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Multivariate Analysis of Selected Metals in Agricultural Soil Receiving UASB Treated Tannery Effluent at Jajmau, Kanpur (India)

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Abstract Tannery effluent and soils samples were collected from 12 different sites of an agricultural area receiving treated tannery wastewater near Kanpur city (India). The samples were analyzed for heavy metals (Fe, Cr, Zn, Mn, Cu, Ni and Pb) content with a view to assess the impact of industrial wastewater on agricultural soils. The results revealed elevated levels of Fe and Cr in agricultural soils irrigated with treated tannery effluents. Cluster analysis of tannery effluent and soil datasets yielded two groups of the metals and demonstrated their relationship in each media. Principal component analysis performed on two datasets yielded two significant factors each for the effluents and soils, suggested tanneries as the probable sources of metals in the soils.

Keywords Heavy metals · Tannery effluents · Agricultural soils · Cluster analysis · Principal component analysis

Leather is a major industrial product made from the natural fabric (the skin) and its processing has evolved from a traditional artisanal practice to an industrial activity. Intensive leather production in small clusters has caused environmental concern. An environmental challenge from leather processing arises from the nature and quantity of the chemicals used along with the amount of wastes generated and discharged (Cooman et al. 2003). Thus, the effluent from tannery industry is considered as a serious environmental threat throughout the world. The use of tannery wastewater is a major problem due to the presence of pollutants particularly metals, however, it is being used for the irrigation of agricultural crops and vegetable. Long-term irrigation can induce changes in the quality of agricultural soil and trace element inputs which sustained over long period (Barman et al. 2000; Gothberg et al. 2002; Sinha et al. 2005, 2006). India is one of the largest producers of leather in the world and about 0.7 million tons of wet salted hides and skins are processed in about 3,000 tanneries. These tanneries discharge 30,000 million L of wastewater with high concentrations of chromium. Uttar Pradesh is an important state in India, holding sizeable population of livestock and is a prominent center for leather processing. There are about 310 tanneries along the banks of river Ganga and about 85,000 tones of cow and buffalo hides are processed annually, which is about 15% of the total quantity. The quantity of effluent generated is about 30 L $kg^{-1}$of hide/skin processed. Although, the tanning industry has been in existence for a long time, the problem of environmental pollution received serious consideration only since last few years due to implementation of restrict control measures laid down by various pollution regulatory agencies.

Thus, it is utmost important to study the distribution of metals in tannery effluent along with receiving soils. To analyze large number of data set, the multivariate technique such as, principal component analysis (PCA) and cluster analysis (CA) are some of the important statistical tool used to evaluate the metals in the two media in terms of their contribution and dependence. In the present

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scenario of industrial revolution in the developing countries, these studies provide useful data on distribution of the metals in the two media (Tariq et al. 2005).

The objective of the present study is based on the above considerations. The levels of selected metals (Fe, Cr, Mn, Zn, Cu, Ni and Pb) in up-flow anaerobic sludge blanket (UASB) treated tannery effluent and agricultural soil from adjoining area receiving tannery wastewater was determined by atomic absorption spectrophotometer (AAS).

These data are statistically evaluated using multiple correlations, along with CA and PCA. A multivariate method such as PCA is a data reduction procedure whose main goal is to provide an easy visualization of the relationship amongst the variables determined in large number of data set. Thus, this approach would provide the correlation patterns of the metals in effluent and soil, which help to evolve an effluent treatment strategy by recovering toxic metals from the effluent.

Materials and Methods

In India, Kanpur (UP) lies in Indo-Gangatic plains between the parallels of 26°28′ N and 80°24′ E. About 310 tanneries are located at Jajmau (Kanpur), which is one of the major centers for the processing of raw hides. The wastewater discharge from these industries and sewage wastewater is being treated in an UASB treatment plant. The treated wastewater is being used for the irrigation of crops and vegetables growing in an area of 2,100 acre. Due to long-term irrigation, the area is highly contaminated and is selected for the present study. This is located about 0.10–10 km distance from the UASB treatment plant and was divided into 12 different sites (S-1 to S-12) where intense cultivation of edible plants is being made.

