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Growth and Metal Uptake Dynamics of *Brassica juncea* in Cadmium and Arsenic Contaminated Soil Irrigated with Industrial Effluent

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Abstract

Brassica juncea (*B. juncea*) is widely recognized for its potential in phytoremediation, yet the combined effects of cadmium (Cd) and arsenic (As) in soil and irrigation water on its growth and metal uptake remain insufficiently explored. This study investigated the germination, growth performance, and Cd–As accumulation of *B. juncea* grown under four combinations of clean and contaminated soil and water using a completely randomized pot experiment. Treatments comprised: control soil with clean water (T1), contaminated soil with clean water (T2), control soil with industrial effluent (T3), and contaminated soil with industrial effluent (T4). Physicochemical characterization confirmed substantially elevated Cd and As levels in industrial soil and wastewater compared with control soil and tap water. Germination percentage decreased from 96% in T1 to 68% in T4, with intermediate reductions in T2 and T3, indicating that both soil and water-borne metals impair seed viability, and their combination exerts the strongest inhibition. Growth traits, including leaf number, plant height, root length, and shoot length, followed the same declining trend from T1 to T4, reflecting pronounced metal-induced stress under dual contamination. Plants grown in contaminated treatments accumulated significantly higher Cd and As concentrations in their tissues than the control, with maximum accumulation under T4, confirming efficient metal uptake under polluted conditions. Overall, the findings demonstrate that while Cd- and As-rich industrial effluents severely reduce the growth and vigor of *B. juncea*, the species maintains sufficient tolerance and uptake capacity to be considered a promising candidate for phytoremediation of industrially contaminated soils and wastewater-irrigated sites.

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Introduction

Industries are the backbone of the country and serve as a fundamental catalyst for economic stability; however, it also constitutes a primary contributor to environmental degradation [1]. The environment has since received progressive toxic compounds, which is a great concern. Heavy metal pollution is one of the most consistent and acute environmental problems from industrial wastes. Unlike organic pollutants, heavy metals, particularly lead (Pb), cadmium (Cd), mercury (Hg), arsenic (As), and chromium (Cr), are non-biodegradable, extremely persistent, and capable of bioaccumulation in the ecosystem [2]. Their continuous accumulation in soils poses significant risks to soil fertility, food safety, and ecosystem health, and thus needs a sustainable and cost-effective remediation approach [3]. Cadmium (Cd) and arsenic (As) are two heavy metals that are of the most important concern because of their high toxicity, widespread prevalence, and harmful effects [4].

Conventional techniques for removing pollutants from wastewater, like soil excavation and chemical precipitation, are expensive, ineffective, and harmful to the environment. The process of using plants to remove pollutants from the environment, known as phytoremediation, has drawn more attention as a viable and affordable option [5]. The employment of green plants to eliminate, transfer, stabilize, or destroy pollutants in soil and water is known as phytoremediation [6]. Among various hyperaccumulator species, *Brassica juncea* (Indian mustard) is prominent due to its rapid growth, high biomass, and good capacity to uptake heavy metals [7]. Its robust nature and adaptability to various soil conditions make it a suitable species for remediation efforts in diverse environments. However, the efficacy of *B. juncea* in real-world, multi-contaminant scenarios, particularly under the stress of complex irrigation sources like industrial effluents, remains insufficiently characterized [8]. The majority of the studies have been done with single-metal pollution using a controlled laboratory environment or by applying different synthetic wastewater, and are limiting knowing the performance in a more realistic natural environment. Additionally, plant growth and metal uptake dynamics over time are important for the development of phytoextraction. The uptake kinetics, the rate, time, and amount of metal accumulation in different plant parts (roots and shoots) during the growth cycle determine the time to harvest for effective removal.

The study aims to assess the phytoremediation ability of *B juncea* in a heavy metal-contaminated soil. *B juncea* was selected with respect to its cadmium and arsenic uptake and growth performance. Through a careful examination of plant growth responses and heavy metal accumulation, this study provides practical guidelines for the implementation of Indian mustard in contaminated agroecosystems as a sustainable choice for remediation. The findings are expected to contribute not only to the advancement of phytoremediation science but also to the development of practical strategies for managing heavy metal pollution in industrially impacted regions.

