of forage being consumed and the rate of the consumption of forages.

TABLE 7. Daily intake of metal and health risk index of cobaltin plant samples.

Forage	DIM		HRI	
	Irrigation water			
	Tap	Sewage	Tap	Sewage
Summer				
Z. mays	0.0033	0.0035	0.074	0.083
P. typhoideum	0.0030	0.0044	0.071	0.103
C. tetragonoloba	0.0021	0.0036	0.049	0.086
S. rostrata	0.0022	0.0023	0.052	0.055
E. colona	0.0013	0.0031	0.030	0.074
S. bicolor	0.0025	0.0028	0.059	0.067
S. vulgare	0.0026	0.0030	0.063	0.071
Winter				
B. campestris	0.0031	0.0037	0.072	0.087
B. napus	0.0028	0.0041	0.067	0.086
T. resupinatum	0.0177	0.026	0.412	0.610
B. juncea	0.0009	0.0033	0.022	0.078
M. sativa	0.0034	0.0046	0.074	0.096
T. alexandrinum	0.0019	0.0032	0.046	0.075

Conclusion

Although sometimes easily reachable on arable lands, irrigation with sewage water can pollute the soil basically. In the current study, plant and soil irrigated with sewage water accumulate dsignificantly higher amounts of cobalt than those irrigated with tap water. The cobalt contents were transferred from soil to forage but without following any particular pattern and changed with treatment. However, the increase in cobalt contents did not exceed the permissible limit. Therefore, with respect of cobalt, it may be safe for livestock to consume these forages.

Conflicts of interest: No conflicts of interest among authors for the publication of this paper.

Authors contribution: HS and HB was responsible for conducting the experiments and the data analysis. IU, MN and MM were responsible for analyzing and interpreting the data. IU and YD were responsible for writing the manuscript. KA and ZIK supervised the study. All authors read and approved the final manuscript.

Ethical approval: Not applicable.

References

Ahmad, K., Khan, Z.I., Yasmin, S., Ashraf, M., Ishfaq, A. (2014) Accumulation of metals and metalloids in turnip (*Brassica rapa* L.) irrigated with domestic wastewater in the peri-urban areas of Khushab City, Pakistan. *Pakistan Journal of Botany,* **46**(2), 511-514.

Ahmad, K., Ashfaq, A., Khan, Z.I., Bashir, H., Sohail, M., Mehmood, N., Dogan, Y. (2018a) Metal accumulation in *Raphanus sativus* and *Brassica rapa*: An assessment of potential health risk for inhabitants in Punjab, Pakistan. *Environmental Science and Pollution,* **25**(8), 16676-16685.

Ahmad, K., Nawaz, K., Khan, Z.I., Nadeem, M., Wajid, K. et al. (2018b) Effect of diverse regimes of irrigation on metals accumulation in wheat crop: An assessment-dire need of the day. *Fresenius Environmental Bulletin,* **27**(2), 846-855.

Ahmad, K., Wajid, K., Khan, Z.I., Ugulu, I., Memoona, H., Sana, M., Nawaz, K., Malik, I.S., Bashir, H., Sher, M. (2019) Evaluation of potential toxic metals accumulation in wheat irrigated with wastewater. *Bulletin of Environmental Contamination and Toxicology*, **102**, 822-828.

Alloway, B.J., Ayers, D.C. (1997) "*Chemical Principles of Environmental Pollution*". 2nd ed. London, New York: Blackie Academic & Professional.

Baslar, S., Kula, I., Dogan, Y., Yildiz, D., Ay, G. (2009) A study of trace element contents in plants growing at Honaz Dagi-Denizli, Turkey. *Ekoloji*, **18**(72), 1-7.

Chary, N.S., Kamala, C.T., Raj, D.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.

Chiroma, T.M., Ebewele, R.O., Hymore, F.K. (2014) Comparative assessment of heavy metal levels in soil, vegetables and urban grey wastewater used for irrigation in Yola and Kano. *International Refereed Journal of Engineering Science*, **3**, 1-9.

Cui, Y.J., Zhu, Y.G., Zhai, R.H., Chen, D.Y., Huang, Y.Z., Qui, Y., Liang, J.Z. (2004) Transfer of metals from near a smelter in Nanning, China. *Environment*

International, **30**, 785-791.

Dogan, Y., Ugulu, I., Baslar, S. (2010) Turkish red pine as a biomonitor: A comparative study of the accumulation of trace elements in needles and barks. *Ekoloji,* **19**(75), 88-96.

Dogan, Y., Baslar, S., Ugulu, I. (2014a) A study on detecting heavy metal accumulation through biomonitoring: Content of trace elements in plants at Mount Kazdagi in Turkey. *Applied Ecology and Environmental Research*, **12**(3), 627-636.

