

Spectroscopic Quantification of Heavy Metal Bioaccumulation, Soil-to-Plant Partitioning, and Dietary Toxicity in Four Economic Crops Irrigated with Textile Industrial Wastewater in Bhilwara, Semi-Arid Rajasthan

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ABSTRACT

In the semi-arid industrial basin of Bhilwara, Rajasthan, effluent discharge from textile wet-processing, dyeing, and finishing units contaminates ephemeral drainage channels and unconfined aquifers. This investigation quantifies toxic trace heavy metal concentrations (Cd, Cr, Pb) across the irrigation water–soil–crop continuum under a factorial field design ($n = 36$) comparing an unamended groundwater control (C1–C4), vermicompost-amended organic management (OS1–OS4), and composite textile effluent irrigation (TS1–TS4) across four contrasting economic taxa: Wheatgrass (*Triticum aestivum* L.), Fenugreek (*Trigonella foenum-graecum* L.), Spinach (*Spinacia oleracea* L.), and Spearmint (*Mentha spicata* L.). Flame atomic absorption spectroscopy (AAS) following matrix-specific wet acid digestions revealed high contaminant loadings in composite textile wastewater: Pb (4.332 mg L^{-1}), Cr (3.388 mg L^{-1}), and Cd (1.492 mg L^{-1}), exceeding permissible national irrigation thresholds (CPCB/BIS) by 43.3-, 33.9-, and 149.2-fold, respectively. In harvested foliar biomass, trace metal bioaccumulation followed the species-dependent hierarchy: *Spinach* (TS3) $\approx$ *Spearmint* (TS4) $>$ *Fenugreek* (TS2) $>$ *Wheatgrass* (TS1). Foliar lead levels in *S. oleracea* and *M. spicata* reached $14.610 \text{ mg kg}^{-1}$ and $13.070 \text{ mg kg}^{-1}$ dry weight, substantially exceeding the international WHO/FAO benchmark (0.30 mg kg^{-1}) and the national Indian FSSAI statutory threshold (2.50 mg kg^{-1}). Cadmium exhibited high soil-to-plant transfer efficiency, yielding Bioaccumulation Factors (BAF) greater than unity (5.83--8.09) in all effluent-irrigated crops. In contrast, foliar metal levels in organic control treatments remained below the solid-phase Limit of Quantification for Cd ($< 0.25 \text{ mg kg}^{-1}$) and within safe baselines for Pb and Cr. Human dietary risk assessments yielded Target Hazard Quotients (THQ) of 20.57 for Pb and 13.94 for Cd in *S. oleracea*, demonstrating serious non-carcinogenic health hazards through the food chain. These findings confirm that untreated textile wastewater serves as a potent vector for heavy metal accumulation in leafy produce, while vermicompost-based organic management mitigates toxic metal entry into the agricultural food chain.

Keywords: Atomic Absorption Spectroscopy (AAS), Heavy Metal Bioaccumulation, Cadmium, Lead, Bioaccumulation Factor, Target Hazard Quotient, Bhilwara

INTRODUCTION

Agricultural intensification across semi-arid urbanizing regions increasingly encounters severe environmental constraints linked to industrial wastewater reuse. The textile manufacturing sector represents a major consumer of freshwater and a primary producer of chemically complex wastewater containing refractory dyestuffs, finishing reagents, dissolved salts, and toxic transition metals (Al-Tohamy et al., 2022; Bhagat & Tiyyasha, 2013). In Bhilwara district, recognized

as the "Textile City of Rajasthan," more than 500 wet-processing, dyeing, and rotary printing establishments discharge 20 to 30 million liters per day (MLD) of industrial wastewater into municipal drainage networks like the Guwardi Drain and ephemeral riverbeds of the Kothari and Banas basins (Dhaker & Mehta, 2020; Pareek & Srivastava, 2025; Sharma, 2024; Singh et al., 2025). Under conditions of chronic freshwater scarcity, peri-urban farmers frequently divert these wastewater-impacted surface flows to

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irrigate food and fodder crops (Dhaker et al., 2025; Kaur et al., 2019).

