

Improving beetroot growth and heavy metal tolerance with biochar under wastewater irrigation across multiple sites

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ABSTRACT

Wastewater containing heavy metals can lead to soil and crop accumulation, thus posing a risk to the environment. Biochar, being a rich carbon-containing organic fertilizer, can reduce these adverse impacts by increasing soil fertility. This study was conducted to assess the biochar-treated wastewater for beetroot cultivation using *Beta vulgaris* L. at three locations in Faisalabad, namely, Directorate of Farms UAF, Uchkera Farm, and Satyana Farm, Faisalabad, using an RCBD factorial design with three wastewater treatments, namely, UAF sewage, Madhuana, and Paharang drains. As a result, pigments increased (Chl a/b 25%, carotenoids 25%), and physiological characteristics improved (RWC 25%, electrolyte leakage -20%). The elevated levels of heavy metals were lowered by biochar. Heavy metal buildup in beets was significantly enhanced by wastewater irrigation, but most metals were successfully reduced by applying biochar, improving crop safety and lowering environmental risk. In general, 2024 performed better than 2023. While biochar further improves performance and reduces the dangers of heavy metals, wastewater irrigation increases beetroot growth, yield, and stress tolerance, encouraging safer and more sustainable crop production.

Keywords: biochar, wastewater irrigation, heavy metal, beetroot, environmental risk.

INTRODUCTION

Water scarcity is one of the important ecological limitations that can affect global agricultural productivity and food security (Sajjad et al., 2025). In many regions, wastewater use for irrigation purposes is considered a valuable option compared to water resources (Naz et al., 2020). Wastewater is also a source of appreciable quantities of macro and micronutrients, thus a cheaper alternative for use in the production of crops. However, apart from the beneficial macro and micronutrients, there are also potentially toxic heavy metals (HMs) such as zinc, mercury, cobalt, cadmium, and iron (Shahzad et al., 2023; Nazeer et al., 2024). Across the globe, wastewater, both treated and untreated, is used to irrigate over 20 million hectares of agricultural lands, especially for the growth of vegetables, which could lead to metal contaminants in food products (Singh, 2021).

The HMs present in wastewater are known to be a major threat to plant growth and development. Higher accumulation of heavy metals has adverse effects on chlorophyll, chloroplast ultrastructure, and other physiological processes, such as photosynthesis and water relations (Nazeer et al., 2020; Mourato et al., 2015). Additionally, heavy metals cause the overproduction of ROS, resulting in oxidative stress, damage to the cell membrane, as well as reduced crop yields and quality (Małkowski et al., 2019). The transfer of these heavy metals to edible parts of the plants poses a risk to the sustainability of human health and the environment (Chowdhury et al., 2020).

Biochar, a carbon-rich product derived from the thermochemical decomposition of biomass in a limited oxygen environment, has been recognized as a viable alternative for improving soil sustainability. The high surface area of biochar and presence of certain functional groups allow for the adsorption of heavy metals, thereby

reducing their bioavailability and uptake by the plants (Glaser and Lehr, 2019). Biochar also plays a role in improving soil aggregation, nutrient retention, and fertility, thus offering a viable solution for addressing heavy metal stress caused by wastewater irrigation.

Beetroot (*Beta vulgaris* L.) of the Amaranthaceae family is globally cultivated for its edible taproot, rich in fiber, minerals, vitamins, and bioactive compounds like betalains, polyphenols, and nitrates (Ceclu and Nistor, 2020). Betalains, particularly betacyanins and betaxanthins, are responsible for the characteristic red color of beetroot and possess strong antioxidant and anti-inflammatory properties. Beetroot is nutritionally valuable but its root nature makes it prone to heavy metal accumulation under wastewater irrigation (Kazimierzczak et al., 2014). on the potential of biochar to reduce heavy metal accumulation and oxidative stress in beetroot (*Beta vulgaris* L.) grown with wastewater irrigation is limited. Moreover, the combined influence of site-specific wastewater quality and biochar amendment on beetroot growth, physiological responses, antioxidant defense mechanisms, and heavy metal accumulation under field conditions has not been thoroughly investigated. Therefore, the present study was designed to evaluate the effects of biochar application on heavy metal immobilization, plant growth, and physiological performance of beetroot under wastewater irrigation, with the ultimate goal of improving crop safety and promoting soil sustainability.

MATERIALS AND METHODS

A two-year field trial was conducted at UAF, Uchkera, and Satyana Farms, sowing seeds in early September at 8 kg ha⁻¹. Gross and net plot sizes were 10.8 m² and 9 m², respectively, with a total area of 611.6 m². Row and plant spacing were 30 cm and 10 cm, using three wastewater sources: UAF campus, Madhuana, and Paharang drains.

