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Environmental Health Risk Estimation of Heavy Metals Accumulated in Soil and Cultivated Plants Irrigated with Industrial Effluents

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Abstract

The lands of Kalipur area are contaminated with industrial wastewater and dust particles. Anticipating a possible health hazard through contaminated food crops with toxic metals like chromium, nickel and cadmium was investigated. The mobilization of metal from rhizosphere soil to plant tissues was calculated to determine the enrichment factors of soil and plants. Scanning Electron Microscope and Fourier Transform Infrared Spectroscopy were used for comparative study of surface soil contamination. The Ni, Cr and Cd in soil varied from 24.5 to 44.5, 42.4 to 65.5 and 14.2 to 31.6 µg g^{-1} enriched by a factor of 3.81, 4.64 and 20.94. Their corresponding values in cultivated plants and weeds were 12.6 ± 1.04 to 44.5 ± 2.84, 26.3 ± 1.64 to 67.5 ± 4.82 and 6.6 ± 0.84 to 22.3 ± 1.46 µg g^{-1} dry wt. enriched by a factor of 7.49, 6.89 and 22.08 respectively. All these metals are causing toxicity of soil while in plant tissues exceed the phytotoxicity limit and fall in the critical range. The causes of wide variation in metal uptake and accumulation in above ground plant parts and how weeds are growing luxuriously in spite of pollution and metal stress condition through evolution are well explained. Thus our study suggests that there is a health risk due to consumption of plants containing higher amounts of toxic metals resulting in asymptomatic chronic disorders in humans and cattle.

Keywords: Wastewater irrigation; Heavy metals; Soil-plants; SEM; FTIR; Toxicity; Health risk

Introduction

Indiscriminate release of industrial wastewater/mining wastes into the water bodies and reuse of this polluted wastewater for agriculture purpose is a topic of debate for sustainable development [1]. These wastewater containing heavy metals (density of >5 g cm^{-3}) in toxic levels is of great health concern for human and animal through food web [2,3]. Durgapur Industrial Belt (DIB) of India is discharging effluent into the Tamala Drain (TD) leading to metal pollution of water-soil system and also affects biota of the area. Locals farmers are using for cultivating food crops, pulses and vegetables because they are ignorant about the hidden toxicity of the factory discharged and their subsequent negative impacts such as loss of soil fertility/crop yield, bio-concentration of metals in edible parts or forage plants [4]. Although, some of the trace metals viz Fe, Mn, Zn and Cu are essential to plant life but Ni, Cr, Cd are toxic even at low concentrations. However, all these metals are toxic beyond a certain threshold value though it may vary with nature and species of metals and kinds of plants [5,6].

Nevertheless, metallic compounds are persistent and accumulate increasingly in soil media. The subsequent uptake and distribution of these metals in edible and fodder plants enhance the chances of health risk through food chain [7,8] though the extent of these toxic metals and the adverse impact thereof on human beings varies from one situation to another. Studies have established that metals are toxic to crops, animals and humans when plants are grown on contaminated soil using wastewater irrigation [9]. Cereals and vegetables constitute an important part of the human diet since they contain proteins, carbohydrates and vitamins as well as minerals and essential trace elements needed for different enzyme activities [10]. Consumption of metal containing crops and vegetables obliviously pose a chronic health hazard problem threat to human health [11]. At present, there is lack of information about the fatal limits of the toxic metals in human beings related to limits in plants but intake of toxic metals is known to induce chronic disorder in human being and others. Therefore it becomes imperative to undertake comprehensive studies for evaluation of metal pollution of water, soil, crops and vegetables grown on industrial sites.

The present investigation was conducted with the following objectives:

(i) To generate information about the accumulation of nickel, chromium and cadmium in the agricultural soil receiving wastewater irrigation and metal mobilization to cultivated crops and weeds grown in contaminated Kalipur area,

(ii) To find out the degree of contamination of soil and plants of Kalipur area with respect to soil and plants of uncontaminated Madhabpur area,

(iii) To assess the possible health risk through consumption of food crops containing higher amounts of toxic metals. Besides these objectives, Scanning Electron Microscope (SEM) is used for surface morphology of contaminated and uncontaminated soil and Fourier Transform Infrared Spectroscopy (FTIR) is used to study the functional groups of soil. Since plant differs in their ability to absorption; mobilization of metals from soil media to plants has been calculated.

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Materials and Methods

Geographical position and climatic condition of study area

The Kalipur area of DIB (23.48°N and 87.32°E, 65 m above mean sea level) is located on the river Damodar of West Bengal, India (Figure 1). DIB experiences transitional climate between the tropical wet and dry climate and the more humid subtropical climate. Summer season is hot (28-35°C) lasting from March to June. It is followed by the monsoon season (July to October) with precipitation and lower average temperature. It receives most of its annual rainfall (150 mm) during this season. A mild, dry winter (15-25°C) prevails from November to January which is followed by short spring during February.

Contamination sources of agricultural lands and crops

DIB area is richer with minerals and natural resources including abundance availability of coal. Therefore, Durgapur area is converted to DIB (which stretches out 50 km along the river Damodar). It is an industrial hub of 500 industries including steel, sponge iron, cement, coal washeries, power plant, chemical, fertilizer, pharmaceuticals as well as cluster of medium and small scale auxiliary industries [12]. Some of these are labeled as red colored industries and unable to compliance the discharge norms of pollution control board [13].

Farmers are cultivating their crops with TD wastewater. Tamala drain, the natural water drain gradually converted to wastewater channel carrying the effluent of DIB and is flowing across the agricultural lands before merging and discharging its wastes to the Damodar river at a distance of 25 km. During monsoon TD gets overflowed contaminating the whole area. While summer season, it receives wastewater irrigation and rest of the year is either contaminated with atmospheric fallout of dust particles bearing chemicals and metal oxides or with direct discharge of industrial wastes [13]. Hence, it is assumed that agricultural lands and crops are contaminated with hazardous chemicals and metals.

Collection of plant and soil samples

Plant and soil samples were collected from effluen contaminated fields of Kalipur area during March, 2012. Eleven plant (11) species of 8 cultivated plants {1 cereal (rice), 1 spice (coriander), 6 vegetables (brinjal, spinach, okra, radish, cabbage and amaranth) and 3 weeds (commelina, croton and parthenium)} were sampled from Kalipur area receiving wastewater irrigation (Table 1). These plants were sampled randomly. Brinjal plant did not bear brinjal. Spinach radish, cabbage and amaranth were leafy vegetables. The Plant samples analyses were collected from polluted site and also from unpolluted site (control site). For each plant sample, 10 to 15 plants of the same species were collected randomly from each of the locations in the polluted area and unpolluted area from where soil samples were drawn. It was ensured that the different samples of each plant species had the same physiological age and identical in appearance. Most of the plants were in flowering stage. Morphological status of plant species were taken into consideration based on numbers of leaves, shoot length, root length, color of the leaves, stage of growth, response to environmental stress condition. The morphological index was categorized as normal, healthy and luxurious. Rhizosphere soils were collected from surface soil to a depth of 30 cm. Similarly, control soil and plants were drawn from uncontaminated Madhabpur area receiving pond water irrigation at a distance of 20 km is devoid of industry and road intersections that may contribute significant amount of metals.