In 2005, the UASB treated tannery effluent was collected in acid washed plastic containers and brought to the laboratory for physico-chemical analysis (APHA 1992). For tannery effluent digestion, 50 mL of sample was digested with 10 mL of 70% HNO_3 using Microwave Digestion System MDS 2000. Composite soil samples were vertically collected with the depth of 5–10 cm from contaminated site, which was dried and ground. After homogenization and sieving (2 mm mesh), a small portion (ca. 0.50 g) of soil samples were digested in 10 mL double distilled water (ddw) +5 mL HCl +4 mL HF +1 mL HNO_3 in Microwave Digestion System MDS 2000 in closed Teflon vessels. Six replicates were taken for each sample analysis. After digestion, the solutions were adjusted to 25 mL with 0.6% HNO_3 solution. The metal content of digested samples was determined with GBC Avanta Σ AAS. The Analytical Grade reagents were used. Certified aqueous standards of the elements (SIGMA) were used to prepare standard curve for AAS. All the standards, reagents solution and samples were stored in polyethylene containers previously cleaned with 4 M HNO_3 and rinsed with ddw.

Analytical data quality was ensured through repeated analysis (n = 6) of EPA quality control samples (Lot TMA 989) for metals (Cd, Cr, Cu, Pb) in water and the results were found to be within ±2.68% of certified values. The reference solution of multi-elements and single element was used for the calibration of equipment and validation of test methods provided by National Physical Laboratory (NPL), New Delhi, India and their certified and observed values. Method validation (accuracy and repeatability) was performed by analyzing the certified reference materials solution (BND 1101.02) of multi-elements (Zn, Fe, Cu) and single element reference solution, BND 102.03 (Pb); BND 402.02 (Cr) and BND 1001.02 (Ni) and the results were found to be within ± 1.7% of certified values (n = 10). For sludge samples, analytical data quality of metals was ensured through repeated analysis of sewage sludge samples of Resource Technology Corporation (EPA Certified Reference material) (Catalog No. CRM 029-050; Lot No. JC029a) and results were found to be within Prediction Intervals. The blanks were run in triplicate to check the precision of the method with each set of samples.

Principal component analysis was performed on experimental data standardized through *z*-scale transformation in order to avoid misclassification due to wide differences in data dimensionally (Singh et al. 2004, 2005). It was applied to the metals of different sites in order to examine the interactions between metals at different sites. PCA was performed on a varimax normalized original matrix composed of 72 lines and 7 columns (Fe, Cr, Zn, Mn, Cu, Ni, Pb). These columns can be considered as statistically dependent variables or factors. Hierarchical agglomerative CA was performed on the normalized data set by means of the Ward's method, using Euclidean distance as a measure of similarity. This method uses the analysis of variance approach to evaluate the distance between clusters, attempting to minimize the sum of squares of any two clusters that can be formed at each step. All the mathematical and statistical computations were made using Excel 97, STATISTICA 5.0 software.

Results and Discussion

Examinations of Table 1 showed that highest mean concentration (mg L^{-1}) was exhibited by Fe (5.37), while the mean levels of Zn (0.39), Mn (0.13) and Cu (0.17) remains comparatively very low. This reflects the use of Fe salts during the preparatory stages of the tanning process. Toxic metals (Cr, 2.80; Ni, 0.60; Pb, 0.13 mg L^{-1}) were present

Table 1 Descriptive statistics of metals in (mg L^{-1}) tannery effluent

	Metals						
	Fe	Cr	Zn	Mn	Cu	Ni	Pb
Mean	5.37	2.80	0.39	0.13	0.17	0.60	0.13
Minimum	3.93	2.0	0.23	0.10	0.11	0.02	0.10
Maximum	6.19	4.32	0.48	0.19	0.30	0.12	0.19
SE	0.05	0.06	0.00	0.00	0.00	0.00	0.00
SD	0.40	0.48	0.04	0.03	0.03	0.04	0.03
Median	5.44	2.65	0.39	0.12	0.18	0.08	0.12
Mode	5.51	3.21	0.38	0.12	0.18	0.02	0.10
Kurtosis	2.10	0.33	4.01	–0.77	4.66	–1.62	1.19
Skew-ness	–0.99	0.86	–0.42	0.61	0.87	–0.13	1.20