Physiological parameters

Relative water content

A fully developed youngest leaf from each of the plants was collected and its fresh weight recorded. All leaves were immersed in distilled water for 10 h, blotted dry, and their weights were recorded to note their turgid weight. Leaves were

dried in an oven at 70 °C for 72 h, and their dry weights were recorded. The calculation of relative water content was performed using the equation provided by (Ghoulam et al., 2002)

$$RWC (\%) = \frac{FW - DW}{TW - DW} \times 100 \quad (1)$$

Electrolyte leakage

Twenty-five leaf discs with a diameter of 0.5 cm each were floated on 15 mL of deionized water with constant shaking. Immediately, the electrolyte content in the solution was measured using a conductimeter (Hama Instruments Ltd., El-Hama, Israel) and noted as C0. After 3 h of incubation at room temperature, the electrolyte content was measured and noted as C3. Similarly, the total electrolyte content was measured by boiling the solution for 10 min and noted as TC (Rolny et al., 2011). Results were expressed as a percentage of electrolyte leakage:

$$\% EL = 100 \times \frac{(C3 - C0)}{TC} \quad (2)$$

Brix content

Brix value was measured using a refractometer (AST Model 1230, Atago Co. Ltd., Tokyo, Japan).

Photosynthetic pigments

The method of Arnon (1949) was adopted for measuring the chlorophyll content, whereas the estimation of carotenoid was done by the method of Davies (1976). A plant sample of 0.1 g was extracted with the help of 2 mL of 80% acetone, maintaining the volume up to 10 mL by using 80% acetone. The absorbance of the supernatant was determined at 663, 645, and 480 nm. The chlorophyll pigments were determined by the use of the following formula:

$$\begin{aligned} \text{Chlorophyll } a \left(\frac{mg}{g} \text{ fresh weight} \right) &= \\ &= (1.27 (OD 663) - 2.69 (OD 645)) \times V/1000 \times W \end{aligned} \quad (3)$$

$$\begin{aligned} \text{Chlorophyll } b \left(\frac{mg}{g} \text{ fresh weight} \right) &= \\ &= (22.9 (OD 645) - 4.68 (OD 663)) \times V/1000 \times W \end{aligned} \quad (4)$$

$$\begin{aligned} \text{Carotenoids} \left(\frac{mg}{g} \text{ fresh weight} \right) &= \\ &= (OD 480 + 0.114 (OD 663) - \\ &\quad - 0.638 (OD 645)) / 2500 \times 1000 \end{aligned} \quad (5)$$

Oxidative stress determinants – biochemical analysis

The hydrogen peroxide (H_2O_2) accumulation was quantified by following the process outlined by Velikova et al. (2000). In order to assess the H_2O_2 , a fresh leaf sample (0.25 g) was crushed in a cold mortar in 3 mL of 0.5% trichloroacetic acid solution. Test tubes were filled with potassium phosphate buffer (0.5 mL), sample extracts (0.5 mL), and KI (1 mL) after a centrifugation process for 15 minutes. A spectrophotometer (Model; WELab 721) recorded the reading at 390 nm after the vortex. A stress indicator, malondialdehyde (MDA), is used as a measure of lipid peroxidation. The MDA content was determined using the method described by Cakmak and Horst (1991). A grinding mixture of 0.5% trichloroacetic acid (TCA) solution was used to grind a fresh leaf sample (0.3 g). After centrifugation of the samples at 12,000 rpm for 15 minutes, the test tubes were filled with 1 milliliter of 0.5% TBA (thiobarbituric acid) in 20% TCA solution. Samples were heated at 95 °C, cooled on ice for 15 min, and absorbance was measured at 532 nm and 600 nm using a WELab 721 spectrophotometer.

Heavy metal determination

Heavy metal concentrations were determined following the protocol described by Khathi et al. (2015) with slight modifications. Dried and finely ground samples (0.5 g) were taken into a 100 ml digestion flask and digested with concentrated nitric acid (HNO_3) on a hot plate until a clear solution was obtained. After cooling, the digest was filtered through Whatman No. 42 filter paper, and the volume was adjusted to 25 ml using deionized water. The filtrate was collected in labeled polyethylene bottles for analysis. The concentrations of Cu^{2+} , Zn^{2+} , Fe^{3+} , and Pb^{2+} were determined using an Atomic Absorption Spectrophotometer (PG 990) equipped with appropriate hollow cathode lamps for each element. Analytical reagent (AR) grade standard solutions of the corresponding metals were used to create calibration curves. The corresponding standard calibration curves were used to determine the metal concentrations in the samples.

Statistical analysis

Analysis of the data was carried out using the HSD test at 5% probability according to Steel et

al. (1997). Statistical analysis and testing of the data were carried out using the Statistix software package (V 8.1), and the figures were prepared using Microsoft Excel.

RESULTS

Beetroot showed varying responses to different irrigation treatments. In general, wastewater application enhanced growth, yield, and physiological attributes, whereas biochar application intensified these responses and minimized heavy metal risks, paving the way for detailed results presented in the following sections. The F-values of all parameters for both years are shown in Table 1.