Sample preparation and analysis

Collected soil and plant samples were grounded by hand held Mortar and Pestle and after proper grinding sieved through <0.71 mm. Plant samples were washed first with running tap water to remove extraneous matter and then with distilled water. After washing, the sample was blotted dry, finely chopped and oven dried at 65°C. The dry sample was pulverized with hand held Mortar and Pestle and stored in kraft paper bags for further analysis. One gram of soil and plant sample was added with mixture of nitric and perchloric acids (4: 1 *v/v*) and the beaker were kept for overnight. It was digested on hot plate until a clear solution was obtained. The residue was diluted with 0.1N HNO_3 and then filtered and was assayed by AAS (Atomic Absorption Spectroscopy, AA plus 250, Varian, Australian make). The concentrations of Cr, Cd and Ni of digested soil/plant sample were measured using Varian model Spectr AA-250 plus Atomic Absorption Spectrophotometer (AAS). The major components of AAS are a radiation source, the flame, a monochromatic and a detector circuited with a recorder. Hollow cathode lamp was used as a source of radiation. Air-acetylene flame was employed for atomization of elements. Analyses were performed in triplicates and mean values are processed in Table 6.

Quality assurance and control

Appropriate quality assurance and procedures were followed to ensure the reliability and reproducibility of the results. Errors occurred during this study are linked to instruments, chemical impurity, data

Plant name		Kalipur Contaminated)			Madhabpur (Uncontaminated)		
Botanical	Common	Soil (µg g^{-1})	Plant (µg g^{-1})	Mob.*	Soil (µg g^{-1})	Plant (µg g^{-1})	Mob.
S1. *Amranthus viridis* L.	Amaranth	43.4 ± 1.98	26.5 ± 2.08	0.61	8.2 ± 0.80	3.5 ± 0.72	0.43
S2. *Solanum melongena* L.	Brinjal	39.1 ± 1.64	18.2 ± 1.94	0.47	11.3 ± 1.20	2.6 ± 0.40	0.23
S3. *Spinacea oleracea* L.	Spinach	35.4 ± 1.19	22.0 ± 2.10	0.62	13.7 ± 1.62	4.1 ± 0.52	0.3
S4. *Oryza sativa* L.	Rice	28.4 ± 1.12	12.6 ± 1.04	0.44	7.5 ± 0.74	1.8 ± 0.24	0.24
S5. *Hibiscus esculentus* L.	Okra	42.1 ± 2.18	17.8 ± 1.92	0.42	6.6 ± 0.82	2.4 ± 0.86	0.36
S6. *Raphanus sativus* L.	Radish	39.8 ± 1.42	15.1 ± 1.48	0.38	9.8 ± 1.16	2.5 ± 0.74	0.26
S7. *Brassica oleracea* L.	Cabbage	44.5 ± 2.18	19.7 ± 2.02	0.44	10.6 ± 1.32	2.2 ± 0.55	0.21
S8. *Coriandrum sativum* L.	Coriander	42.5 ± 1.62	14.6 ± 1.56	0.34	11.2 ± 1.42	3.1 ± 0.62	0.28
S9. *Commelina benghalensis* L.	Commelina	32.6 ± 1.45	38.4 ± 2.26	1.18	10.2 ± 1.14	3.3 ± 0.42	0.32
S10. *Croton bonplandianum* Baill.	Croton	24.5 ± 1.04	30.2 ± 2.24	1.23	13.1 ± 1.26	6.9 ± 1.20	0.53
S11.*Parthenium hysterophours* L.	Parthenium	38.6 ± 2.02	44.5 ± 2.84	1.15	11.7 ± 1.24	3.6 ± 1.12	0.31
Min		24.5	12.6	0.34	6.6	1.8	0.21
Max		44.5	44.5	1.23	13.7	6.9	0.53
Avg		37.35 ± 6.47	23.60 ± 10.31	0.66 ± 0.35	10.35 ± 2.22	3.27 ± 1.38	0.32 ± 0.09

Mob.*=Soil to plant mobilization

Table 1: Concentration of nickel in soil and plants of Kalipur and Madhabpur area.

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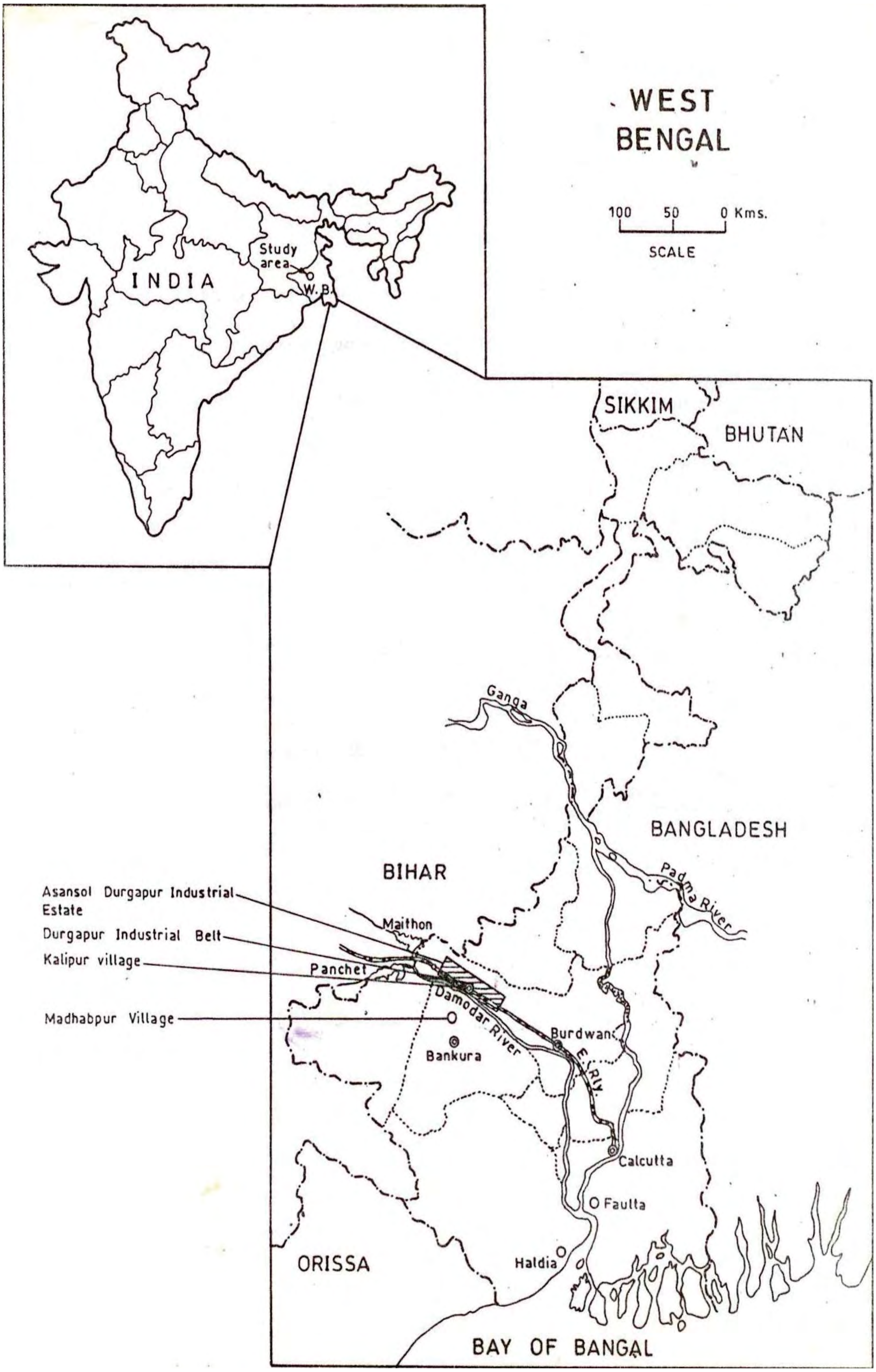