Heavy metals such as cadmium (*Cd*), chromium (*Cr*), and lead (*Pb*) are non-biodegradable and persist in the agro-ecosystem (Atta et al., 2023; Hassan et al., 2024; Khaliq et al., 2022; Waheed et al., 2019). Continuous application of metal-laden wastewater promotes the translocation of free divalent cations and organic complexes across root membranes into edible vegetative organs (Bonanno et al., 2016; Souri et al., 2019). Broad-leaved succulent vegetables (e.g., *Spinacia oleracea*) and perennial aromatic herbs (e.g., *Mentha spicata*) possess high transpiration rates and large root-surface-area-to-biomass ratios, predisposing them to elevated metal uptake compared to fibrous-rooted monocotyledonous grasses (Bonanno et al., 2016; Souri et al., 2019). Ingestion of produce contaminated with non-essential metals like *Pb* and *Cd* poses well-documented chronic health hazards, including nephrotoxicity, neurotoxicity, skeletal demineralization, and cardiovascular impairment (Alsafran et al., 2021; Ojekunle et al., 2022; Sultana et al., 2022).

Conversely, restorative organic soil management utilizing composted bio-fertilizers and vermicompost introduces stabilized humic and fulvic substances capable of chelating transition metals on functional carboxyl and phenolic surfaces, immobilizing them within the soil matrix and reducing phytoavailability (Aslam et al., 2019; Duddigan et al., 2023; Oyege & Bhaskar, 2023; Xu & Mou, 2016). To establish the environmental and health impacts of these contrasting agronomic practices, this paper presents a detailed flame atomic absorption spectroscopic (AAS) investigation evaluating the uptake, soil-to-plant transfer kinetics (*BAF*), and dietary health risks (*EDI*, *THQ*) of *Cd*, *Cr*, and *Pb* in four economically important crops under field conditions in Bhilwara, Rajasthan.

MATERIALS AND METHODS

2.1 Study Area, Experimental Design, and Treatment Matrix

The field experiment was performed at the experimental farm of Krishi Vigyan Kendra (KVK), Agarpura Chouraha, Bhilwara (25.345132° N, 74.717118° E), situated in the semi-arid agro-climatic

Zone IV-A of Rajasthan. The soil was an uncultivated clay loam with no prior history of synthetic fertilizer or industrial wastewater application.

The trial employed a $3 \times 4 \times 3$ completely randomized block design yielding 36 experimental plots ($6\text{ ft} \times 8\text{ ft}$ each; 4.46 m^2). Four economically and medicinally important crops were cultivated:

- Wheatgrass (*Triticum aestivum* L., var. Raj-4238, Poaceae)
- Fenugreek (*Trigonella foenum-graecum* L., var. PRM-45, Fabaceae)
- Spinach (*Spinacia oleracea* L., Amaranthaceae)
- Spearmint (*Mentha spicata* L., Lamiaceae)

Treatments comprised:

- **Control Series (C1–C4):** Cultivated in unamended baseline soil and irrigated with uncontaminated KVK borewell groundwater.
- **Organic Series (OS1–OS4):** Cultivated in baseline soil amended with well-decomposed vermicompost (5 kg/plot basal application, with scheduled split doses) and irrigated with KVK borewell groundwater.
- **Textile Effluent Series (TS1–TS4):** Cultivated in unamended baseline soil and irrigated with composite industrial textile effluent (TW) collected from four strategic discharge locations (Kothari River Sites 1 & 2, Guwardi Drain, and Banas River catchment) in 200 L HDPE drums.