Physiological parameters

At UAF and Satyana, WW irrigation increased relative water content (RWC) by 22% at both sites in 2023 and by 22% (UAF) and 24% (Satyana) in 2024 compared with canal water. These results were illustrated in Figure 1. WB resulted in an increase RWC, with increases of 24% (UAF) and 44% (Satyana) in 2023 and 27% and 49%, respectively, in 2024. At Uchker, industrial WW reduced RWC by 5% in both 2023 and 2024, while WB mitigated this effect, increasing RWC by 16% and 17%, respectively, compared with canal water. At UAF and Satyana, WW irrigation reduced electrolyte leakage by 6% and 18% in 2023 and by 6% and 17% in 2024, respectively, compared with canal water. The addition of biochar (WB) further decreased electrolyte leakage, reaching reductions of 27% and 29% in 2023 and 25% and 27% in 2024 at the respective sites. In contrast, at Uchker, industrial WW markedly increased electrolyte leakage by 96% in 2023 and 92% in 2024. However, WB application effectively mitigated these adverse effects, restoring electrolyte leakage to levels comparable with the control in both years. At UAF and Satyana, WW increased Brix content by 9% and 8% in 2023 and 10% and 9% in 2024, respectively, while WB further increased it to 21% and 26% in 2023 and 23% and 29% in 2024. At Uchker, industrial WW reduced Brix by 6% in 2023 and 7% in 2024, while WB reversed this effect, increasing it by 19% and 21%, respectively, compared with canal water. At the UAF farm, WW increased TDS by 52% in both 2023 and 2024, whereas WB reduced it by 24% in 2023 and 22%

Table 1. F value of following traits of beet root for year 2023

ParameterS	Treatment		Sites		Treatment*Sites	
	2023	2024	2023	2024	2023	2024
Relative water content	42.93**	23.65**	8.97**	4.76*	6.76**	3.48*
Electrolyte leakage	235.51**	251.92**	383.81**	406.68**	187.96**	203.7**
Chlorophyll a	48.95**	48.95**	30.59**	30.59**	14.97**	14.97**
Chlorophyll b	82.99**	82.99**	19.48**	19.48**	7**	7**
Carotenoid	95.4**	71.82**	21.43**	22.67**	8.04**	6.91**
Malondialdehyde leaf	49.76**	49.35**	14.67**	14.71**	9.14**	9.16**
MDA Root	67.81**	68.53**	13.57**	13.74**	6.15**	6.23**
H ₂ O ₂ Leaf	29.3**	29.34**	36.16**	38.02**	12.09**	12.75**
H ₂ O ₂ Root	27.92**	329.59**	12.88**	61.29**	3.29*	44.86**
Co	217.78**	521.31**	297.78**	685.59**	94.16**	256.1**
Cr	1404.96**	1817.77**	3239.14**	4053.7**	1188.86**	1629.28**
Cu	621.38**	486.22**	1971.38**	1326.05**	672.36**	527.46**
Pb	1308.84**	1575.82**	3341.88**	4129.97**	1146.18**	1418.2**
Ni	649.96**	496.95**	1304.11**	1019.07**	396.75**	312.92**
Hg	265.79**	309.1**	249.1**	328.5**	78.41**	105.94**
Zn	1581.28**	906.78**	3357.87**	1942.71**	1252.22**	724.94**
Cd	148.88**	236.26**	193.51**	273**	77.02**	107.58**

in 2024. At Uchkera, WW raised TDS by 56% in 2023 and 55% in 2024, while WB decreased it by 19% and 18%, respectively. At Satyana, WW elevated TDS by 31% both years, and WB lowered it by 31% in 2023 and 29% in 2024.

Photosynthetic pigments

At the UAF site, WW treatment increased Chl a content by 12% in 2023 and 2024 growing seasons, while WB treatment further enhanced it to 33% in 2023 and 34% in 2024, as compared to canal water treatment. The corresponding data appeared in Figure 2. At Uchkera farm, WW treatment decreased Chl a content by 33% in 2023 and 2024; however, WB treatment mitigated this reduction, improving Chl a content by 15% in 2023 and 2024. At Satyana, WW treatment increased Chl a content by 11% in 2023 and 2024, with WB treatment further enhancing it to 21% and 21% in both growing seasons relative to canal water treatment. At the UAF site, WW treatment increased Chl b content by 16% in 2023 and 2024, while WB treatment further enhanced it to 56% and 57%, respectively. At the Satyana site, WW treatment increased Chl b content by 8% in 2023 and 2024, with WB treatment further enhancing it to 41% and 42%, respectively, relative to canal water treatment. Conversely,

at the Uchkera site, industrial WW reduced Chl b content by 30% in 2023 and 2024; however, WB treatment mitigated these effects, improving it by 29% and 30%, respectively. At the UAF site, WW treatment increased carotenoid content by 6% in 2023 and 10% in 2024, while WB treatment further enhanced it to 42% and 45%, respectively. At the Satyana site, WW treatment increased carotenoid content by 6% in 2023 and 2024, with WB treatment further enhancing it to 42% and 40%, respectively, relative to canal water treatment. Conversely, at the Uchkera site, industrial WW reduced carotenoid content by 36% in 2023 and 38% in 2024; however, WB treatment mitigated these effects, improving it by 30% and 35%, respectively.