Figure 1: Layout of West Bengal showing Durgapur industrial estate and sampling locations for soil and plants in Kalipur (Contaminated) and Madhabpur (uncontaminated) field and the river Damodar.

processing, procedures followed and human error. Analytical grade chemicals and reagent blanks were used in all analyses to check reagent impurities. Freshly prepared reagents were standardized for actual strength. Prior sample analysis, instruments were calibrated and validated for erratic readings. Cross contamination is checked during sample grinding. Duplicate samples were read to verify the precision of the analytical method and instrument. CRM multi element standards solution IV (CertiPUR® 1.11355.0100 Lot. No. HC081563, Merck) was used during elemental analysis for validation and calibration of the AAS. Stepwise precautions were followed to minimize the scale of uncertainty which could be <5% of the observed value with respect to true value.

Enrichment factor

The contribution of TD irrigation carrying effluents metal concentrations in soil and plants of the Kalipur area has been determined by working out the enrichment factor related to the Madhabpur area.

$$\text{Soil(EFs) and plants (EFp)} = \frac{\text{Concentration of metal in soil or (Plant) at contaminated site}}{\text{Concentration of metal in soil or (Plant) at un contaminated site}}$$

Statistical analysis

Bivariate relationship between metals concentrations in soils and plants of contaminated and uncontaminated sites were estimated using the Origin 9.1. Coefficient of determination ^{2}) was work out to find out the percentage of best fit data through linear regression The Pearson's correlation coefficient, 'r' was calculated between met level in soils and plants and their significance was tested using 't' test

Results

The accumulation of metals in soil and plants of Kalipur and Madhabpur area, metal mobilization ratio, enrichment factors are shown in Tables 1-4 and in Figures 2-4.

Metal concentration in soil and plant and their mobilization

Nickel in soil: The nickel content of Kalipur soil varied from 24.5 to 44.5 (avg. 37.4 ± 6.5) μg Ni g^{-1} soil (Table 1). This was appreciably higher than the soil of the uncontaminated Madhabpur which contained 6.6 ± 0.82 to 13.7 ± 1.62 (avg. of 10.4 ± 2.2) μg Ni g^{-1} soil. As a result of TD irrigation of Kalipur area caused enrichment of nickel by a factor of 1.87 to 6.38 (avg. 3.81 ± 1.23).

Nickel in plant: Taking both the cultivated plants and weeds together, nickel concentration in plants of Kalipur and Madhabpur area were ranged from 12.6 to 44.5 (avg. 23.6 ± 10.3) and 1.8 to 6.9 (avg. 3.3 ± 1.4) μg Ni g^{-1} dry wt. respectively (Table 1). The nickel concentration in cultivated plants and weeds of the Kalipur area were

Plant name		Kalipur (Contaminated)			Madhabpur (Uncontaminated)		
Botanical	Common	Soil (μg g^{-1})	Plant (μg g^{-1})	Mob.*	Soil (μg g^{-1})	Plant (μg g^{-1})	Mob.
S1. *Amranthus viridis* L.	Amaranth	59.9 ± 2.82	33.9 ± 1.82	0.57	12.9 ± 1.28	3.5 ± 0.80	0.27
S2. *Solanum melongena* L.	Brinjal	65.5 ± 3.44	28.6 ± 1.46	0.44	9.5 ± 1.02	4.9 ± 0.78	0.52
S3. *Spinacea oleracea* L.	Spinach	65.2 ± 3.28	31.2 ± 1.80	0.48	16.3 ± 2.26	5.8 ± 0.98	0.36
S4. *Oryza sativa* L.	Rice	51.4 ± 2.94	26.3 ± 1.20	0.51	13.6 ± 2.02	4.3 ± 0.48	0.32
S5. *Hibiscus esculentus* L.	Okra	54.1 ± 2.62	39.2 ± 2.22	0.72	12.4 ± 1.98	6.3 ± 0.54	0.51
S6. *Raphanus sativus* L.	Radish	58.2 ± 3.26	26.4 ± 1.64	0.45	14.8 ± 1.92	4.5 ± 0.46	0.3
S7. *Brassica oleracea* L.	Cabbage	42.4 ± 2.48	26.8 ± 1.58	0.63	13.6 ± 1.48	5.7 ± 0.82	0.42
S8. *Coriandrum sativum* L.	Coriander	50.3 ± 2.04	30.7 ± 2.20	0.61	8.5 ± 1.22	3.3 ± 0.68	0.39
S9. *Commelina benghalensis* L.	Commelina	55.2 ± 3.42	48.7 ± 3.22	0.88	13.4 ± 1.42	8.9 ± 1.10	0.66
S10. *Croton bonplandianum* Baill.	Croton	59.6 ± 2.80	67.5 ± 4.82	1.13	11.3 ± 1.46	8.6 ± 1.12	0.76
S11. *Parthenium hysterophours* L.	Parthenium	53.6 ± 2.18	67.4 ± 4.28	1.26	10.7 ± 1.12	7.2 ± 0.96	0.67
Min		42.4	26.3	0.44	8.5	3.3	0.27
Max		65.5	67.5	1.26	16.3	8.9	0.76
Avg		55.95 ± 6.76	38.79 ± 15.64	0.70 ± 0.28	12.45 ± 2.30	5.73 ± 1.89	0.47 ± 0.17
Mob.*=Soil to plant mobilization							

Table 2: Concentration of chromium in soil and plants of Kalipur and Madhabpur area.

Plant name		Kalipur (Contaminated)			Madhabpur (Uncontaminated)		
Botanical	Common	Soil (μg g^{-1})	Plant (μg g^{-1})	Mob.*	Soil (μg g^{-1})	Plant (μg g^{-1})	Mob.
S1. *Amranthus viridis* L.	Amaranth	23.9 ± 2.10	9.0 ± 0.80	0.38	1.29 ± 0.40	0.51 ± 0.24	0.4
S2. *Solanum melongena* L.	Brinjal	16.7 ± 1.68	7.7 ± 0.78	0.46	0.82 ± 0.20	0.46 ± 0.12	0.56
S3. *Spinacea oleracea* L.	Spinach	17.4 ± 1.72	7.3 ± 0.68	0.42	1.89 ± 0.22	0.59 ± 0.14	0.31
S4. *Oryza sativa* L.	Rice	16.9 ± 1.24	8.3 ± 0.64	0.49	0.89 ± 0.10	0.47 ± 0.12	0.53
S5. *Hibiscus esculentus* L.	Okra	22.4 ± 1.82	8.2 ± 0.48	0.37	0.89 ± 0.12	0.55 ± 0.16	0.62
S6. *Raphanus sativus* L.	Radish	14.2 ± 1.08	6.6 ± 0.84	0.46	1.48 ± .022	0.79 ± 0.18	0.53
S7. *Brassica oleracea* L.	Cabbage	22.1 ± 1.88	9.7 ± 1.10	0.44	0.58 ± 0.12	0.35 ± 0.11	0.6
S8. *Coriandrum sativum* L.	Coriander	24.4 ± 2.10	11.4 ± 1.20	0.47	0.99 ± 0.16	0.38 ± 0.12	0.38
S9. *Commelina benghalensis* L.	Commelina	31.6 ± 2.24	22.3 ± 1.46	0.71	0.99 ± 0.18	0.52 ± 0.22	0.53
S10. *Croton bonplandianum* Baill.	Croton	17.9 ± 1.68	16.5 ± 1.22	0.92	1.28 ± 0.22	0.66 ± 0.14	0.52
S11. *Parthenium hysterophours* L.	Parthenium	15.7 ± 1.46	16.3 ± 1.20	1.04	0.79 ± 0.14	0.55 ± 0.12	0.7
Min		14.2	6.6	0.37	0.58	0.35	0.31
Max		31.6	22.3	1.04	1.89	0.79	0.7
Avg		20.29 ± 5.12	11.21 ± 5.00	0.56 ± 0.23	1.08 ± 0.37	0.53 ± 0.12	0.52 ± 0.11
Mob.*=Soil to plant mobilization							