2.2 Sample Digestion Protocols and Dilution Formulations

Sample preparation and wet acid digestions followed standardized analytical protocols:

- **Solid Plant Foliar Matrices (C1–C4, OS1–OS4, TS1–TS4):** Harvested leaves were rinsed thoroughly with tap water and double-deionized water, shade-dried to constant weight, and pulverized into a fine powder. Exactly 10.0 g of tissue powder was predigested overnight with 10 mL concentrated analytical-grade HNO_3 (70%), digested with 1.0 mL HClO_4 on a water bath for 3 hours until transparent, filtered through Whatman No. 1 paper, and made up to 100 mL (0.100 L) with double-distilled water:

$$\text{FoliarMetal}(\text{mgk g}^{-1} \text{ drywt.}) = C_{\text{inst}}(\text{mg L}^{-1}) \times \frac{0.100 \text{ L}}{0.010 \text{ kg}} = C_{\text{inst}} \times 10$$

- **Pre-Cultivation Baseline Soil Matrix (SS):** 1.0 g of air-dried, sieved (<2 mm) soil was digested in 12 mL aqua regia (3 HCl:1 HNO₃) at

70–80 °C, filtered through Whatman No. 42 paper, and diluted to 50 mL (0.050 L):

$$\text{SoilMetal}(\text{mgk g}^{-1} \text{ drywt.}) = C_{\text{inst}}(\text{mg L}^{-1}) \times \frac{0.050 \text{ L}}{0.001 \text{ kg}} = C_{\text{inst}} \times 50$$

- **Liquid Textile Wastewater (TW):** 100 mL of acidified wastewater was digested with 5 mL concentrated HNO₃ to near-dryness and reconstituted to 100 mL, yielding a direct conversion factor:

$$\text{WastewaterMetal}(\text{mg L}^{-1}) = C_{\text{inst}} \times 1.0$$

2.3 Spectroscopic Instrumentation and Quality Control

Heavy metal determinations (Cd, Cr, Pb) were executed on a PG Instruments AA500 Atomic Absorption Spectrophotometer equipped with element-specific hollow-cathode lamps and deuterium background correction at the Materials Research Centre (MRC), MNIT Jaipur. Calibration curves were constructed from multi-point certified elemental standards (0.0--10.0 mg L⁻¹, $r^2 \geq 0.998$). Solid-phase Limits of Quantification (LOQ) were established as:

- Cadmium (Cd): 0.25 mgk g⁻¹
- Chromium (Cr): 1.00 mgk g⁻¹
- Lead (Pb): 1.00 mgk g⁻¹

Values falling below these thresholds are designated as Below Limit of Quantification (BLQ).

2.4 Bioaccumulation and Dietary Risk Assessment Formulations

Bioaccumulation Factor (BAF): Quantifies transfer efficiency from soil to foliar biomass:

$$BAF = \frac{C_{\text{plant}}(\text{mgk g}^{-1} \text{ drywt.})}{C_{\text{soil}}(\text{mgk g}^{-1} \text{ drywt.})}$$

1. where C_{soil} represents baseline soil concentrations:

Cd 0.350 mgk g⁻¹, Cr 6.050 mgk g⁻¹, and Pb 7.650 mgk g⁻¹.

Estimated Daily Intake (EDI):

$$EDI = \frac{C_{\text{plant}} \times IR \times EF \times ED}{BW \times AT}$$

2. where IR is daily adult intake of leafy greens (0.345 kg person⁻¹ day⁻¹); EF is exposure frequency (365 days year⁻¹); ED is exposure duration (70 years); BW is average adult body weight (70 kg); and AT is averaging time for non-carcinogens (25,550 days).

$$\text{Target Hazard Quotient (THQ): } THQ = \frac{EDI}{RfD}$$

3. where oral reference doses (RfD) are 0.0035 mgk g⁻¹ day⁻¹ for Pb, 0.001 mgk g⁻¹ day⁻¹ for Cd, and 0.003 mgk g⁻¹ day⁻¹ for Cr (US EPA). A $THQ > 1.0$ indicates potential non-carcinogenic health hazards.