Biochemical stress indicators

In 2023, WW treatment increased shoot MDA content by 23% at UAF and 30% at Satyana; in 2024, the increases were 23% and 30%, respectively. The associated data were displayed in Figure 3. The WB treatment further elevated these increases to 34% and 51% in 2023 and 33% and 51% in 2024. At Uchkera, industrial WW reduced shoot MDA content by 11% in 2023 and 2024 due to the presence of heavy metals. However, WB treatment mitigated these effects, leading to a

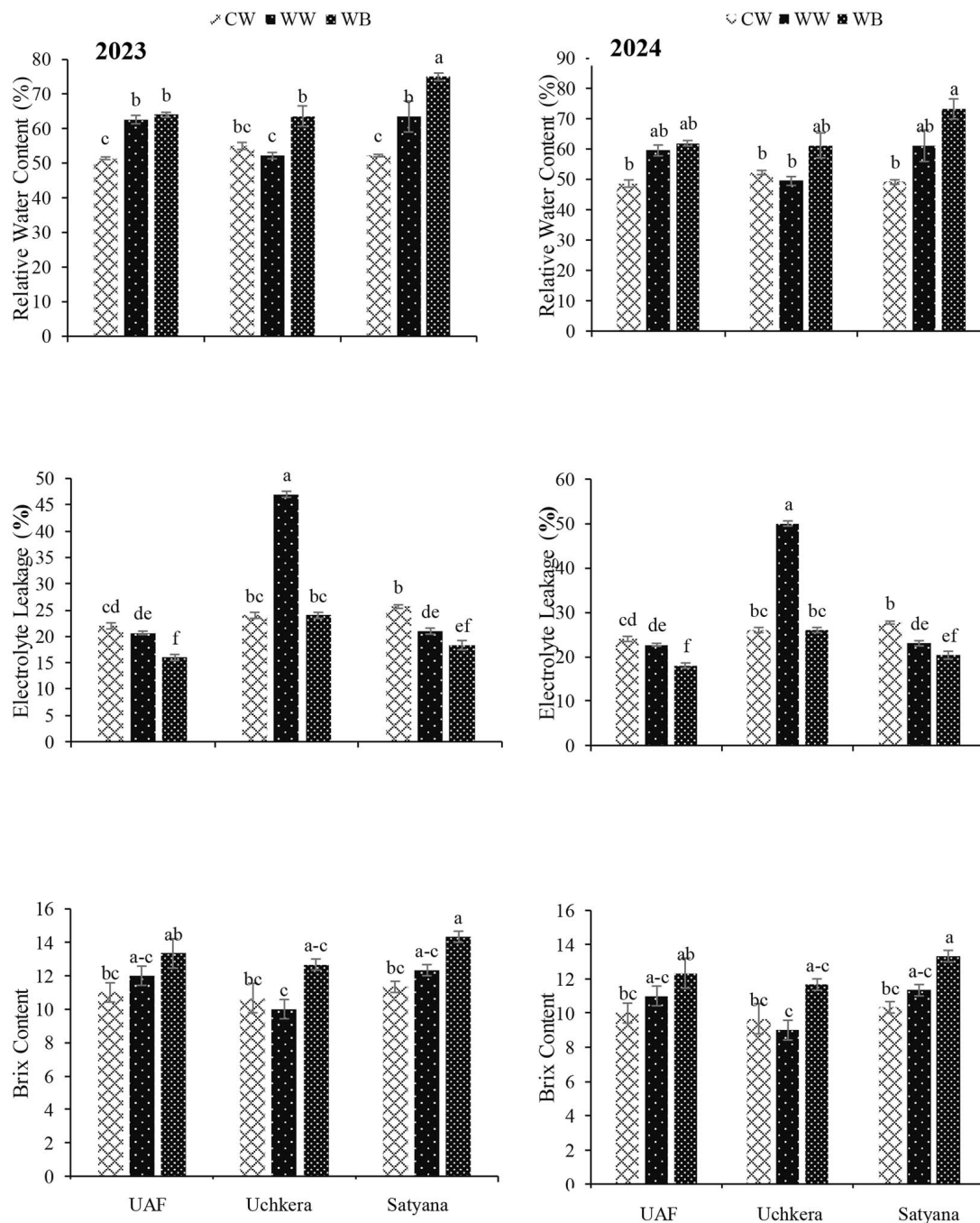


Figure 1. Effect of biochar-amended wastewater irrigation on relative water content, electrolyte leakage, and brix content across multiple sites

13% increase in 2023 and 2024 as compared to canal water treatment. At UAF, WW increased root MDA by 8% in both 2023 and 2024, while WB further raised it to 34% in both years. At Uchkera, industrial WW reduced root MDA content by 17% in both years. However, WB treatment mitigated these effects, leading to 13% increase in 2023 and 2024. At Satyana, WW increased root MDA by 7% in both 2023 and 2024, while WB further increased it to 27% in both years compared

with canal water. At UAF and Satyana, WW treatment increased leaf H_2O_2 content by 5% and 8% in 2023 and by 4% and 8% in 2024, respectively. The WB treatment further elevated these increases to 8% and 14% in 2023 and 8% and 13% in 2024. At Uchkera, industrial WW reduced leaf H_2O_2 by 15% in both 2023 and 2024 due to toxic contaminants. However, WB treatment mitigated these effects, leading to 9% increase in 2023 and 2024 compared to canal water treatment. WW