Table 3: Concentration of cadmium in soil and plants of Kalipur and Madhabpur area.

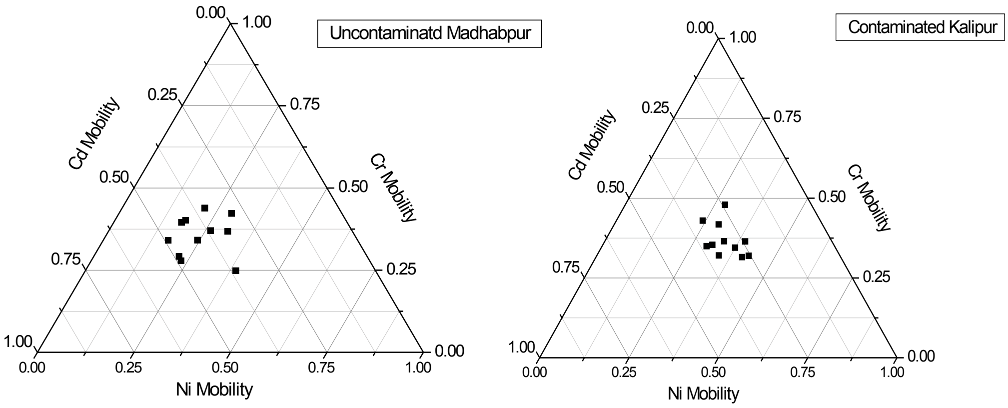

Figure 2: Mobilization of Ni, Cd and Cr from rhizosphere soil to plant in (i) Kalipur area and (ii) Madhabpur area.

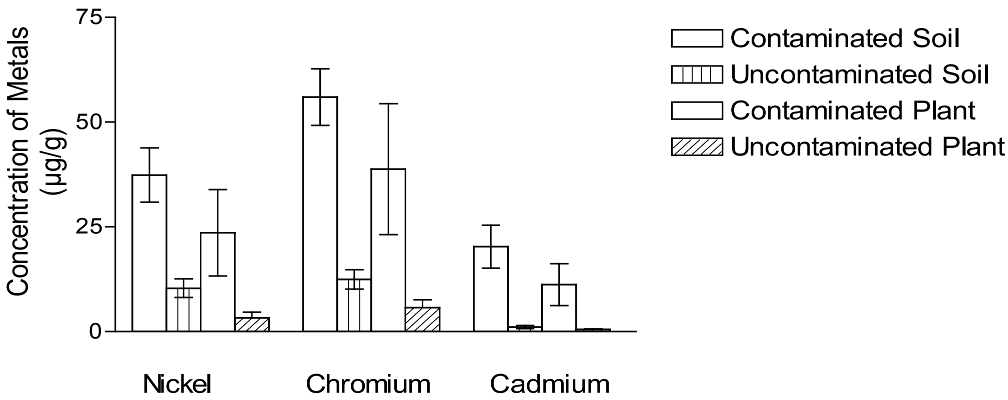

Figure 3: Metal concentrations in soil and plants of Kalipur (contaminated) and Madhabpur (uncontaminated) areas.

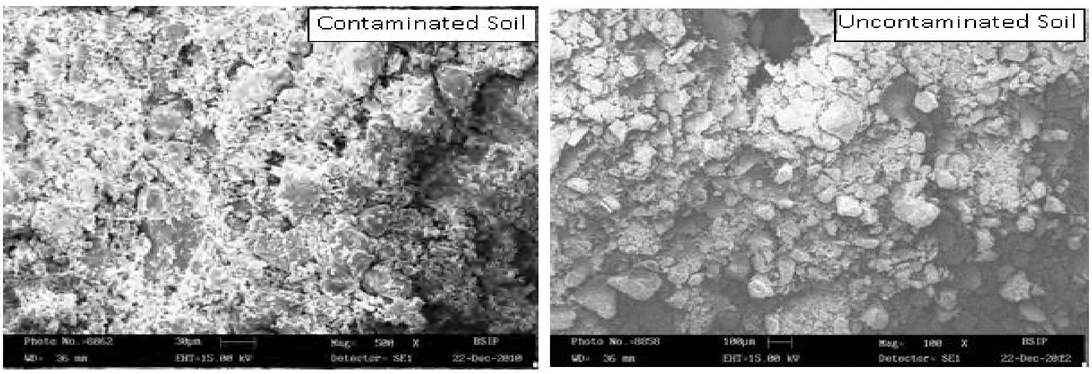

Figure 4: Photomicrographs showing the surface morphology of soils collected from contaminated-Kalipur and uncontaminated-Madhabpur areas.

ranged from 12.6 ± 1.04 to 26.5 ± 2.08 (avg. 18.3 ± 4.5) and 30.2 ± 2.24 to 44.5 ± 2.84 (avg. 37.9 ± 7.1) µg g^{-1} dry wt. and in Madhabpur, it was 1.8 ± 0.24 to 4.1 ± 0.52 (avg. 2.8 ± 0.7) and 3.3 ± 0.42 to 6.9 ± 1.20 (avg. 4.6 ± 2.0) µg g^{-1} dry wt. respectively. Plants of Kalipur area showed nickel enrichment by a factor of 4.38 to 12. The enrichment was relatively high (EFp: 12.36) in case of Parthenium and low (EFp: 4.38) in case of Croton.

Nickel mobilization: The soil to plant nickel mobilization ratio (MR) in cultivated plants and weeds of the Kalipur area were ranged from 0.34 to 0.62 (avg. 0.47 ± 0.1) and 1.15 to 1.23 (avg. 1.19 ± 0.04) respectively (Figure 2). In the Madhabpur area, it is varied from 0.21 to 0.43 (avg. 0.29 ± 0.07) in the cultivated plants and 0.31 to 0.53 (avg. 0.38 ± 0.12) in the weeds. The soil to plant MR had about the same in the Kalipur and the Madhabpur area.

Chromium in soil: The chromium concentration in Kalipur soil ranged between 42.4 to 65.5 (avg. 55.9 ± 6.76) µg g^{-1} soil (Table 2). This was appreciably higher than the chromium concentration in the Madhabpur soil which contained 8.5 to 16.3 (avg. of 12.45 ± 2.3) µg g^{-1} soil. As a result of TD irrigation, the Kalipur soil showed enrichment by a factor of 3.12 to 6.89 (Table 4).