RESULTS AND DISCUSSION

3.1 Trace Heavy Metal Burden in Irrigation Matrices and Baseline Soil

The composite textile wastewater (TW) carried high heavy metal concentrations: Pb (4.332 mg L⁻¹),

Cr (3.388 mg L⁻¹), and Cd (1.492 mg L⁻¹). These levels substantially exceeded the Indian Central Pollution Control Board (CPCB) and Bureau of Indian Standards (BIS: IS 2296) irrigation limits (0.10 mg L⁻¹ for Pb, 0.10 mg L⁻¹ for Cr, and 0.01 mg L⁻¹ for Cd) by factors of 43.3 ×, 33.9 ×, and 149.2 ×, respectively. The metal burden reflects the widespread use of lead chromate pigments, potassium dichromate oxidizing agents, and cadmium-stabilized complex azo dyestuffs in Bhilwara textile units (Dagne, 2020; Meena & Meena, 2020; Odipe et al., 2019). In contrast, local groundwater used for the

control and organic treatments exhibited non-detectable metal levels ($<0.001 \text{ mg L}^{-1}$).

AAS instrumental readouts and calculated concentrations are compiled in Table 1.

Table 1: AAS Instrumental Parameters, Raw Absorbances, Dilution Factors, and Converted Elemental Concentrations *Values with an asterisk fall below the established Limit of Quantification (LOQ: Cd = 0.25 mg/kg ; Cr = 1.00 mg/kg ; Pb = 1.00 mg/kg).

Sample Code	Matrix Description	Cd Inst. (ppm)	Cd Converted (mg/kg)	Cr Inst. (ppm)	Cr Converted (mg/kg)	Pb Inst. (ppm)	Pb Converted (mg/kg)	QA/QC Status
TW	Textile Effluent	1.492	1.492 mg/L	3.388	3.388 mg/L	4.332	4.332 mg/L	>Permissible Standard
SS	Baseline Soil	0.007	0.350 mg/kg	0.121	6.050 mg/kg	0.153	7.650 mg/kg	Baseline Soil Burden
C1	Wheatgrass (Control)	0.013	0.130 mg/kg*	0.121	1.210 mg/kg	0.153	1.530 mg/kg	Baseline Trace Level
OS1	Wheatgrass (Organic)	0.003	0.030 mg/kg*	0.121	1.210 mg/kg	0.076	0.760 mg/kg*	Below LOQ Threshold
TS1	Wheatgrass (Textile)	0.216	2.160 mg/kg	0.344	3.440 mg/kg	0.538	5.380 mg/kg	Bioaccumulated
C2	Fenugreek (Control)	0.010	0.100 mg/kg*	0.121	1.210 mg/kg	0.076	0.760 mg/kg*	Below LOQ Threshold
OS2	Fenugreek (Organic)	0.003	0.030 mg/kg*	0.121	1.210 mg/kg	0.076	0.760 mg/kg*	Below LOQ Threshold
TS2	Fenugreek (Textile)	0.204	2.040 mg/kg	0.269	2.690 mg/kg	0.923	9.230 mg/kg	Bioaccumulated
C3	Spinach (Control)	0.010	0.100 mg/kg*	0.195	1.950 mg/kg	0.076	0.760 mg/kg*	Below LOQ Threshold
OS3	Spinach (Organic)	0.010	0.100 mg/kg*	0.121	1.210 mg/kg	0.153	1.530 mg/kg	Baseline Trace Level
TS3	Spinach (Textile)	0.283	2.830 mg/kg	0.492	4.920 mg/kg	1.461	14.610 mg/kg	Hyper-accumulator
C4	Spearmint (Control)	0.007	0.070 mg/kg*	0.195	1.950 mg/kg	0.153	1.530 mg/kg	Baseline Trace Level
OS4	Spearmint (Organic)	0.007	0.070 mg/kg*	0.195	1.950 mg/kg	0.230	2.300 mg/kg	Baseline Trace Level
TS4	Spearmint (Textile)	0.246	2.460 mg/kg	0.492	4.920 mg/kg	1.307	13.070 mg/kg	Hyper-accumulator

3.2 Foliar Heavy Metal Bioaccumulation Across Botanical Taxa

In foliar biomass harvested from effluent plots (TS1–TS4), trace metal accumulation was significantly elevated over controls, following the species order:

Spinach (TS3) $\approx$ *Spearmint* (TS4) $>$ *Fenugreek* (TS2)

- **Lead Accumulation:** Foliar Pb reached $14.610 \text{ mg kg}^{-1}$ dry weight in spinach (TS3) and $13.070 \text{ mg kg}^{-1}$ in spearmint (TS4). Relative to organic controls (OS1–OS4: 0.760 – 2.300 mg kg^{-1}), effluent irrigation increased foliar lead by $7.1 \times$ in wheatgrass,

$12.1 \times$ in fenugreek, $9.5 \times$ in spinach, and $5.7 \times$ in spearmint.

- **Chromium Dynamics:** Tissue Cr in TS3 and TS4 reached 4.920 mg kg^{-1} dry weight, higher than wheatgrass (3.440 mg kg^{-1}) and fenugreek (2.690 mg kg^{-1}).
- **Cadmium Phytoavailability:** Cadmium accumulated across all effluent-treated crops, reaching 2.830 mg kg^{-1} in spinach and 2.460 mg kg^{-1} in spearmint, compared to 2.160 mg kg^{-1} in wheatgrass and 2.040 mg kg^{-1} in fenugreek. In organic

plots (OS1–OS4), foliar Cd was consistently below the LOQ threshold ($<0.25 \text{ mg kg}^{-1}$).

The observed accumulation differences reflect plant anatomical characteristics. Broad-leaved dicots (*S. oleracea*) and perennial herbs (*M. spicata*) maintain higher stomatal conductance and transpiration rates, driving substantial mass-flow xylem transport of divalent cations into foliar tissues (Bonanno et al., 2016; Souri et al., 2019). Conversely, monocotyledonous wheatgrass (*T. aestivum*) develops an endodermal Casparian strip with suberized secondary thickenings that restricts apoplastic radial ion flux into the stele, limiting heavy metal translocation to above-ground parts (Bonanno et al., 2016; Souri et al., 2019).

3.3 Soil-to-Plant Partitioning and Bioaccumulation Factors (BAF)

Soil-to-shoot transfer efficiencies were determined against pre-cultivation baseline soil concentrations (Table 2).

Cadmium exhibited Bioaccumulation Factors well above unity ($BAF = 5.83 - 8.09$) across all effluent-irrigated crops. The high mobility of Cd^{2+} stems from its chemical resemblance to divalent cations (Ca^{2+} , Fe^{2+}), facilitating entry into root symplasts via ZIP-family permeases and non-selective cation channels (Bankole et al., 2024; Orosun et al., 2020). Under sodic conditions ($SAR = 15.43$), high Na^+ loads displace Cd^{2+} from soil exchange sites into the soil solution, enhancing bioaccumulation (Bankole et al., 2024; Siddique et al., 2021).

Lead concentrated preferentially in spinach ($BAF = 1.91$) and spearmint ($BAF = 1.71$). Conversely, chromium exhibited $BAF < 1.0$ ($0.44 - 0.81$) in all taxa. In well-aerated alkaline soils ($pH 8.5 - 9.0$), soluble hexavalent chromate reduces to trivalent chromium (Cr^{3+}), precipitating as insoluble chromium hydroxide [$Cr(OH)_3$] or binding to mineral lattices, moderating translocation into edible foliage (Bankole et al., 2024; Chen & Lena, 2001; Gaudino et al., 2007).

