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# Estimation of Chromium in Soil-Plant-Animal Continuum: A Case Study in Ruminants of Punjab, Pakistan

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#### ESTIMATION OF CHROMIUM IN SOIL-PLANT-ANIMAL CONTINUUM: A CASE STUDY IN RUMINANTS OF PUNJAB, PAKISTAN

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#### ABSTRACT

The increased use of waste water for agriculture purposes has increased around the globe. There are toxic metals present in waste water which affect plants, animals and human health. But at the same time it also contains useful nutrients which increase growth of plants. Different indices were applied to evaluate the metals present in water soil plant and milk. Various and correlation were determined with the help of SPSS, mean significance was found at the probability levels of 0.05, 0.001 and 0.01. The highest value of Cr was found in Avena sativa (0.7872 mg/kg) collected from site 5 while lower concentration of Cr was observed in Brassica campestris (0.0743 mg/kg) at site 4. In soil samples, Trifolium alexandrinum showed highest value of Cr (0.9887 mg/kg) at site 1 while lowered concentration was observed in Zea mays (0.1862 mg/kg) at site 3. Milk samples of site 5 had higher value of Cr (0.2898 mg/kg) and lowest at (0.1540 mg/kg) site 2. Water samples of site 3 had high value of Cr in them (1.849 mg/kg) and lowered concentration was found in water samples of site 5 (0.219 mg/kg). Cr concentration in fodders ranges from 0.0743 to 0.7872 mg/kg, soil 0.1862 to 0.9887 mg/kg, milk 0.1540 to 0.2898 mg/kg and water 0.219 to 1.849 mg/kg. In water samples, Cr level were above than permissible limit. In milk samples, concentration of Cr was greater than permissible limit which shows that it is hazardous to human health, may be due to pollution of environment like air, water and soil. This study concluded that if animals are allowed to graze on contaminated fodders and drink wastewater then metals were accumulated in their tissues and milk which causes toxicity to human health.

Keywords: Waste water, chromium, soil, plant, human health.

#### **INTRODUCTION**

The excess usage of waste water agriculture field is the serious issue of today worldwide. The untreated sewage effluents are released into drainage channels which then reached to the soil and pollute it. Industrial and urban activities are also the reason of heavy metal accumulation (Mehdi et al., 2003). Trace elements continuously enters the soil and accumulate so, it becomes toxic for the plants growing in it. Through these plantations they enter the food chain and hence very serious issues arose (Shukry, 2001). Most fodder plants minerals and heavy metals from soil and also from air due to dust and vehicular emission .Use of fertilizers also add metals in soil .Most of these metals exceed their limit and causes harmful effects on health of livestock (Tokalioglu et al., 2000). Crops grown on soil which is normally irrigated by sewage water contain lot of metals which are harmful for animals and human (Ward and Savage, 1994). Animals grazing on contaminated fodder accumulate heavy metals which then move towards body muscles and also in milk. Human health is on risk when they consume contaminated meat from those animals who consumes contaminated food (Chitmanat and Traichaiyaporn, 2010). Milk is an important part of diet contains all the essential nutrients around the world (Buldini et al., 2002). Industrial revolution technical progress and increasing traffic results in increasing pollution (Licata et al., 2004). Heavy metal concentration is increasing in environment speedly (Staniskiene and Garaleviciene, 2004). Metals found in soil like lead and cadmium enter into plants through food chain and increase the risk of health for the life of organisms (Licata et al., 2004). Metal are soluble in water and plants have natural tendency of accumulating them. Heavy metals are found everywhere in environment .Industries release lot of waste water which contain heavy metal plants uptake these metals and store them these metals cause different diseases like problems in neurotransmitter production effect on metabolic processes, neurological functions disturbance (Singh et al., 2004; Chen et al., 2005). Different indices were applied to evaluate the metals present in water, soil, plant and milk samples. The objective of the study was to determine concentration of heavy metals in soil, fodders, water and milk.

## MATERIALS AND METHODS:

## Study area

This study was done to evaluate the effects of usage of sewage water in soil and selected crops from Mandi-Bahauddin district. The climate of this region is moderate. Six samples of fodders were collected from five sites. These are: *Trifolium alexendrinium, Brassica* 

campestris, Pennisetum glaucum, Zea mays, Sorghum bicolor and Avena sativa.