Chromium in plant: In the eleven plant species grown in Kalipur area, the chromium ranged from 26.3 to 67.5 (avg. 38.79 ± 15.64) µg

Plant name		Nickel		Chromium		Cadmium	
Botanical	Common	Soil (μg g^{-1})	Plant (μg g^{-1})	Soil (μg g^{-1})	Plant (μg g^{-1})	Soil (μg g^{-1})	Plant (μg g^{-1})
S1. *Amranthus viridis* L.	Amaranth	5.29 ± 1.10	7.57 ± 1.42	4.64 ± 0.88	9.69 ± 1.20	18.53 ± 2.20	17.65 ± 1.98
S2. *Solanum melongena* L.	Brinjal	3.46 ± 0.80	7.00 ± 1.28	6.59 ± 0.48	5.84 ± 0.86	20.37 ± 2.24	16.74 ± 1.44
S3. *Spinacea oleracea* L.	Spinach	2.55 ± 0.64	5.37 ± 0.88	4.00 ± 0.38	5.38 ± 0.46	9.21 ± 1.28	12.37 ± 1.02
S4. *Oryza sativa* L.	Rice	3.79 ± 0.44	7.00 ± 1.10	3.78 ± 0.34	6.12 ± 0.48	18.99 ± 2.20	17.66 ± 1.28
S5. *Hibiscus esculentus* L.	Okra	6.38 ± 0.48	7.42 ± 1.12	4.36 ± 0.32	6.22 ± 0.42	25.17 ± 2.46	14.91 ± 1.64
S6. *Raphanus sativus* L.	Radish	4.06 ± 0.42	6.04 ± 0.88	3.93 ± 0.28	5.87 ± 0.38	9.59 ± 1.46	8.35 ± 1.20
S7. *Brassica oleracea* L.	Cabbage	4.20 ± 0.24	8.95 ± 0.42	3.12 ± 0.32	4.70 ± 0.28	38.10 ± 2.68	27.71 ± 2.42
S8. *Coriandrum sativum* L.	Coriander	3.79 ± 0.22	4.71 ± 0.26	5.92 ± 0.86	9.30 ± 1.22	24.65 ± 1.80	30.00 ± 2.86
S9. *Commelina benghalensis* L.	Commelina	3.20 ± 0.20	11.64 ± 1.20	4.12 ± 0.46	5.47 ± 0.88	31.92 ± 2.22	42.88 ± 3.20
S10. *Croton bonplandianum* Baill.	Croton	1.87 ± 0.10	4.38 ± 0.28	5.27 ± 0.42	7.85 ± 0.38	13.98 ± 1.82	25.00 ± 2.98
S11. *Parthenium hesterophours* L.	Parthenium	3.30 ± 0.20	12.36 ± 1.38	5.01 ± 0.24	9.36 ± 1.12	19.87 ± 1.48	29.64 ± 2.66
Min		1.87	4.38	3.12	4.7	9.21	8.35
Max		6.38	12.36	6.89	9.69	38.1	42.88
Avg		3.81 ± 1.23	7.49 ± 2.60	4.64 ± 1.08	6.89 ± 1.82	20.94 ± 8.97	22.08 ± 9.97

Table 4: Enrichment factor in soil and plant sampled from Kalipur.

g^{-1} dry wt and 3.3 to 8.9 (5.73 ± 1.89) μg g^{-1} dry wt. in Madhabpur area respectively (Table 2). Chromium concentration in the cultivated plants and weeds of the Kalipur area were ranged from 26.3 ± 1.20 to 39.2 ± 2.22 (avg. 30.39 ± 4.46) and 48.7 ± 3.22 to 67.5 ± 4.28 (avg. 61.2 ± 10.83) μg g^{-1} dry wt. respectively. The highest concentration was observed in Croton collected from Kalipur area. The plants of Kalipur showed enrichment by a factor of 4.70 to 9.69 (Table 4).

Chromium mobilization: The eleven plant species were taken together, MR were varied from 0.44 to 1.26 (avg. 0.70 ± 0.28) in Kalipur area and 0.27 to 0.76 (avg. 0.47 ± 0.17) in Madhabpur area (Table 2 and Figure 2). Thus, it is evident that soil to plant MR in the Kalipur area was higher than in the Madhabpur area.

Cadmium in soil: The Kalipur soil contained 14.2 to 31.6 (avg. 20.29 ± 5.12) μg g^{-1} soil. This was higher than in the Madhabpur soil, which contained 0.58 to 1.89 (avg. 1.08 ± 0.37) μg g^{-1} (Table 3). As a result of wastewater irrigation, the Kalipur soil showed enrichment by a factor of 9.21 to 38.10.

Cadmium in plant: The concentration in the eleven plant species of Kalipur and Madhabpur were ranged 6.60 to 22.30 (avg. 11.21 ± 5.0) and 0.35 to 0.79 (avg. 0.53 ± 0.12) μg g^{-1} dry wt. (Table 3). The concentration in the cultivated plants and weeds of Kalipur were ranged from 6.6 ± 0.84 to 11.4 ± 1.20 (avg. 8.53 ± 1.51) and 16.3 ± 1.22 to 22.3 ± 1.46 (avg. 18.37 ± 3.41) μg g^{-1} dry wt. respectively. The plants of the Kalipur area showed enrichment due to TD wastewater irrigation by a factor of 8.35 to 42.88 (avg. 22.08 ± 9.97) (Table 4).

Cadmium mobilization: The MR in the eleven plant species taken together varied from 0.37 to 1.04 (avg. 0.56 ± 0.23) in the Kalipur area and from 0.31 to 0.70 (avg. 0.52 ± 0.11) in the Madhabpur area (Table 3 and Figure 2).

Enrichment factor in soil and plants of Kalipur area

Enrichment factors varied in soil and plants but it showed the same order of enrichment both in soil and plants i.e., Cd<Cr<Ni. TD irrigation is increasing the metal concentrations in cultivated plants and weeds growing wild at the Kalipur with respect to their reference values. The elevated levels of Cd, Cr and Ni in plants grown in the Kalipur area may likely to constitute a significant chronic health hazard problem to the consumers in the long run. Therefore, daily intake of such plants which accumulate more toxic metals should be avoided from consumption. This is true for edible plant species particularly leafy vegetables. Usually, the root values of metals are one to two orders of magnitude higher than their shoot parts. Whatever the amount accumulated by leafy and root/tuber vegetables, there is a fair chance of direct entry into human diet. In few cases fodder plants may also increase dietary intake of metals through grazing animals and thereby subsequent tropic levels [14].

Discussion

The soil and plant samples of contaminated Kalipur area showed relatively higher accumulation of cadmium>chromium>nickel than soil and plant samples of the uncontaminated Madhabpur area.

Nickel toxicity to soil and plants

The nickel concentration in normal soil has been reported to range from 2 to 750 mg kg^{-1} [15], with a typical value of 40 μg g^{-1} [16] or 50 μg g^{-1} [17]. The critical limit for toxicity has been suggested to be around 100 μg g^{-1} soils by [18-21]. The average concentration 37.35 ± 6.47 in the Kalipur soil was well below the reported critical limit for its toxicity.