Dogan, Y., Unver, M.C., Ugulu, I., Calis, M., Durkan, N. (2014b) Heavy metal accumulation in the bark and leaves of *Juglans regia* planted in Artvin City, Turkey. *Biotechnology & Biotechnological Equipment,* **28**(4), 643-649.

Durkan, N., Ugulu, I., Unver, M.C., Dogan, Y., Baslar, S. (2011) Concentrations of trace elements aluminum,boron, cobalt and tin in various wild ediblemushroom species from Buyuk Menderes River Basin of Turkey by ICP–OES. *Trace Elements and Electrolytes,* **28**(4), 242-248.

FAO/WHO. (2001) Maximum levels for lead. Joint FAO/WHO food standards programme, Codex Alimentarius Commission, 24th session, Geneva, 2–7 July 2001, Codex Alimentarius Commission. Food and Agriculture Organization of the United Nations, Rome.

Fleming, G.A. (1970) The influence of stage of maturity and season on trace element levels in perennial ryegrass. *Agricultural Digest,* **19**, 25-32.

Khan, Z.I., Ashraf, M., Ahmad, N., Ahmad, K., Valeem, E.E. (2009) Availability of nutritional minerals (cobalt, copper, iron, manganese and zinc) in pastures of central Punjab for farm livestock. *Pakistan Journal of Botany*, **41**(4), 1603-1609.

Khan, Z.I, Ahmad, K., Ashraf, M., Parveen, R., Mustafa, I., Khan, A., Bibi, Z., Akram, N.A. (2015) Bioaccumulation of heavy metals and metalloids in luffa (*Luffa cylindrica* L.) irrigated with domestic wastewater in Jhang, Pakistan: A prospect for human nutrition. *Pakistan Journal of Botany*, **47**(1), 217-224.

Khan, Z.I., Ugulu, I, Sahira, S, Ahmad, K., Ashfaq, A., Mehmood, N., Dogan, Y. (2018a) Determination of toxic metals in fruits of *Abelmoschus esculentus* grown in contaminated soils with different irrigation sources by spectroscopic method.*International Journal of Environmental Research,* **12**(3), 503-511.

Khan, Z.I., Ugulu, I., Umar, S., Ahmad, K., Mehmood, N., Ashfaq, A., Bashir, H., Sohail, M. (2018b) Potential toxic metal accumulation in soil, forage and blood plasma of buffaloes sampled from Jhang, Pakistan. *Bulletin of Environmental Contamination and Toxicology*, **101**, 235-242.

Khan, Z.I., Ugulu, I., Ahmad, K., Yasmeen, S., Noorka, I.R., Mehmood, N., Sher, M. (2018c) Assessment of trace metal and metalloid accumulation and human health risk from vegetables consumption through spinach and coriander specimens irrigated with wastewater. *Bulletin of Environmental Contamination and Toxicology*, **101**, 787-795.

Khan, Z.I., Ahmad, K., Safdar, H., Ugulu, I., Wajid, K., Bashir, H., Dogan, Y. (2018d) Manganese bioaccumulation and translocation of in forages grown in soil irrigated with city effluent: An evaluation on health risk. *Research Journal of Pharmaceutical, Biological and Chemical Sciences*, **9**(5), 759-770.

Khan, Z.I., Ahmad, K., Rehman, S., Ashfaq, A., Mehmood, N., Ugulu, I., Dogan, Y. (2019a). Effect of sewage water on accumulation of metals in soil and wheat in Punjab, Pakistan. *Pakistan Journal of Analytical & Environmental Chemistry,* **20**(1), 60–66.

Khan, Z.I., Nisar, A., Ugulu, I., Ahmad, K., Wajid, K., Bashir. H., Dogan, Y. (2019b). Determination of cadmium concentrations of vegetables grown in soil irrigated with wastewater: evaluation of health risk to the public. *Egyptian Journal of Botany,***59**(3), 753–762.

Khan, Z.I., Iqbal, S., Ahmad, K., Ashfaq, A., Bashir, H., Dogan, Y. (2019c). Assessment of heavy metal content of wheat irrigated with wastewater in Sargodha, Pakistan: Implications for human health. *Trace Elements and Electrolytes,* **36**(2), 82–92.

Khan, Z.I., Arshad, N., Ahmad, K., Nadeem, M., Ashfaq, A., Wajid, K., Bashir, H., Munir, M., Huma, B., Memoona, H., Sana, M., Nawaz, K., Sher, M., Abbas T., Ugulu. I. (2019d) Toxicological potential of cobalt in forage for ruminants grown in polluted soil: a health risk assessment from trace metal pollution for livestock. *Environmental Science and Pollution Research*, **26**, 15381-15389.