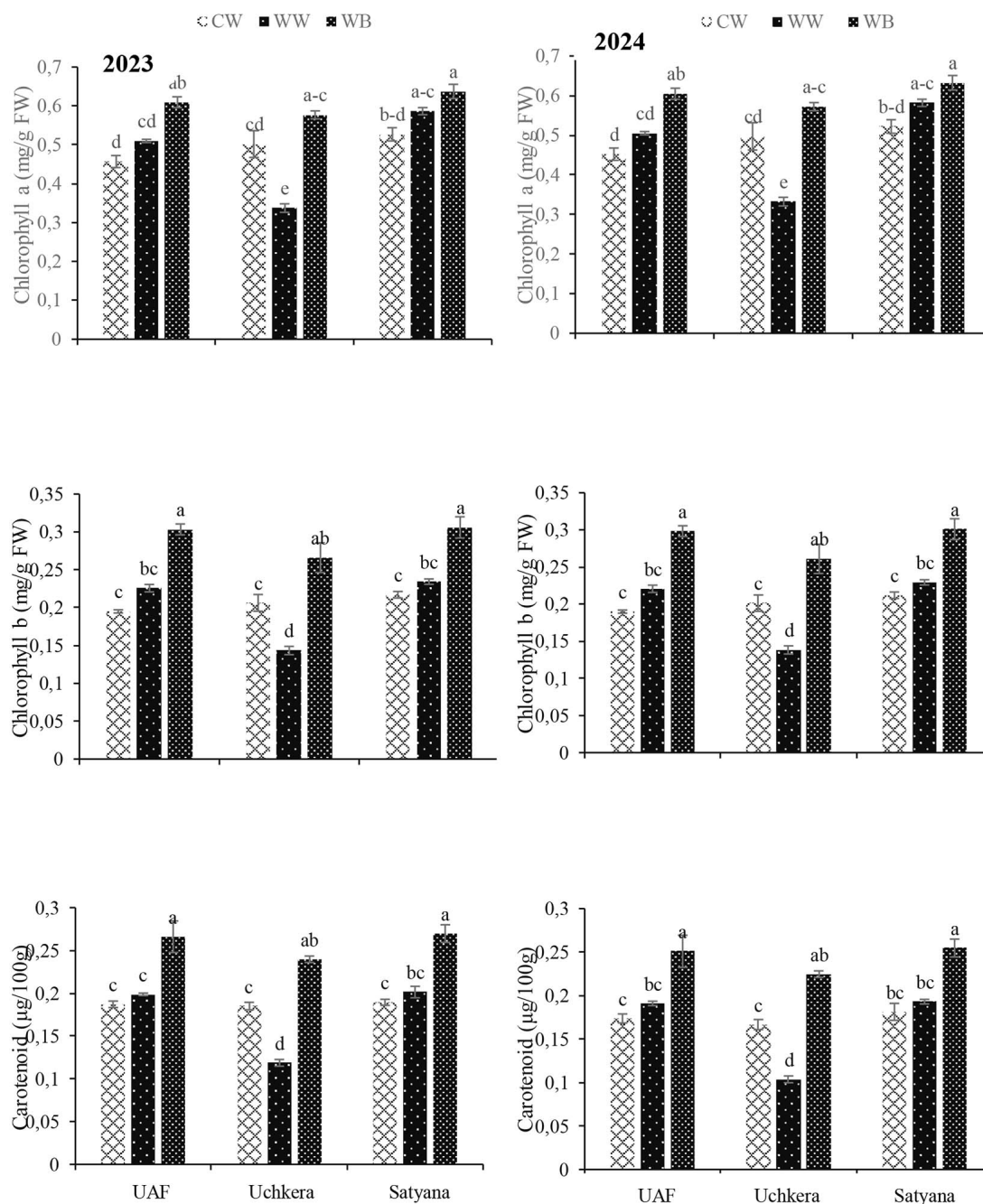


Figure 2. Effect of biochar-amended wastewater irrigation on chlorophyll a, chlorophyll b, and carotenoid across multiple sites

treatment increased root H_2O_2 content by 6% at UAF and 15% at Satyana compared to the canal water treatment in 2023, resulting in 7% increase at UAF and 8% increase at Satyana in 2024. Conversely, WB further reduced H_2O_2 content by 6% at UAF and 4% at Satyana in 2023 and led to 10% decrease at UAF and 12% at Satyana in 2024. The plants irrigated with WW treatment at Uchkera resulted in 30% increase in 2023 and 22% in 2024, while WB treatment led to 6% increase in 2023 and 8% increase in 2024.

Heavy metal accumulation

At the UAF site, WW treatment increased Cd levels by 19% in 2023 and 2024 while WB treatment reduced Cd by 3% in 2023 and 2024. Supporting data were shown in Figure 4. At Uchkera Farm, WW increased Cd levels by 130% and 127% in 2023 and 2024, while WB increased them by 37% and 33%, respectively. At the Satyana Farm, WW increased Cd by 5% in 2023 and 10% in 2024, while WB reduced it