Nickel concentration in plants of Kalipur area was much higher than the normal concentration of nickel in plants as reported by Ref. [15,22]. The former reported 1 μg g^{-1} and the latter reported 0.02 to 5 μg g^{-1} in normal plants. In 6 out of 11 plant species of Kalipur exceeded 10 μg g^{-1}, which denotes the threshold of its toxicity. The critical toxic concentration in plants has been reported to range from 8 to 220 μg g^{-1} according to Ref. [23] while 10 to 100 μg g^{-1} according to Ref. [24].

Chromium toxicity to soil and plants: The chromium concentration in normal soil has been reported to range from 5 to 1500 μg g^{-1} [15], with a typical value of 100 μg g^{-1} [16] or 70 μg g^{-1} [17]. The critical limit for chromium toxicity has been suggested to 100 μg g^{-1} soil by Ref. [18-21] and 75 μg g^{-1} by Ref. [19]. The concentration 55.95 ± 6.76 μg g^{-1} of Kalipur soil was well below the reported critical limit for its toxicity.

Chromium concentration in plants of the Kalipur area also suggestive of its toxicity. Ref. [22] reported normal concentration in plants varies from 0.2 to 1.0 μg g^{-1} and Ref. [15] reported 0.03 to 14.0 μg g^{-1} Cr in normal plants. The critical toxic concentration ranges from 2.0 to 18.0 μg g^{-1} [23] and 5.0 to 30.0 μg g^{-1} [24]. The concentrations of Kalipur plant fall in this range.

Cadmium toxicity to soil and plants: The cadmium concentration in normal soil has been reported to range from 0.01 to 20.0 μg g^{-1} [15], with a typical value of 0.06 μg g^{-1} [16] or 0.35 μg g^{-1} [17]. The critical limit for toxicity has been suggested to 5.0 μg g^{-1} soil by Ref. [18,20] and

3.0 μg g^{-1} by Ref. [21]. The concentration 20.29 ± 5.12 μg g^{-1} in Kalipur soil was below reported critical limit for its toxicity.

The normal cadmium concentrations in plants are 0.2 to 0.8 μg g^{-1} dry wt. [22] and 0.1 to 2.4 μg g^{-1} [15]. Ref. [23] had suggested toxic concentration 4.0 to 200.0 μg g^{-1} in plants. According to Ref. [24], this limit is 5.0 to 30.0 μg g^{-1}. The concentration in plants of Kalipur area (6.6 to 22.3 μg g^{-1} dry wt.) tended to fall in the range of the critical toxic concentration.

Cause of variation of metals in soil and plants: Accumulation of different metals in plants, related to their metal pool in soil, revealed to their differences in mobility of metals from species to species and soil to soil even in same field (Figure 3). Metal uptake by plants has, however, been reported to depend, not only on the concentration in soil but also on the form in which they are present in the soil [25]. The metal level in plants is associated with exchangeable metals in the soil, which is affected by physical, chemical and biological factors. The availability of a metal in the soil-plant system is influenced by soil properties such as pH, organic matter and cation exchange capacity [26]; the nature of plant species, stage of growth and season of growth; type of microbial biomass, mycorrhizae, root exudates and environmental health (soil flora/ fauna and terrestrial animals)

Ref. [27] reported a difference in copper accumulation by legumes and grasses. Metal variation in plants is due to the availability of metals to plants that depend on total concentration in the soil and by the forms in which they occur [28]. Weeds of Kalipur area showed higher metal accumulation than the cultivated plants and vegetables. In spite of this, weeds of Kalipur area demonstrated luxurious growth and healthy morphology than the cultivated ones. This is possible only when weeds of polluted area develop greater tolerance to pollution stress condition and altering its biochemical pathways and physiological function occurred during the evolution. Wide differences in the mobilization ratio of Kalipur plants suggest that some species can efficientl restrict the passage of these elements from the contaminated soil to safeguard their progeny against their toxicity. Such a restriction could be attributed to their genetic makeup, expressed in their structural and functional attributes such as morphological and anatomical feature and mechanism of ion uptake and transport [29].

Effects of heavy metal on plant growth and development

Cultivated plants of Kalipur showed stunted growth. Excess accumulation of metal hampers normal physiological functions especially its metabolic cycles resulted in the abnormal or stunted growth. Figure 4 indicates that soil particles are densely packed and porosity is less in case of contaminated soil whereas particles are loosely bound in case of uncontaminated soil. Salts are deposited on the surface soil receiving TD wastewater. These salts get deposited in spaces exist among soil particles and densely packed with them. This reduces soil porosity and makes soil unfertile and unsuitable for root growth and overall development. The leaf and fruit size, length and branching of root/shoot were restricted. A large percentage of plants showed general or intervenal chlorosis and necrosis of leaves. In case of brinjal, fruiting phenomenon was drastically reduced while in case of tomato, size and numbers of fruit were restricted. Cabbage also showed abnormal growth. Specific and characteristic symptoms of toxicity of a particular element however, have not been observed in any plant species examined. Plants subjected to toxic symptoms of metals may not show visible symptoms, yet undergo hidden injury due to the toxic effects of metals. Such plants may also show metabolic changes

Ref. [30] reported that deposition of cement dust on cherry plants prevented the germination of its pollen grains on the stigmas. The loss of fruiting phenomenon in brinjal, hampering of fertilization and about 20% reduction in crop yield observed in Kalipur area is might be an outcome of deposition of chemical dust. Metal accumulation in soil is of great concern in agricultural production due to adverse effect on food quality (safety and marketability), crop growth, and soil fertility and hence affects agricultural economics of third world countries [31]

FTIR spectra interpretation of soil samples

The FTIR spectra of contaminated soil and control soil are shown in Figure 5. Apart from the metallic ions (frequency region 400-750 cm^{-1}), many shoulders of different range (626.56-3400.5) were observed in contaminated soil spectrum [32]. A broad shoulder around the 3400 cm^{-1} indicates the O-H (H- bonded) stretching of water vapor. The peak around 3019.46 cm^{-1}, 2400 cm^{-1} and 1629.4 cm^{-1} are due to C-H (alkenes), C≡N (cyano) and C=O (amide) functional groups respectively. It was found that the contaminated soil showed greater number of shoulders of different functional groups than control soil. The metals detected in soil by AAS are in accordance with the FTIR spectrum which confirms the presence of metals by the shoulders region 450-750 cm^{-1}. Thus, FTIR study suggests that soil of Kalipur is grossly polluted with many pollutants including metals.

Statistical interpretation: We have attempted to find out whether there exists any correlation between metal levels in soil and plant by determining the coefficien of determination (R^2) and correlation coefficien (r) (Table 5 and Figure 6). About 40% Ni data of uncontaminated area and 49% Cd data in contaminated area could reliably be explained by the linear regression. Nickel data of contaminated area and Cr of both contaminated and uncontaminated areas showed least relationship. Significant positive correlations were observed between Ni in contaminated plant and Cr in contaminated plant ($r=0.836$, $p<0.01$), Ni in contaminated plant and Cd in contaminated plant ($r=0.808$, $P<0.01$), Cd in uncontaminated soil and Cd in uncontaminated plant ($r=0.648$, $p<0.025$). In contrary, weak correlation was found between Ni in contaminated plant and Cd in contaminated soil ($r=0.233$, $p<0.085$). It can be inferred that in field conditions there may be many influencing factors other than soil metal pool which control the plant's concentration.