n = 72, *S*E standard error, *SD* standard deviation

Table 2 Correlation coefficient matrix for selected metals in tannery effluent

Metals	Metals						
	Fe	Cr	Zn	Mn	Cu	Ni	Pb
Fe	1.00						
Cr	0.16	1.00					
Zn	–0.11	0.10	1.00				
Mn	0.06	0.04	0.21	1.00			
Cu	0.11	0.00	0.19	0.04	1.00		
Ni	0.10	–0.01	0.05	–0.07	0.06	1.00	
Pb	**–0.34**	–0.28	0.23	0.03	0.18	0.05	1.00

n = 72, bold values are significant at $p < 0.001$

Table 3 Descriptive statistics of metals (mg kg^{-1} dw) in soil samples

	Metals						
	Fe	Cr	Zn	Mn	Cu	Ni	Pb
Mean	20,401.3	293.7	230.8	288.0	68.5	26.9	16.9
Minimum	11,002.0	37.9	48.2	198.1	28.6	6.3	9.2
Maximum	30,953.0	859.7	476.7	390.4	152.08	63.8	28.2
SE	494.2	29.2	14.0	5.5	3.9	1.7	0.5
SD	4,193.3	247.6	118.6	46.8	33.	14.8	4.2
Median	20,909.5	181.5	207.4	289.5	56.5	26.3	16.9
Mode	–	–	–	–	–	12.0	18.5
Kurtosis	–0.5	–0.8	–1.0	–0.7	–0.3	–0.8	0.1
Skew-ness	0.1	0.8	0.5	0.1	0.9	0.5	0.5

n = 72

Table 4 Correlation coefficient matrix for selected metals in soils

Metals	Metals						
	Fe	Cr	Zn	Mn	Cu	Ni	Pb
Fe	1.00						
Cr	–0.17	1.00					
Zn	0.20	0.26	1.00				
Mn	**0.33**	0.16	0.23	1.00			
Cu	0.28	**0.40**	**0.88**	**0.32**	1.00		
Ni	**0.42**	0.19	**0.78**	**0.52**	**0.74**	1.00	
Pb	0.23	–0.26	0.14	0.08	0.02	0.09	1.00

n = 72, bold values are significant at $p < 0.001$

in high concentrations. The data (Table 1) reflect a wide dispersion in the metal contents of the tannery effluent samples as indicated by high standard deviation (SD). Also, large skew-ness and kurtosis values evidenced that the trace metals in tannery effluent are not normally distributed.

Thus, this situation unveils a random distribution of the metals in the effluents, which is in agreement with the sequence of chemical operations conducted in different batches during the hide/skin processing in most of the tannery industries. The correlation matrix (Table 2) of metals in treated tannery effluent showed that no significant relation exist between metals except Fe, which is negatively ($p < 0.001$) correlated with Pb. The data (Table 3) showed highest mean concentration (mg kg^{-1} dw) of Fe (20401.3), however, the mean level of metals, Zn, Mn and Cu remain comparatively very low, i.e., 230.8, 288.5 and 68.5 mg kg^{-1} dw, respectively. Toxic metals (Cr, Ni and Pb) were found in significantly high concentrations (mg kg^{-1} dw), i.e., 293.7 (Cr), 26.9 (Ni) and 16.9 (Pb). The high value of standard deviation (Table 4) reflects wide dispersion in metal contents of soil samples. In soil, the trace metals are not normally distributed as evidenced from high value of skew-ness and kurtosis. Thus, the random distribution of the metals in the soil receiving tannery wastewater was due to the sequence of chemical operation conducted in different batches of processing. The analysis of correlation matrix (Table 4) showed that Fe in soil have shown positive ($p < 0.001$) correlation with Mn and Ni. Similarly, Cu have shown significant positive ($p < 0.001$) correlation with Cr, Zn and Mn. In case of Ni, it is positively ($p < 0.001$) correlated with Zn, Mn and Cu. The treated tannery effluent (e) versus soil (s) trace metals content relationship is also studied (Table 5). The correlation coefficients have shown non-significant correlation with metals in effluent versus metals in soil except Zn, which showed positive significant ($p < 0.001$) correlation with Mn and Ni. This is due to long term irrigation of soil with treated tannery effluent since last many decades.