#### Water Samples

5 water samples were taken in bottles from waste water sites. These samples of water were those which were used to irrigate the fodders samples. The bottles were washed earlier to avoid contamination.

## Soil Samples

90 samples of soil were collected from where fodders samples were collected. Soil samples were taken from each site at a depth of 15 cm. After that, it was placed in polythene bags and brought to laboratory for further analysis.

#### Milk Samples

Fresh milk samples of about 5 bottles were collected from selected locations. Milking was done at morning. Milk was collected in polythene bottles that were already washed with de-ionized water to avoid contamination. 100 ml milk was collected from cows and stored at -20 °C until analysis was done.

## **Preparation of Water Samples**

Water samples were digested with a mixture of 10ml of HNO<sub>3</sub> and 5ml  $H_2O_2$ in a digestion tube. After that, they were allowed to cool down, collected in a volumetric flask with a final volume of 50ml and passed to atomic absorption spectrophotometer to measure chromium concentration (Yu et al., 2006).

## **Preparation of Soil Samples**

Firstly the soil samples were placed in air for drying and then put in oven at 70°C. After that, add soil sample (0.5 g) and HNO<sub>3</sub> (20 ml) in a digestion tube. Heat the mixture for 30 min. Then add 10 ml  $H_2O_2$  in it and heated again until mixture became a colorless solution and passed to atomic absorption spectrophotometer to measure chromium concentration (Dey et al., 1999).

## **Preparation of Fodder Samples**

Firstly the fodder samples were placed in air for drying and grinded into fine powder. After that samples were put into oven (70°C) for 72 hours. Fodder sample (1 g) was added with 68 % of HNO<sub>3</sub> (5 ml) in a digestion chamber and left this mixture for a night. Samples were put on heat and cool down. Then, added 30 % H<sub>2</sub>O<sub>2</sub> (5 ml) and heated until became a clear solution. Allowed it to cool down, solution was made to about 50 ml and passed to atomic absorption spectrophotometer to measure chromium concentration (Dey et al., 1999).

## **Preparation of Milk Samples**

Milk sample (1ml), 10ml of  $HNO_3$ and 3 ml of  $H_2O_2$  was taken in the digestion tube. Heat the mixture of digestion tubes until became colorless. Filtered the solution and diluted with 50 ml of distilled water. Then, solution was passed to atomic absorption spectrophotometer to measure chromium concentration (Qin et al., 2009).

## Statistical Analysis

Variance and correlations were determined with the help of SPSS (special program for social sciences) and software version no 20 was used. By using one way ANOVA variance of fodder, soil, water and milk was calculated. Mean significance was at 0.05, 0.001 and 0.01 probability levels represented by (Steel and Torrie, 1980).

## **Bio-concentration Factor**

Amount of metals present in fodders which is transferred through soil is

measured by this formula (Cui et al., 2004):

Bio-concentration = Concentration of metals in fodders / Concentration of metals in soil

## Daily intake of Metals (DIM)

Daily intake of metal was measured by Chary et al. (2008) formula:

 $C_{metal}$  presents chromium concentration in forage,  $C_{daily intake of food}$  presents fodder intake in animals in kg/day, Conversion factor was 0.085 mg/kg and B<sub>average weight</sub> showed cattle weight selected at the study area. Average fodder consumption was 12.5 kg and cattle weight was 550 kg (Briggs and Briggs, 1980).

## Health Risk Index (HRI)

HRI index was applied to check the health risk in animals as a result of consuming polluted fodder (USEPA, 2002)

HRI= Daily intake of metal/Oral reference dose

## **Pollution Load Index (PLI)**

Metals level present in soil is checked by PLI (Liu et al., 2005)

PLI= Metals concentration in soil/ Metal reference value in soil.

## Enrichment Factor (EF)

Enrichment factor deals with metal holding power of soil (Hwaiti and Khashman, 2015).

EF = Metals present in the contaminated soil/ Metals present in the less contaminated soil

#### RESULTS

#### Water

In water data, Cr metal showed non-significant effect in sites (Table 1). According to the table, high concentration of Cr was found in site 3 of water samples and least concentration was found at site 5 (Table 2).

 Table 1: Analysis of Variance for Cr in sewage

 water irrigated sites.

SOV	DF	Mean
		Square
Sites	4	1.236 <sup>ns</sup>
Error	10	0.980

ns= non-significant

Table 2: Mean Cr concentration in sewagewater irrigated sites.