Possible mechanisms of root metal uptake and role of soil properties

The metal ions get entry into the root hairs either by active absorption due to metabolic energy, transpiration or passive adsorption due to the diffusion of ions from the soil solution into the root endodermis or both the active and passive forces work coherently. The trace elements and toxic metals enter into the root system in competitive way; it means non-essential metals often bind with the cofactor instead of essential metal. It is presumed that plants readily take up essential metals than non-essential that are dissolved in soil solution in either ionic or chelated and complexes forms. The bioavailability and toxicity is mostly governed by soil genesis and properties as well as surrounding microbial community.

Interactions and antagonistic effects of metals: Interactions amongst different elements exist in soil media could be also account for differential accumulation of elements in plants. Cadmium, manganese and iron are known to be antagonistic to zinc. In this investigation, it is not unlikely that accumulation of cadmium and lead in toxic concentrations could have prevented accumulation of zinc in plants in toxic concentrations. Nickel and manganese are reported to show antagonistic interaction. The higher accumulation of nickel could have prevented high accumulation of manganese in plants. Copper

			Ni				Cr				Cd			
			Contaminated		Uncontaminated		Contaminated		Uncontaminated		Contaminated		Uncontaminated	
			Soil	Plant	Soil	Plant	Soil	Plant	Soil	Plant	Soil	Plant	Soil	Plant
Ni	Contaminated	Soil	1											
		Plant	-0.213	1										
	Uncontaminated	Soil	-0.266	0.334	1									
		Plant	-0.549[c]	0.475	0.635[b]	1								
Cr	Contaminated	Soil	-0.257	0.078	0.326	0.392	1							
		Plant	-0.451	0.836[a]	0.352	0.717[a]	0.075	1						
	Uncontaminated	Soil	-0.126	-0.086	-0.078	-0.109	0.099	-0.279	1					
		Plant	-0.533[c]	0.713[b]	0.337	0.538[c]	0.042	0.770[a]	0.089	1				
Cd	Contaminated	Soil	0.157	0.233	-0.231	-0.054	-0.298	0.021	-0.084	0.211	1			
		Plant	-0.418	0.808[a]	0.269	0.46	-0.152	0.757[b]	-0.232	0.77[a]	0.527[c]	1		
	Uncontaminated	Soil	-0.229	-0.073	0.364	0.428	0.664[b]	-0.067	0.547[b]	-0.061	-0.224	-0.221	1	
		Plant	-0.344	0.136	0.125	0.385	0.521[c]	0.283	0.409	0.267	-0.441	-0.017	0.648[b]	1

[a]Values significant at 0.01 level of probability
[b]Values significant at 0.025 level of probability
[c]Values significant at 0.05 level of probability

Table 5: Correlation coefficient (r) values for metal level in soils and plants for Contaminated and uncontaminated sites.

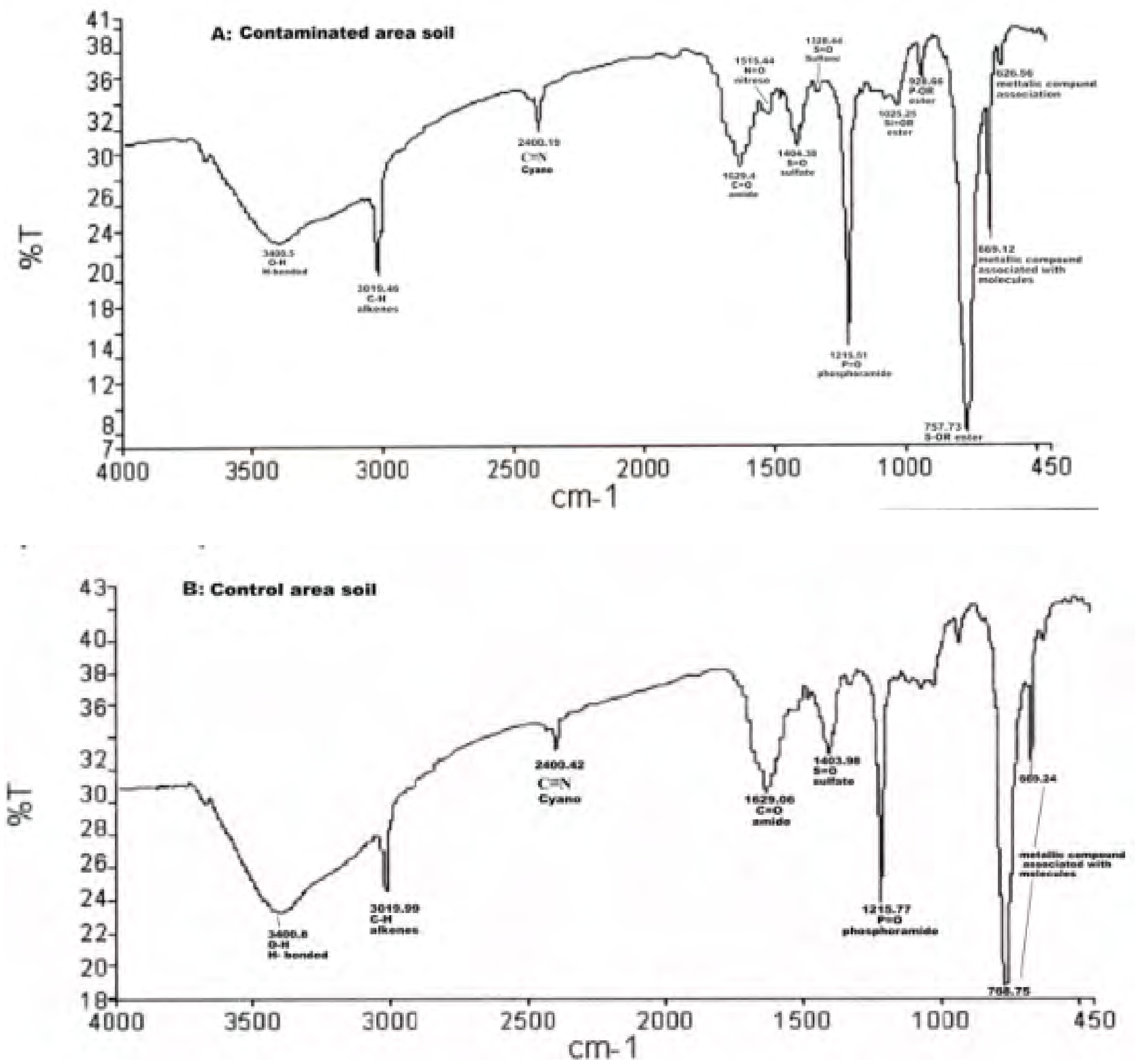

Figure 5: FTIR Spectra of (A) Contaminated soil and (B) Control soil.