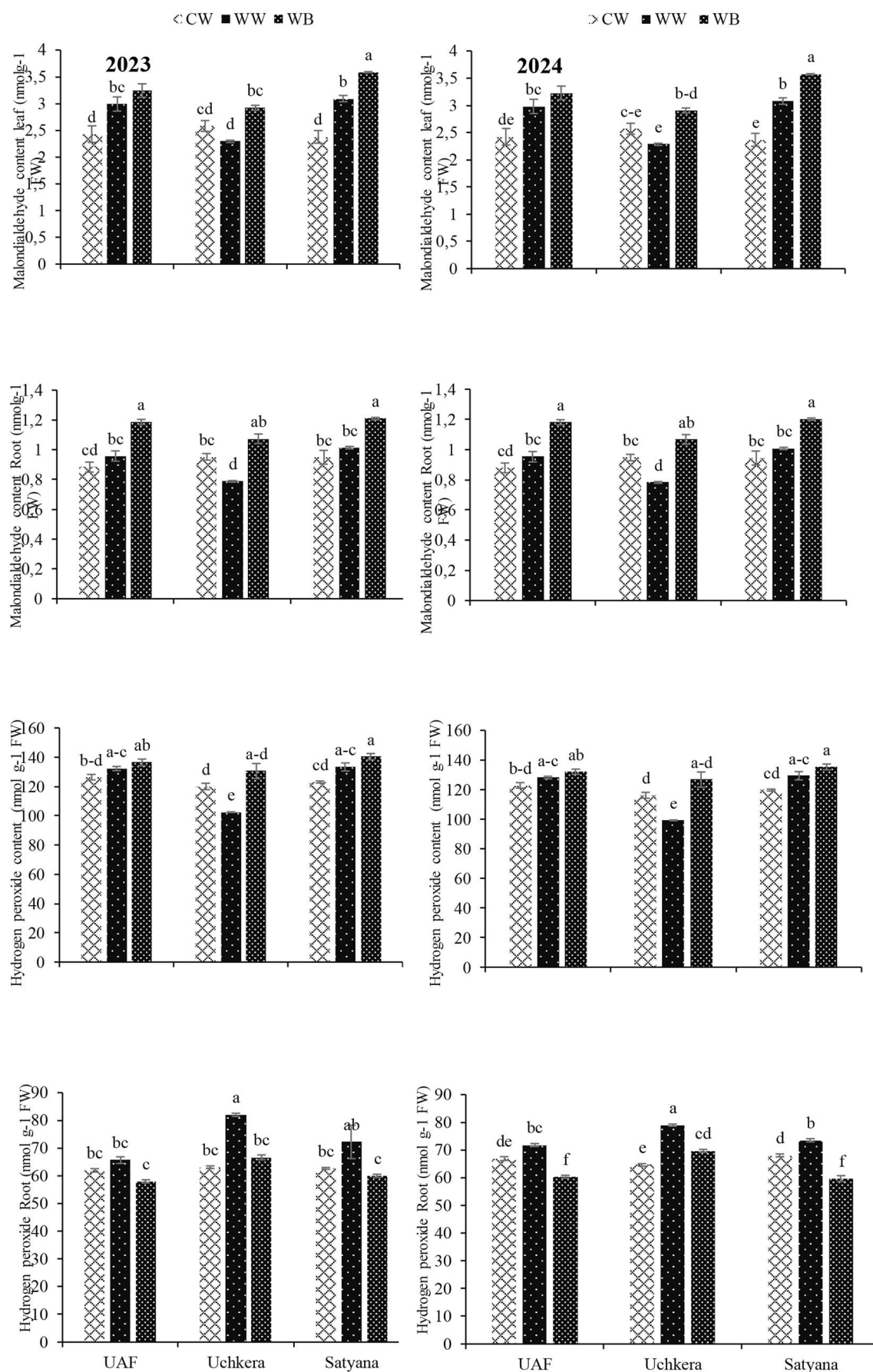


Figure 3. Effect of biochar-amended wastewater irrigation on malondialdehyde content leaf, malondialdehyde content root, hydrogen peroxidase content leaf and hydrogen peroxidase content root across multiple sites