Materials and Methods

Experimental design and study area

The experiment was conducted on the rooftop of the Department of Environmental Science, Bahauddin Zakariya University (BZU), Multan, Pakistan, under natural light and ambient conditions. Multan has a hot semi-arid climate with very hot summers and cool winters. A completely randomized design (CRD) was employed with four treatments (T1–T4), each replicated five times (20 pots in total). Plastic pots (10 × 11 cm) lined with polyethylene bags were filled with 1 kg of prepared soil to minimize leaching and cross-contamination. Before filling, collected soils were air-dried under direct sunlight for 48 hours on the rooftop. The dried soil was then sieved through a 2 mm mesh to remove debris and ensure homogeneity. Industrial wastewater was collected from an outlet discharging untreated effluents from textile, dyeing, and manufacturing units. Contaminated industrial soil was sampled from fields irrigated with this effluent near the discharge point, while uncontaminated control soil was taken from fields irrigated only with clean canal water. Control water for irrigation was obtained from the departmental tap supply at BZU. Immediately after collection, pH, electrical conductivity (EC), total dissolved solids (TDS), organic matter (OM), texture, color, and odor were determined following standard procedures [9], and Cd and As were analyzed by atomic absorption spectrophotometry (AAS). The treatments used were designed as follows: T1, control soil with clean water; T2, contaminated soil with clean water; T3, control soil with industrial wastewater; and T4, contaminated soil with industrial wastewater.

Plant selection and growth conditions

Mustard (*Brassica juncea*) was selected due to its high biomass, rapid growth, and reported capacity to hyperaccumulate metals in roots and shoots [10]. Ten seeds were sown per pot; after germination, seedlings were thinned to five healthy plants per pot, giving five replicate pots per treatment. Plants were grown for approximately 60 days on the rooftop under natural light and ambient temperature. Pots in T1 and T2 were irrigated with control water, while those in T3 and T4 received industrial wastewater regularly to maintain adequate soil moisture without causing waterlogging.

Sample preparation

At the end of the growth period, plants were harvested after recording final plant height to assess biomass and prepare samples for heavy metal analysis. Fresh weights of roots and shoots were recorded immediately after harvesting, followed by oven drying at 55 ± 2°C until constant weight was achieved to obtain dry biomass. The dried plant materials were ground into fine powder and digested using a HNO_3:$HClO_4$ (3:1) wet digestion method. Concentrations of Cd and As in plant tissues were determined using Atomic Absorption Spectrophotometry (AAS).

Statistical analysis

Microsoft Excel was used to record initial data, and SPSS software was used for analysis.

Results and Discussion

Germination rate

The germination rate of *B. juncea* was evaluated to understand the impact of contamination on seed and plant development under treatments T1–T4. T1 had the highest germination rate of 96%, whereas T4 had the lowest germination rate of 68%, where both soil and water were contaminated. Intermediate values were observed for T2 (86%) and T3 (78%), respectively (Fig. 1). The data show that contamination, either in soil, water, or both, has a detrimental effect on the germination. The germination percentage followed the trend: T1 (96%) > T2 (86%) > T3 (78%) > T4 (68%). The declining trend in T1 to T4 highlights the acute toxicity, which is likely to disrupt water uptake and enzymatic activity. Research by Adrees et al. [11] demonstrated that metal-laden water directly damages seed membranes. The poorest performance in T4 (68%), a synergistic stress effect occurs, where metal interactions in soil and water collectively suppress metabolic processes. This is consistent with research by Rizwan et al. [12], who found that dual contamination increases oxidative stress and reduces seed reserves required for germination.