Table 2: Soil-to-Shoot Bioaccumulation Factors (BAF) and Regulatory Limit Comparisons

Crop Taxon	Sample Code & Regime	Tissue Cd (mg/kg)	BAF-Cd	Tissue Cr (mg/kg)	BAF-Cr	Tissue Pb (mg/kg)	BAF-Pb
<i>Triticum aestivum</i> L.	TS1 (Effluent)	2.160	6.17	3.440	0.57	5.380	0.70
<i>Trigonella foenum-graecum</i> L.	TS2 (Effluent)	2.040	5.83	2.690	0.44	9.230	1.21
<i>Spinacia oleracea</i> L.	TS3 (Effluent)	2.830	8.09	4.920	0.81	14.610	1.91
<i>Mentha spicata</i> L.	TS4 (Effluent)	2.460	7.03	4.920	0.81	13.070	1.71
All Crops Baseline	OS1–OS4 (Organic)	< 0.25 (BLQ)	—	1.210–1.950	0.20–0.32	< 1.00 –2.300	0.10–0.30
WHO/FAO Permissible Limit	Edible Vegetable Benchmark	0.20 mg/kg	—	2.30 mg/kg	—	0.30 mg/kg	—
Indian FSSAI Limit	National Food Safety Standard	1.50 mg/kg	—	20.00 mg/kg	—	2.50 mg/kg	—

3.4 Dietary Human Health Risk Assessment (EDI and THQ)

Dietary exposure metrics and non-carcinogenic health hazards calculated for adult consumers are summarized in Table 3.

Table 3: Dietary Exposure (EDI) and Non-Carcinogenic Target Hazard Quotients (THQ) for Adult Consumers

Exposure Scenario	Cd EDI (mg/kg/day)	THQ-Cd	Pb EDI (mg/kg/day)	THQ-Pb	Cumulative Hazard Index (HI = Σ THQ)
TS1 Wheatgrass (<i>T. aestivum</i>)	0.0106	10.64	0.0265	7.57	18.21
TS2 Fenugreek (<i>T. foenum-graecum</i>)	0.0100	10.05	0.0455	12.99	23.04
TS3 Spinach (<i>S. oleracea</i>)	0.0139	13.94	0.0720	20.57	34.51
TS4 Spearmint (<i>M. spicata</i>)	0.0121	12.12	0.0644	18.40	30.52
Safe Toxicological Threshold	—	≤ 1.0	—	≤ 1.0	≤ 1.0 (No Significant Non-Carcinogenic Risk)

Target Hazard Quotients for both lead and cadmium exceeded the safe threshold ($THQ > 1.0$) across all crops receiving untreated textile wastewater. In *Spinacia oleracea*, $THQ-Pb$ reached 20.57 and $THQ-Cd$ reached 13.94, yielding a cumulative Hazard Index of 34.51. In *Mentha spicata*, the cumulative Hazard Index was 30.52. Because leafy vegetables are dietary staples consumed with minimal processing, irrigating peri-urban fields with untreated textile wastewater presents potential food-chain contamination risks for local populations (Alsafran et al., 2021; Ojekunle et al., 2022; Sultana et al., 2022).

CONCLUSION

Flame atomic absorption spectrophotometry confirmed that composite textile industrial wastewater in Bhilwara contains high concentrations of toxic heavy metals ($Pb\ 4.332\ mg\ L^{-1}$, $Cr\ 3.388\ mg\ L^{-1}$, $Cd\ 1.492\ mg\ L^{-1}$), substantially exceeding permissible discharge and agricultural reuse limits. Irrigation with this wastewater led to significant metal accumulation in edible foliar biomass, following the hierarchy:

Spinach $\approx$ *Spearmint* $>$ *Fenugreek* $>$ *Wheatgrass*.

Foliar lead in *S. oleracea* reached $14.610\ mg\ kg^{-1}$ dry weight, exceeding the WHO/FAO permissible food safety threshold by nearly 48-fold and yielding a Target Hazard Quotient (THQ) of 20.57. Cadmium demonstrated net soil-to-plant bioaccumulation ($BAF\ 5.83--8.09$) in all effluent-irrigated crops. In contrast, vermicompost-amended organic management maintained foliar cadmium levels below the

analytical quantification limit ($< 0.25\ mg\ kg^{-1}$) and kept lead and chromium within safe baselines. These findings underscore the importance of enforcing industrial effluent treatment standards in the Kothari and Banas river basins and support the adoption of organic soil management and appropriate crop choices to reduce heavy metal entry into peri-urban food systems.

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