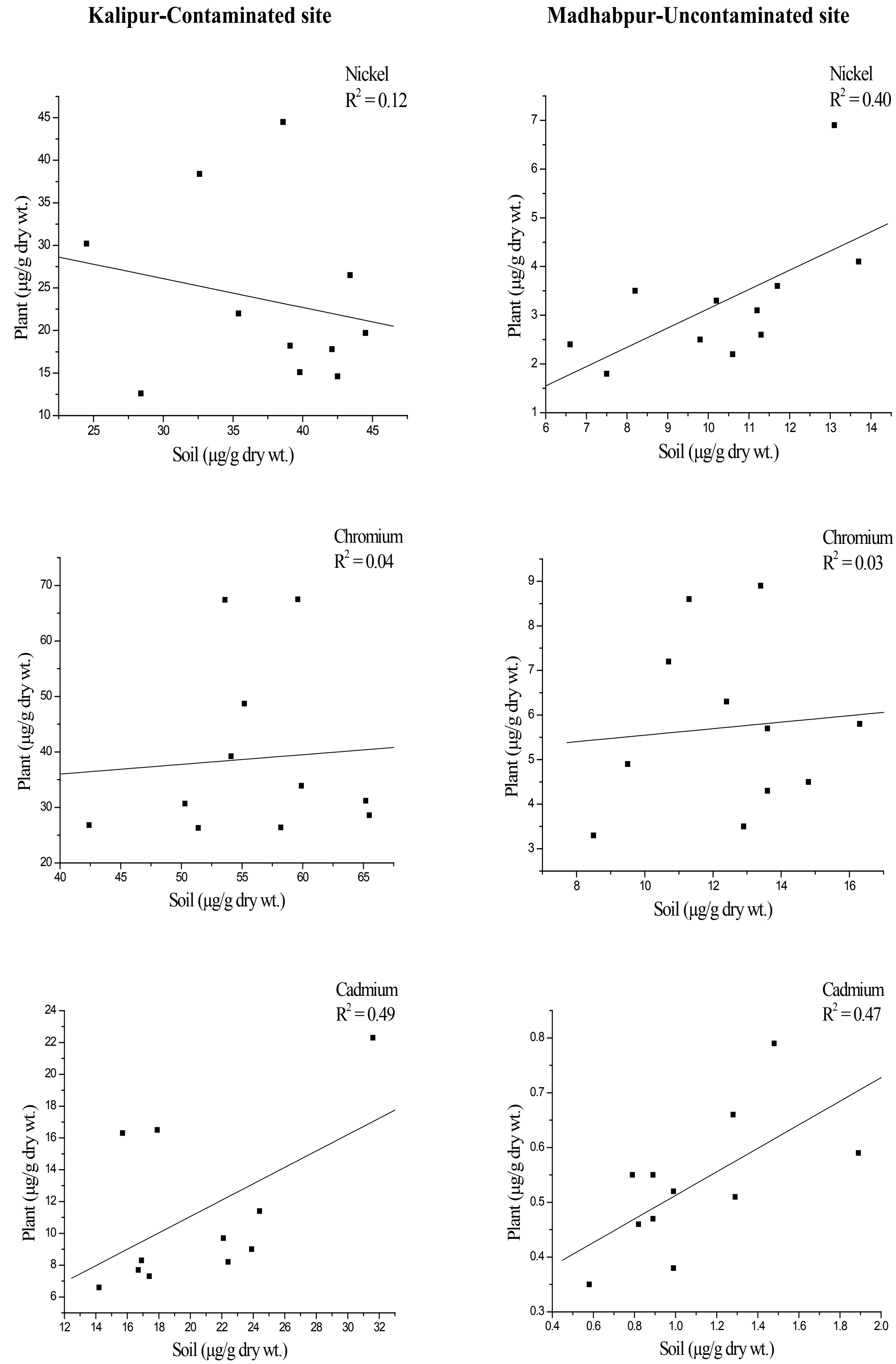

Figure 6: Relationship between heavy metals (Ni, Cr and Cd) concentrations (µg/g dry wt.) in soil and plant samples drawn from contaminated and uncontaminated sites.

Metal	Wave length (nm)	Current (Milli ampere)
Nickel	232.0	25
Chromium	357.9	25
Cadmium	228.8	4

Table 6: The conditions of the AAS for determination of the Cr, Cd and Ni were as follows.

and chromium show antagonistic effects. There is a possibility that accumulation of copper in toxic concentrations could have prevented accumulation of chromium in toxic concentrations.

Carcinogenic potential of nickel, chromium and cadmium: The US EPA has classified chemicals into five groups for carcinogenicity. Group A (Human Carcinogen), B1 and B2 (Probable Human Carcinogen), C (Possible Human Carcinogen), D (Not Classifiable as to Human Carcinogenicity), E (Evidence of Non-Carcinogenicity for Humans). Group B1 is for agents with limited evidence of carcinogenicity from epidemiological studies in humans. The agents with "sufficient evidence from animal studies and for which there is "inadequate evidence" or "no data" from epidemiologic, studies would usually be categorized under Group B2. Cr^{6+} is classified as group A (Human Carcinogen), so it is assumed that of the total Cr concentration, Cr^{6+} contributes about one seventh. USEPA has classified nickel carbonyl as Group B2, probable human carcinogen (1999A) and cadmium as Group B1, probable human carcinogen.

Minimal risk levels of cadmium, chromium and nickel: According to the Agency for Toxic Substances and Disease Registry [33], the Minimal Risk Levels (MRLs) is an estimate of the daily human exposure to a hazardous substance that is likely to be without appreciable risk of adverse non-cancer health effects over a specified duration of exposure. The MRLs of Cr^{6+} for intermediate (1-15 days) and chronic exposure (1 Year or longer) are 0.005 and 0.0009 mg/kg per day while for Cd 0.0005 and 0.0001 mg/kg per day respectively. It has not estimate the value for nickel by oral ingestion. Once metals absorbed by the body are only slowly excreted and as a consequence, metal toxicity is cumulative.

The daily intake of nickel from food will vary widely because of different dietary habitats and can range from 100-800 µg/day; the mean dietary intake in most countries is 100-300 µg/day. The average daily recommended value of Ni via food which is 250 µg/day [34]. According to Ref. [35] more than 5 ppm of Ni in edible plant parts may pose pollution problem. The average daily intake recommended value of Cr via food which is 60 µg/day. Chromium (Cr^{3+}) is considered least toxic among essential trace metals on the basis of essential to toxic ratio. Nephritis, anuria and extensive lesions in kidney and gastrointestinal ulceration are noticed in human suffering from C^{6+} toxicity.

The joint expert committee on food additives has established a provisional tolerable weekly intake of 7 µg/kg body weight of cadmium i.e., daily tolerable intake level of 70 µg per day for 70 kg man and 60 µg of per day for 60 kg women [36]. Ref. [37] reported the mean daily intake of Cd 90 µg /day from different places in the US. Ref. [39] have stated that in most countries, the daily intake of Cd is in the range 25 to 75 µg/day. Symptoms of chronic Cd toxicity include growth retardation and impaired kidney function. Consumption of cadmium containing cereals covering two to three decades caused a Itai-Itai disease among Japanese [38,39].

Conclusions

Findings could be summed up that the soil and plants of Kalipur of DIB receiving industrial wastewater are polluted with metals and tended to approach the toxic levels. Bioaccumulation of hazardous metals like nickel, chromium and cadmium in cultivable crops, weeds and fodder without showing visible sign of phytotoxicity may pose serious problem of toxicity to human and animals. Our expositions presented in this paper call for rigorous imposition of pollution control measures in relation to discharge of untreated or partially treated effluent from the industries before allowed to mixing in water bodies may help in providing water for irrigation purpose for the farmers without harming their life. Entry of metals through wastewater into cultivated crops may be reduced by proper soil and crop management practices. For prospective utilization of contaminated agricultural lands or to reuse wastewater for irrigation; mobilization ratio, enrichment factor and statistical techniques for soil-plant relationship should be the criteria of plant selection which can grow not only in metal contaminated land but also restrict efficiently the passage of metal from soil to plan

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