The principal component loading varimax normalized rotation for both treated effluent and soil samples was also performed. The principal factor loading for treated effluent

Table 5 Correlation coefficient matrix of selected metals in effluent vs soil samples

Metals	Metals						
	Fe (e)	Cr (e)	Zn (e)	Mn (e)	Cu (e)	Ni (e)	Pb (e)
Fe (s)	–0.029	–0.032	0.257	–0.120	0.077	0.067	–0.069
Cr (s)	–0.258	–0.261	–0.148	0.130	**–0.328**	0.053	–0.023
Zn (s)	–0.138	–0.181	0.186	0.059	0.071	0.097	–0.021
Mn (s)	**–0.860**	0.034	**0.339**	–0.066	0.285	0.083	0.066
Cu (s)	–0.073	–0.197	0.052	0.115	0.034	0.128	–0.107
Ni (s)	–0.133	–0.070	**0.361**	–0.046	0.131	0.033	–0.079
Pb (s)	–0.170	–0.156	0.277	–0.164	–0.045	0.080	0.156

n = 72, bold values are significant at $p < 0.001$

metal data using varimax normalized rotation is shown in Fig. 1. Two factors were obtained with eigenvalues >1, cumulatively contributing 42.26% of variance. The first factor with a total variance of 22.76% manifests the contributions from Fe and Cr, which emanate from chemicals used in tanning process. The second factor with significant loadings for Zn contributes to 19.5% of total variance, was considered to originate from tannery process, contaminating the adjoining agricultural soil. Similarly, two factors are obtained for soil samples with eigenvalues >1, cumulatively explaining more than 74% of variance (Fig. 2). Factor 1, accounts for 21.18% of total variance has significant loading for Zn, Cu and Ni in the soil samples which may be soil based or may be derived from effluent. Factor 2 with high loading for Fe and Pb with 23.13% of total variance may also be associated with chemical used in tanning process. The CA was also performed to check the relationship among the elements and graphically viewed by the dendrograms for effluent (Fig. 3) and soil samples (Fig. 4), which supports the finding of PCA and correlation analysis. Tariq et al. (2005) also

Fig. 1 Principal component analysis applied to metals content in treated tannery effluent loading values of the initial variables

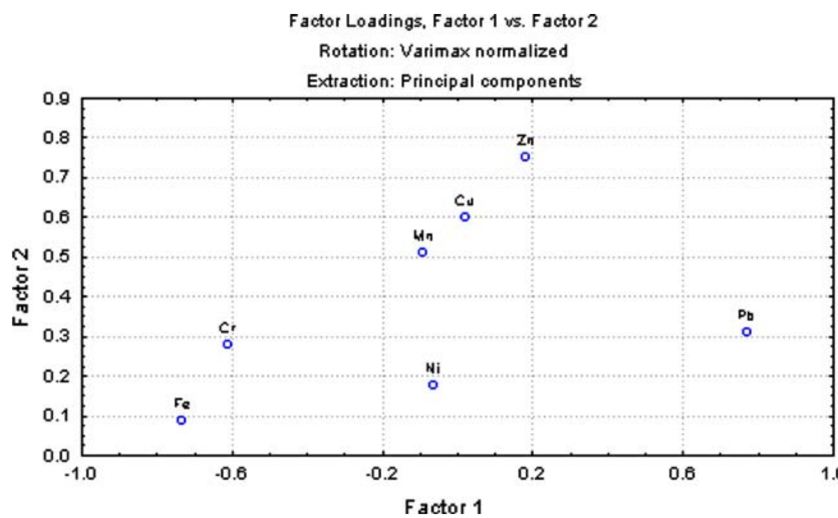
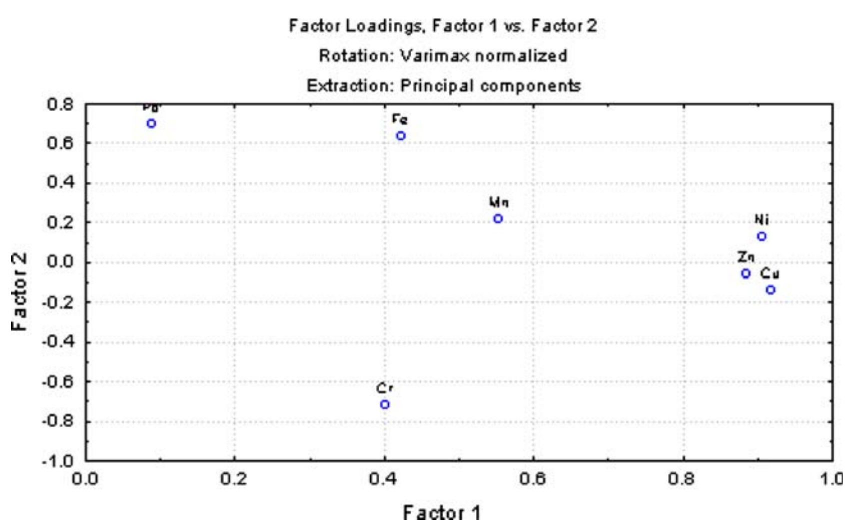