Sites	Mean square ± Standard Error
1	$0.879 \pm 0.157$
2	0.703±0.092
3	1.849±1.261
4	0.357±0.089
5	0.219±0.033

#### Soil

In soil data, Cr showed significant effect in Sites, Fodders and Sites \* Fodders (Table 3). Table showed that highest Cr value was detected in *T. alexandrinum* at site 1 and lesser value was observed in *Z. mays* at site 3 (Table 4)

## Fodders

In fodder data, Cr significant effects was shown by sites, fodders and

Sites \* Fodders (Table 3). High Cr concentration was observed at site 5 in *A. sativa* and lowered concentration was found at site 4 in *B. campestris* (Table 4).

## Milk

In milk data, significant effect of Cr was observed on sites (Table 5). A higher Cr level was found in milk samples of site 5 (Table 6).

#### **Pollution Load Index**

Highest PLI for Cr was observed in *T. alexandrinum* at site 1 and least amount was observed in *Z. mays* at site 3 (Table 7).

#### **Bio Concentration Factor**

Chromium higher values for BCF were obtained in *Z. mays* of site 3 and lowered quantity was observed in *B. campestris* of site 4 (Table 7).

#### **Enrichment Factor**

Enrichment factor of Cr in high quantity was present in *Z. mays* at site 3 and *B. campestris* showed the lowest EF value at site 4 (Table 7).

## Daily Intake and Health Risk Index

Daily intake of metal for Cr was present higher in Z. mays of site 4 and P. glaucum of site 2 showed the lowest Cr concentration. Health risk index of Cr gave higher concentration in A. sativa of site 5 and B. campestris presented the lowest concentration at site 1 (Table 8). Ahmad et al. (2022). Evaluation of Chromium in soil, Plant and Animal in Punjab. *J Biores Manag.*, 9(1): 81-89.

SOV	Soil samples		Fodder samples	
	DF Mean Square		DF	Mean Square
Sites	4	0.347***	4	0.063***
Fodders	5	0.080*	5	0.037***
Sites * Fodders	20	0.123***	20	0.047***
Error	60	0.026	60	0.012

Table 3: Analysis of variance	for Cr in soil and	fodder samples of s	ewage water irrigated sites.

\*\*\*, \*= significant at 0.001 & 0.05 level

#### Table 4: Cr in soil and fodder samples collected from five selected sites of sewage water.

Sites	Т.	<i>A</i> .	Ζ.	<i>B</i> .	<i>P</i> .	S.bicolor			
	alexandrinum	sativa	mays	campestris	glaucum				
Soil	Soil								
Site1	0.9887	0.2915	0.3137	0.3287	0.4310	0.7663			
Site2	0.4202	0.3637	0.3270	0.3202	0.4130	0.3812			
Site3	0.4375	0.3647	0.1862	0.2380	0.3813	0.2310			
Site4	0.4415	0.4290	0.6690	0.9020	0.7430	0.7663			
Site5	0.6690	0.9548	0.3670	0.3710	0.3910	0.3737			
Fodde	er								
Site1	0.4137	0.3482	0.3527	0.1627	0.2945	0.2547			
Site2	0.3740	0.3598	0.4092	0.4503	0.2587	0.3820			
Site3	0.2447	0.1818	0.3772	0.2480	0.4267	0.2927			
Site4	0.2412	0.3120	0.4555	0.0743	0.4068	0.3850			
Site5	0.4097	0.7872	0.4225	0.4330	0.2898	0.2659			

#### Table 5: Data analysis for Cr in Milk samples.

SOV	DF	Mean Square
Sites	4	0.010**
Error	10	0.001

\*\*= significant at 0.01

#### Table 6: Mean Cr concentration in Milk samples from five selected sites of sewage water.

Sites	Mean square ± Standard Error
1	0.1960±0.000
2	0.1540±0.029
3	0.2251±0.018
4	0.2766±0.018
5	0.2898±0.029

#### Table 7: PLI, BCF and EF of Cr in soil collected from five selected sites of sewage water.

Site	T. alexandrinum	A. sativa	Z. mays	B. campestris	P. glaucum	S.bicolor		
PLI								
1	0.00988	0.00291	0.00313	0.003287	0.00431	0.00766		
2	0.00420	0.00363	0.00327	0.003202	0.00413	0.00381		
3	0.00437	0.00364	0.00186	0.00238	0.00381	0.00231		
4	0.00441	0.00429	0.0066	0.00902	0.00743	0.00766		
5	0.0066	0.00954	0.00367	0.00371	0.00391	0.00373		
BCF	BCF							
1	0.4184	1.1945	1.1243	0.4949	0.6832	0.3323		
2	0.8900	0.9892	1.2513	1.4063	0.6263	1.0020		

Ahmad et al. (2022). Evaluation of Chromium in soil, Plant and Animal in Punjab. *J Biores Manag.*, 9(1): 81-89.