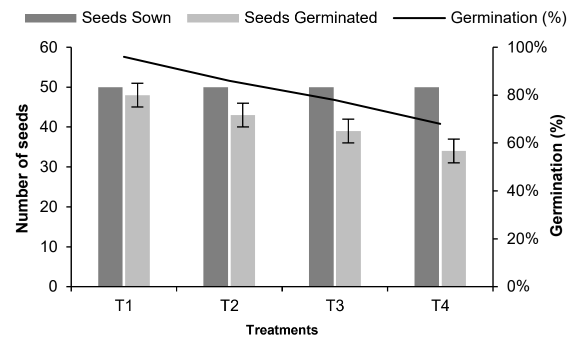

Fig. 1 Germination performance of mustard (*Brassica juncea*) seeds under different treatments (T1–T4).

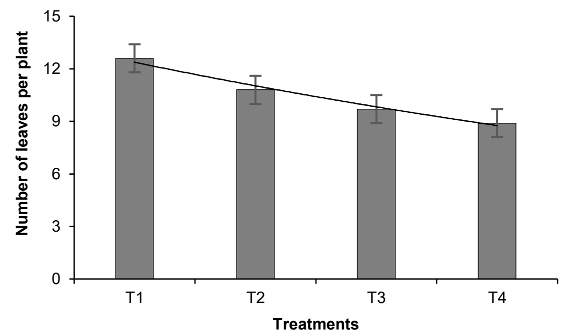

Fig. 2 Effect of different treatments (T1–T4) on the number of leaves per mustard (*Brassica juncea*) plant.

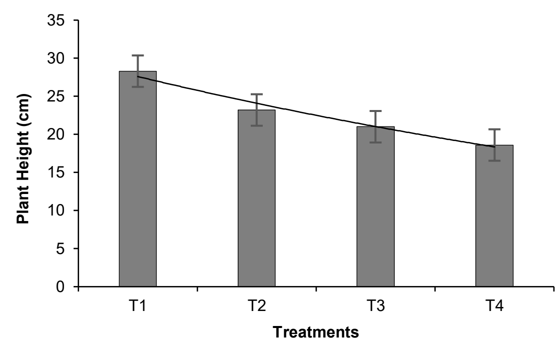

Fig. 3 Effect of different treatments on plant height of mustard (*Brassica juncea*).

Growth parameters

The growth parameters of *B. juncea* vary significantly across treatments, that demonstrate the detrimental effects of heavy metal contamination on plant development. The number of leaves in *B.*

juncea decreased steadily from T1 to T4, indicating progressive growth inhibition under contamination (Fig. 2). A gradual decline in leaf numbers from T1 to T4 indicates a dose-dependent response, where the most contaminated treatment (T4) caused roughly a one-third reduction in leaf production relative to T1. Increasing metal load negatively affected canopy development and photosynthetically active surface area.

Plant height of *B. juncea* decreased markedly with increasing contamination and effluent stress. Fig. 3 shows that plants in T1 achieved the greatest mean height (28 cm), whereas T2, T3, and T4 reached about 23, 21, and 19 cm, respectively. This pattern demonstrates that combined Cd–As contamination and industrial effluent irrigation severely constrained vertical growth relative to the control.

Root length was significantly reduced from 14.5 ± 1.8 cm (T1) to 9.5 ± 1.2 cm (T4). The intermediate reductions in T2 and T3 were 17% and 22%, respectively (Fig. 4). With the highest value in T1 (13.8 ± 2.0 cm) and the lowest in T4 (9.1 ± 1.0 cm), shoot length showed a similar drop. Treatments T2, T3, and T4 showed reductions of 20%, 30%, and 34%, respectively (Fig. 5).

The pronounced decline under Cd and As stress exerts an inhibitory effect on *B. juncea* growth, where heavy metals reduced plant growth by impairing photosynthetic efficiency, nutrient uptake, and turgor-driven cell expansion. Cadmium and arsenic interfere with root function and disrupt water relations, leading to reduced xylem transport of assimilates and hormones such as gibberellins that regulate stem elongation, ultimately shortening plants. Overall, the results indicate that contamination in either the soil or water medium negatively affects plant growth, but the most profound effect is seen when both media are polluted simultaneously.