Khan, Z.I., Safdar, H., Ahmad, K., Wajid, K., Bashir, H., Ugulu, I., Dogan, Y. (2020) Copper bioaccumulation and translocation in forages grown in soil irrigated with sewage water. *Pakistan Journal of Botany*, **52**(1), 111-119.

Khaskhoussy, K., Hachicha, M., Kahlaoui, B., Messoudi-Nefzi, B., Rejeb, A., Jouzdan, O., Arselan, A. (2013) Effect of treated wastewater on soil and corn crop in the Tunisian area. *Journal of Applied Sciences Research*, **9**(1), 132-140.

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

McDowell, L.R. (1997) "*Minerals for Grazing Ruminants in Tropical Regions*", 3rd ed. University of Florida, Ganesville FL.

Nadeem, M., Qureshi, T.M., Ugulu, I., Riaz, M.N., An, Q.U., et al. (2019) Mineral, vitamin and phenolic contents and sugar profiles of some prominent date palm (*Phoenix dactylifera*) varieties of Pakistan. *Pakistan Journal of Botany*, **51**(1), 171-178.

Sajjad, K., Farooq, R., Shahbaz, S., Khan, M.A., Sadique, M. (2009) Health risk assessment of heavy metals for population via consumption of vegetables. *World Applied Sciences Journal,* **6**, 1602-1606.

Siddique, S., Ahmad, K., Khan, Z.I., Wajid, K., Bashir, H. et al. (2019) Sodium status of soil, forages, and small ruminants of Punjab, Pakistan. *Pure and Applied Biology*, **8**(3), 1950-1961.

Tung, G., Temple, P.J. (1996) Uptake and localization of lead in corn (*Zea mays* L.) seedlings: A study by histochemical and electron microscopy. *Science ofthe Total Environment*, **188**, 71-85.

Tunisian Standards (1989) Environment protection-use of reclaimed water for agricultural purposes-physical, chemical and biological specifications (in French), Tunisian standards, ground water discharge of waste water. INNORPI, NT 106.03.

Ugulu, I. (2015a) Determination of heavy metal accumulation in plant samples by spectrometric techniques in Turkey. *Applied Spectroscopy Reviews,* **50**(2), 113-151.

Ugulu, I. (2015b) A quantitative investigation on recycling attitudes of gifted/talented students. *Biotechnology & Biotechnological Equipment,* **29**, 20-26.

Ugulu, I., Baslar, S., Dogan, Y., Aydin H. (2009) The determination of colour intensity of *Rubia tinctorum* and *Chrozophora tinctoria* distributed in Western Anatolia. *Biotechnology & Biotechnological Equipment,* **23** (SE), 410-413.

Ugulu, I., Dogan, Y., Baslar, S., Varol, O. (2012) Biomonitoring of trace element accumulation in plants growing at Murat Mountain. *International Journal of Environmental Science and Technology*, **9**, 527-534.

Ugulu, I., Unver, M.C., Dogan, Y. (2016) Determination and comparison of heavy metal accumulation level of *Ficus carica* bark and leaf samples in Artvin, Turkey. *Oxidation Communications*, **39**(1), 765-775.

Ugulu, I., Unver, M.C., Dogan, Y. (2019a) Potentially toxic metal accumulation and human health risk from consuming wild *Urtica urens* sold on the open markets of Izmir. *Euro-Mediterranean Journal for Environmental Integration,* **4**, 36.

Ugulu, I., Khan, Z.I., Rehman, S., Ahmad, K., Munir, M., Bashir, H., Nawaz, K. (2019b) Appraisal of trace element accumulation and human health risk from consuming field mustard (*Brassica campestris* Linn.) grown on soil irrigated with wastewater. *Pakistan Journal of Analytical & Environmental Chemistry*, **20**(2), 107-114.

Ugulu, I., Khan, Z.I., Rehman, S., Ahmad, K., Munir, M., Bashir, H., Nawaz, K. (2019c) Trace metal accumulation in *Trigonella foenum-graecum* irrigated with wastewater and human health risk of metal access through the consumption. *Bulletin of Environmental Contamination and Toxicology,* **103**, 468-475.

Unver, M.C., Ugulu, I., Durkan, N., Baslar, S., Dogan, Y. (2015) Heavy metal contents of *Malva sylvestris* sold as edible greens in the local markets of Izmir. *Ekoloji,* **24**(96), 13-25.

USEPA. (1997) Exposure Factors Handbook. Volume II-Food Ingestion Factors. EPA/600//P-95/002Fa. Office of Research and Development, US Environmental Protection Agency, Washington, DC.

USEPA. (2010) Risk-based concentration table. Office of Research and Development, Washington, DC.: US Environmental Protection Agency.

WHO. (1996) Trace elements in human nutrition and health. Prepared in collaboration with the FAO of the UN and the IAEA. World Health Organization, Geneva.