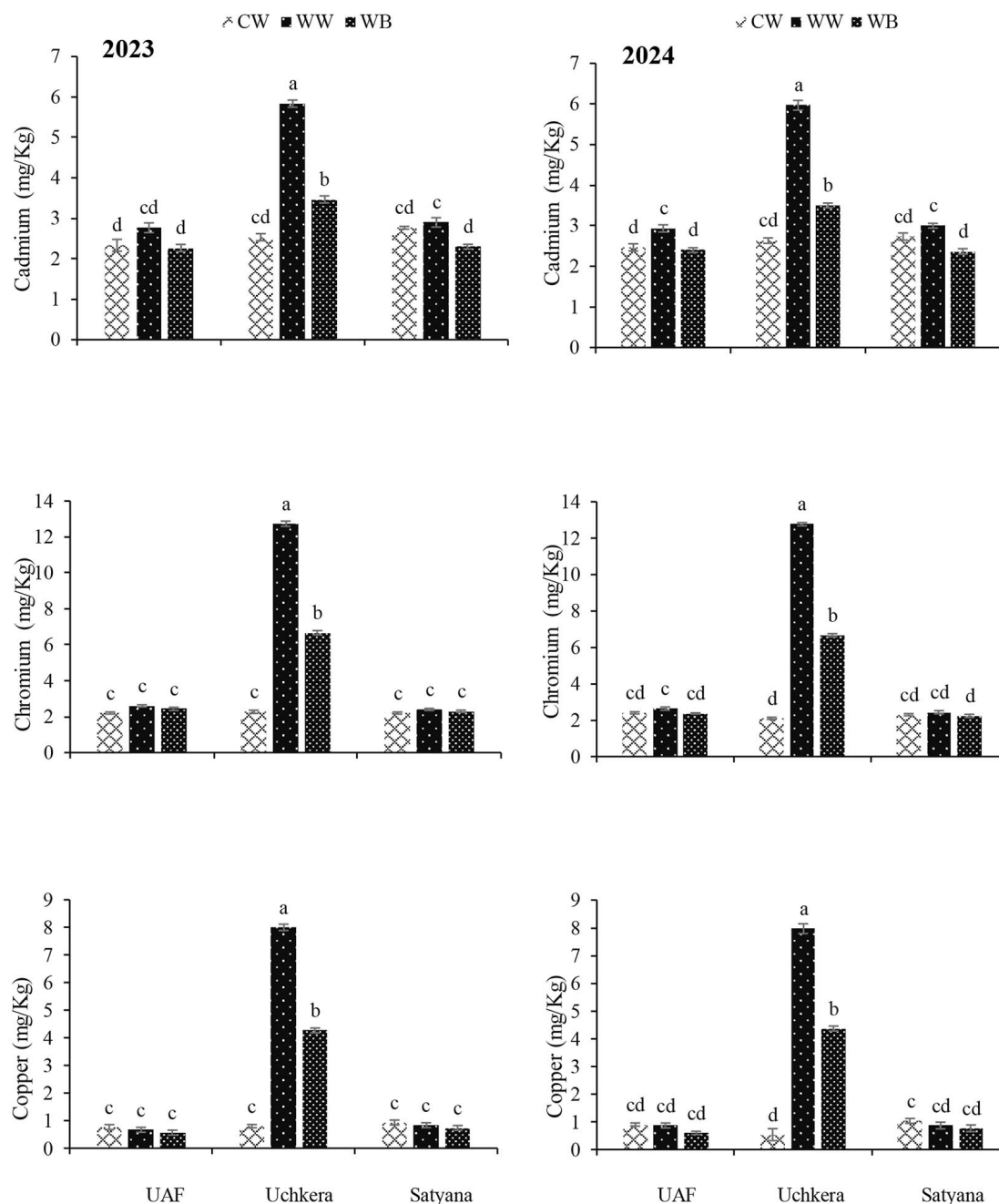


Figure 4. Effect of biochar-amended wastewater irrigation on cadmium, chromium, and copper across multiple sites

by 17% and 13%, respectively, compared with canal water. At UAF, WW increased Cr by 71% in 2023 and 78% in 2024, while WB reduced it by 42% in 2023 and 100% in 2024 compared with canal water. At the Uchkera site, WW treatment elevated Cr levels by 137% in 2023 and 140% in 2024, while WB treatment resulted in 58% increase in both years. At Satyana Farm, WW increased Cr by 100% in 2023 and 128% in 2024, while WB reduced it by 50% and 150%, respectively, compared with canal water. At

UAF, WW increased Cu by 13% in 2023 but decreased it by 4% in 2024, while WB reduced Cu by 26% and 33% compared with canal water. At Uchkera, WW treatment elevated Cu levels by 900% in 2023 and 1384% in 2024, while WB treatment increased Cu levels by 433% in 2023 and 714% in 2024. At Satyana, WW treatment increased Cu by 11% in 2023 but reduced it by 16% in 2024, whereas WB treatment decreased Cu levels by 21% and 26%, respectively, relative to canal water.

At UAF and Satyana, WW increased Pb by 21% and 14% in 2023, and by 17% and 10% in 2024, while WB reduced it to 5% and 10% in 2023, and by 3% and 5% in 2024 compared with canal water. These results were illustrated in Figure 5. Conversely, at Uchkera Farm, industrial WW increased Pb levels by 811% in 2023 and 802% in 2024. However, WB treatment mitigated this increase, leading to a 401% rise in 2023 and 404% in 2024 compared to canal water treatment. At UAF, WW increased Ni by 58% in 2023 and

49% in 2024, while WB increased it by 50% and 40% compared with canal water. At Uchkera Farm, WW treatment significantly elevated Ni accumulation by 419% in 2023 and 377% in 2024. However, WB treatment reduced Ni accumulation to 188% in 2023 and 168% in 2024 relative to canal water. At Satyana, WW increased Ni by 36% in 2023 and 27% in 2024, while WB raised it by 22% and 13% compared with canal water. At UAF and Satyana Farms, WW treatment increased Hg levels by 617% and 230% in 2023