Fig. 2 Principal component analysis applied to metals content in soil samples loading values of the initial variablesv



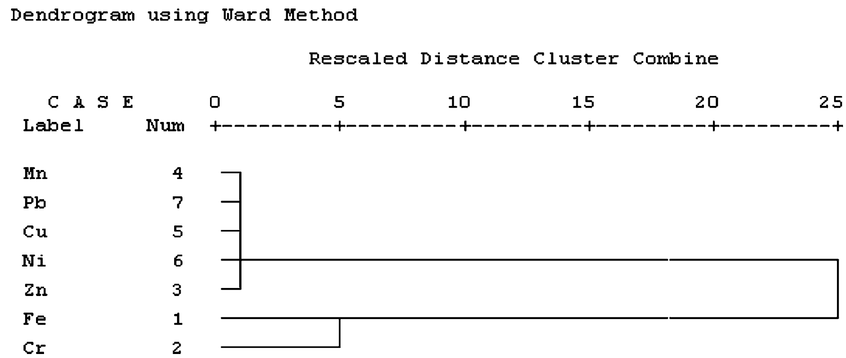


Fig. 3 Dendrogram of hierarchical cluster analysis between metals in treated effluent

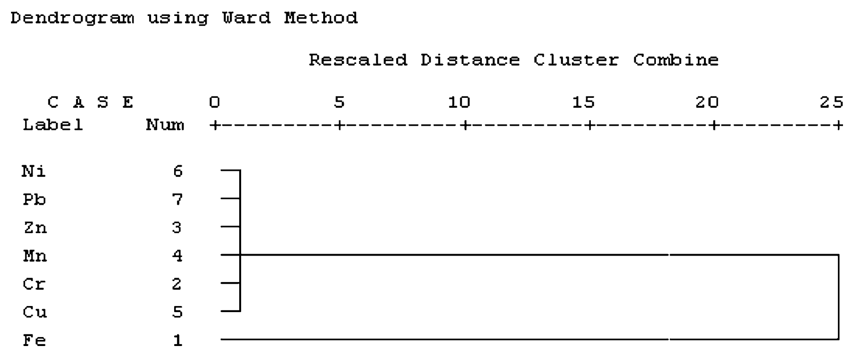


Fig. 4 Dendrogram of hierarchical cluster analysis between different metals in soil

studied contamination of soils receiving untreated effluents from 38 tanneries housed in Kasur, Pakistan and reported very high levels of metals, particularly Cr and Na in the effluents as well as in soils in the close proximity to the tanneries. High level of metals in soil was attributed to their elevated levels in untreated effluents discharged from tanneries.

The present study on trace metals distribution in UASB treated tannery effluent irrigated agricultural area of Jajmau, Kanpur evidences high concentration (mg L^{-1}) of Fe (6.19) and Cr (4.32) in the effluent and eventually in agricultural soils, with high concentration (mg kg^{-1} dw) of Fe (30,953) and Cr (859.7), which were enough to pose serious pollution stress to the environment in the vicinity of the tanneries. Other tested metals (Ni and Pb) were also found significantly high in treated tannery effluent and in agricultural soil where metal containing wastewater is being used for irrigation. Such alarming levels of metals may pose a direct health hazard to the people inhabiting that area. Sinha et al. (2006) reported the implications of metal contamination of agricultural soils due to long term irrigation with treated industrial wastewater and their subsequent accumulation in the vegetables/crops growing on such soils during the period 2002–2003. The present work (study period 2005–2006) demonstrated the usefulness of multivariate methods towards classifying the metals as groups in terms of their independent behavior and identifying their probable origin in effluents. The results of CA also support the finding of PCA and correlation analysis. It is suggested that the recovery and recycling of chemicals used in the tanning process may be undertaken in order to protect further contamination of soil.

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