Fresh and dry biomass of mustard plants

The effect of soil and water contamination on total plant productivity was evaluated by measuring the fresh and dry biomass of mustard plants. Under heavy metal contamination, the fresh biomass of *B. juncea* roots and shoots showed notable decreases (Fig. 6). Fresh shoot biomass (6.5 ± 0.8 g) and fresh root biomass (3.2 ± 0.4 g) were highest in T1. The biomass of fresh shoot and fresh root dropped by 20% and 16% in T2, 29% and 31% in T3, 42% and 41% in T4 treatments, respectively.

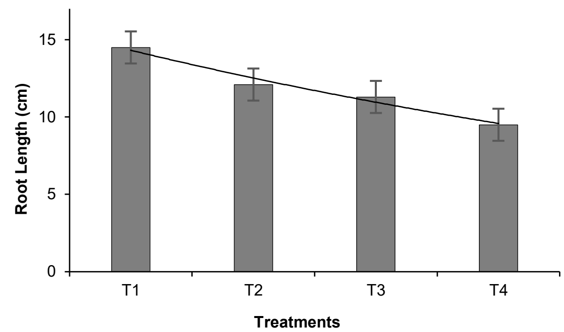

Fig. 4 Effect of different treatments on root length of mustard (*Brassica juncea*) plants.

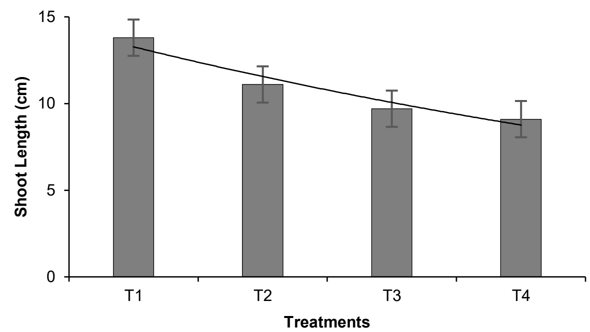

Fig. 5 Effect of different treatments on shoot length of mustard (*Brassica juncea*) plants.

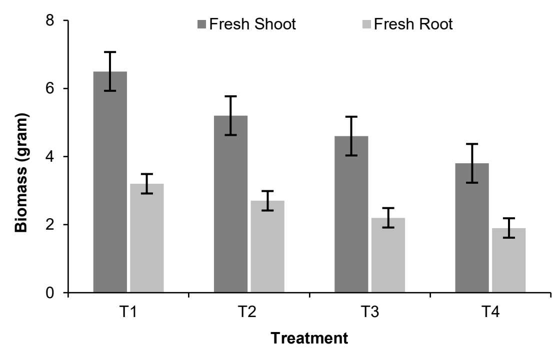

Fig. 6 Fresh biomass of roots and shoots of mustard plants under different treatments.

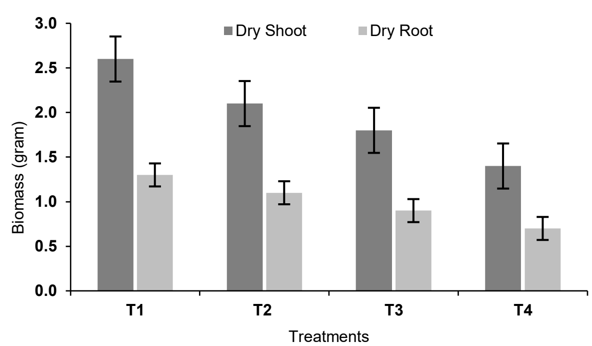

Fig. 7 Dry biomass of roots and shoots of mustard plants under different treatments.

Table 1 Cadmium (Cd) and Arsenic (As) concentrations (mg/kg) in roots and shoots of mustard plants (*Brassica juncea*) under different treatments.