3	0.5593	0.4984	2.0257	1.0420	1.1190	1.2671
4	0.5463	0.7272	0.6808	0.0823	0.547	0.5024
5	0.6124	0.8244	1.1512	1.1671	0.7411	0.7115
EF						
1	32.1867	91.885	86.4863	38.0754	52.5611	25.5674
2	68.4655	76.0982	96.2597	108.1776	48.1840	77.0845
3	43.0241	38.3455	155.8291	80.1551	86.0820	97.4692
4	42.0245	55.9440	52.3743	6.3363	42.1161	38.6472

Table 8: Daily intake of Cr and health risk index of fodders collected from five selected sites of sewage water.

-							
Site	DIM&HRI	T.alexandrinum	A.sativa	Z.mays	B.campestris	P.glaucum	S.bicolor
1	DIM	0.000799	0.000673	0.000681	0.000314	0.000569	0.000492
	HRI	0.000533	0.000448	0.000454	0.00021	0.000379	0.000328
2	DIM	0.000722	0.000695	0.000790	0.00087	0.0005	0.000738
	HRI	0.000482	0.000463	0.000527	0.00058	0.000333	0.000492
3	DIM	0.000472	0.000351	0.000728	0.000479	0.000824	0.000565
	HRI	0.000315	0.000234	0.000486	0.000319	0.00055	0.000377
4	DIM	0.000465	0.000603	0.000879	0.000144	0.000786	0.000744
	HRI	0.000311	0.000402	0.000587	0.0000957	0.000524	0.000496
5	DIM	0.0007914	0.00152	0.000816	0.000836	0.00056	0.000514
	HRI	0.000528	0.001014	0.000544	0.000558	0.000373	0.000342

#### DISCUSSION

In current work the concentration of Cr was higher as compared to values found by Ahmad et al., (2018). Lower values of heavy metals Cr, in waste water was given by Alghobar et al. (2014) as compared to found in this work. The mean concentration of Cr as observed by Adekiya et al. (2018) was found greater as found in this study. The trend of Cr proposed by Awokunmi et al. (2015) was found lower. Pathak et al. (2010) studied concentration of chromium metals in soil were higher than present study. As proposed by Adekiya et al. (2018) values of heavy metals like Cr were lower as compared to values observed in present study. Bhatti et al., (2016) reported greater Cr mean concentration in fodder samples while in this study the Cr concentration of fodder plants was lower. The mean concentration of Cr was greater in this study as compared to reported by Bousbia et al. (2019). Yu et al. (2015) suggested concentration of Cr were lower than concentration of Cr in current study. The trend for PLI of Cr was observed high in

this work as detected (Cr 0.02 mg/kg) by Ashfaq et al. (2015). If PLI is found greater than 1 soil is more contaminated when Pollution load index detected lower than 1 indicate that the soil is less polluted and PLI= 1 showed only baseline level of pollutants present. BCF for Cr found in this work study was higher than suggested range of Asdeo (2014). Enrichment factor for Cr was in extremely high significant Alghobar enrichment category. and Suresha (2015) showed lower enrichment factor of Cr then present study. According to the values of DIM of Cr reported by Ali et al. (2017) were lowered than values found in present study. The level of daily intake of Cr was observed lower in this study then observed Guerra et al. (2011). The trend of heavy metal Cr was high in this study as evaluated by Ali et al,. (2017). When the values of HRI were found to be lower than 1 then there is no risk to health of livestock but when values exceeds the limit 1 then it is an indicator of risk to animal health (Sajjad et al., 2009).

## CONCLUSION

The reason of higher concentration of heavy metals in samples is the long term use of sewage water for irrigation of fodder crops. Fodders and soil buildup heavy metals when animals consume these fodders, heavy metals enter into the milk and may cause toxicity to human health.

## **CONFLICT OF INTEREST**

Authors declare no conflict of interest in the publication of this manuscript.

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