Treatments	Roots		Shoots	
	Cd	As	Cd	As
T1	0.12 ± 0.02[d]	0.05 ± 0.01[d]	0.08 ± 0.01[d]	0.02 ± 0.00[d]
T2	1.32 ± 0.11[c]	1.01 ± 0.09[c]	0.85 ± 0.09[c]	0.64 ± 0.06[c]
T3	1.56 ± 0.13[b]	1.28 ± 0.11[b]	0.96 ± 0.10[b]	0.73 ± 0.07[b]
T4	2.45 ± 0.19[a]	2.08 ± 0.17[a]	1.62 ± 0.14[a]	1.19 ± 0.10[a]

The fact that fresh shoots' biomass was reduced more than roots in T3 indicates that metals are quickly transferred to aerial tissues, where they hinder cell growth and photosynthetic efficiency. Cd and As can disrupt chloroplast function and reduce carbon assimilation. The fresh root biomass decline was less severe in T2 than in T3, indicating that soil-bound metals may be partially immobilized, limiting direct root damage. However, T4 showed the most severe suppression, indicating that exposure to synergistic metals increases biomass loss, most likely as a result of compounded oxidative stress and inhibition of food uptake.

The dry biomass revealed a pattern that matched the fresh biomass data, with T1 having the highest dry shoot weight (2.6 ± 0.3 g) and T4 having the lowest (1.4 ± 0.1 g). Similarly, dry root biomass dropped from 1.3 ± 0.2 g in T1 to 0.7 ± 0.1 g in T4 (Fig. 7). Dry biomass represents the actual organic matter accumulation and structural growth. The higher dry weight in T1 suggests enhanced photosynthetic efficiency and better overall plant vigor under this treatment. The biomass reduction patterns observed clearly depict the cumulative stress effects induced by heavy metal contamination on mustard plants. T2 has decreased 20-25% biomass when compared to the control, which is consistent with an observation made by Thakur et al. [13], wherein it was reported that soil-bound metals primarily affect root development and nutrient uptake efficiency. The more pronounced reduction in T3 (29-34% decrease) validates the emerging hypothesis from Page and Feller [14] that water-soluble metals would be absorbed more readily by plant tissues across which they are distributed and hence pose broader physiological disruptions [15].

Heavy metal uptake in plant tissues

The concentrations of cadmium (Cd) and arsenic (As) in the root and shoot tissues were analyzed to evaluate the uptake and translocation potential (Table 1). Cadmium and arsenic concentrations were minimal in T1, suggesting little background contamination. Both metals showed significantly higher accumulation in T2, T3, and especially T4, with greater accumulation in roots compared to shoots across all treatments. *B. juncea*'s ability to absorb metals is demonstrated by the progressive metal buildup across treatments, which also reveals significant bioavailability patterns. The results of Ashraf et al. [16] about the potential for metal buildup in mustard are supported by the 11–20 times increase in root Cd concentrations. However, the consistently higher Cd uptake compared to As (despite similar treatment conditions) supports the model of Kaur et al. [17] that Cd's chemical similarity to essential nutrients like Zn facilitates its absorption through shared transport systems. Higher accumulation of metals in T3 compared to T2 (18% for Cd and 27% for As) indicates that the dissolved form of metal in water could be more bioavailable to plants than soil-bound forms [12]. The synergistic accumulation in T4 suggests that combined contamination creates additive exposure pathways [18]. The maintained root: shoot concentration ratios across treatments suggest *B. juncea* employs consistent metal sequestration strategies regardless of exposure route, possibly through constitutive expression of metal-binding proteins [19].

Conclusion

The threat to human health and the ecosystem from heavy metal pollution is becoming an important global issue. The study was carried out to assess the phytoremediation potential of arsenic (As) and cadmium (Cd). The parameters such as growth, biomass production, and metal accumulation were used. The results show that the contaminant significantly reduced germination, growth, and biomass of *B. juncea*. However, *B juncea* was able to endure and effectively accumulate Cd and As. Despite limited metal movement to shoots, bioaccumulation and translocation patterns further supported mustard's suitability as an accumulator species. Overall, *B juncea* proves to be a cost-effective, economical, and environmentally sound choice for phytoremediation. Its fast growth rate, tolerance to stress, and ability to accumulate metals make it also suitable for large-scale remediation.

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Conflict of Interest

The authors had no conflicts of interest to disclose.

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