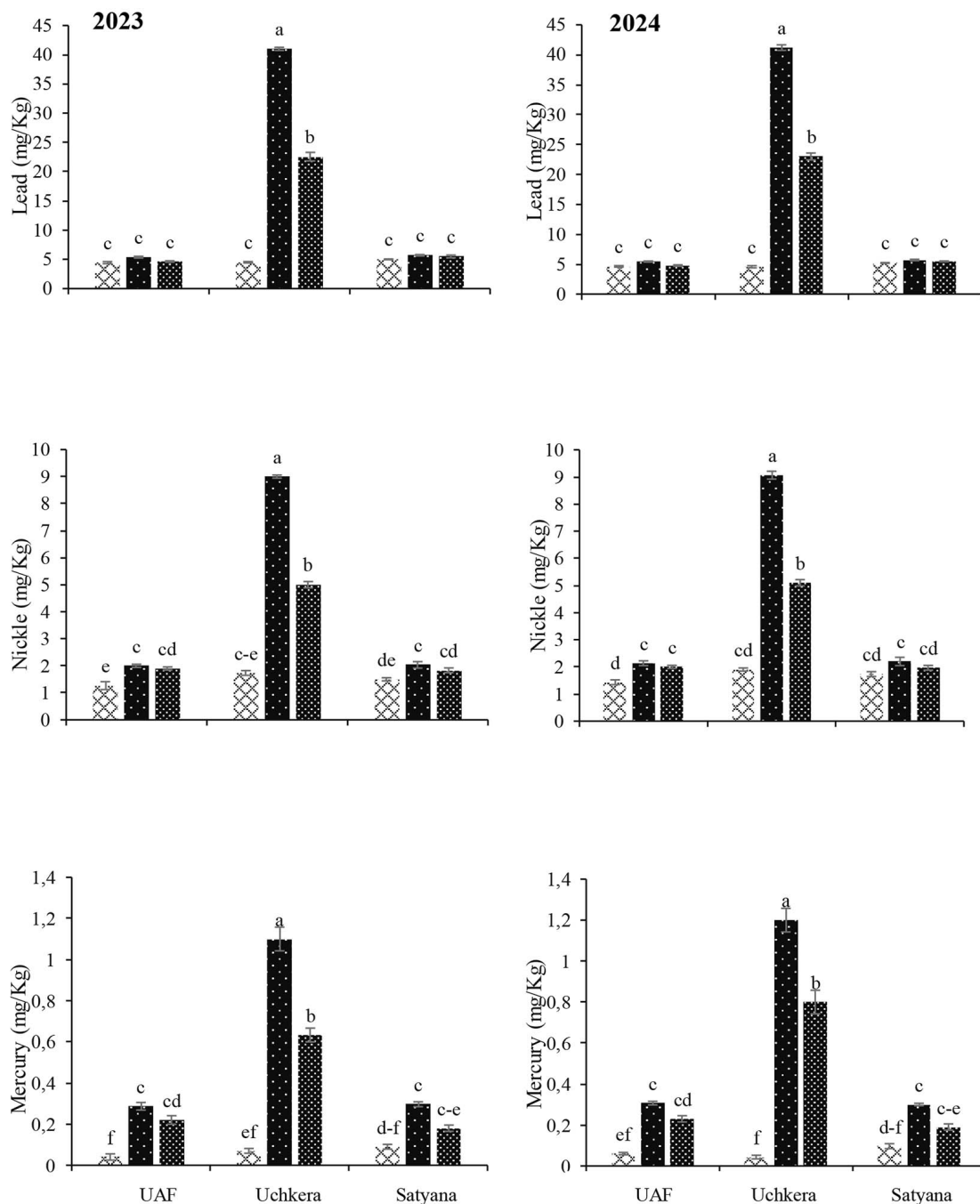


Figure 5. Effect of biochar-amended wastewater irrigation on lead, nickel, and mercury across multiple sites

and 411% and 210% in 2024, respectively, while WB treatment reduced Hg accumulation to 450% and 100% in 2023 and to 283% and 97% in 2024, relative to canal water, compared to canal water. Conversely, at Uchkera Farm, industrial WW significantly elevated Hg levels by 1471% in 2023 and 2669% in 2024. However, WB treatment mitigated this increase, reducing it to 805% in 2023 and 1746% in 2024 compared to canal water.

Figure 6 shows that at UAF and Satyana, WW increased Zn by 42% and 17% in 2023 and 40% and 14% in 2024, while WB reduced it to 21% and 6% in 2023 and 22% and 4% in 2024 compared with canal water. At Uchkera, industrial WW raised Zn by 777% in 2023 and 2024, while WB reduced it to 288% and 295% compared with canal water. At the UAF site, WW treatment increased Co levels by 82% in 2023 and 68% in 2024, while WB treatment significantly reduced Co accumulation by 55% in 2023 and 40% in 2024, compared to canal water. At the Uchkera Farm, WW treatment led to a substantial increase in Co levels by 1115% in 2023 and 1264% in 2024. However, WB treatment mitigated this rise, reducing it to 519% in 2023 and 616% in 2024. At the Satyana Farm, WW treatment increased Co levels by 233% in 2023 and 122% in 2024, while WB treatment did not alter Co levels in either year relative to canal water.

Correlation matrix and principal component analysis

The correlation analysis for 2023 and 2024 showed consistent relationships among the measured traits. These results indicated that better growth can lead to greater productivity. Chlorophyll content was also positively correlated with growth and yield parameters. These results indicated that better photosynthesis was required for better growth and productivity. Oxidative stress parameters, like malondialdehyde and hydrogen peroxide content, showed negative correlations with growth and yield parameters, which indicated their adverse effect on growth and productivity. These results showed that these parameters are consistent over both years, which is important for better growth and productivity. Detailed results are shown in Figure 7.

DISCUSSION

This study aimed to examine the complex responses of beetroot plants to wastewater irrigation and biochar amendments under various contamination conditions. From the study, complex responses of beetroot plants to wastewater and biochar amendments are evident.

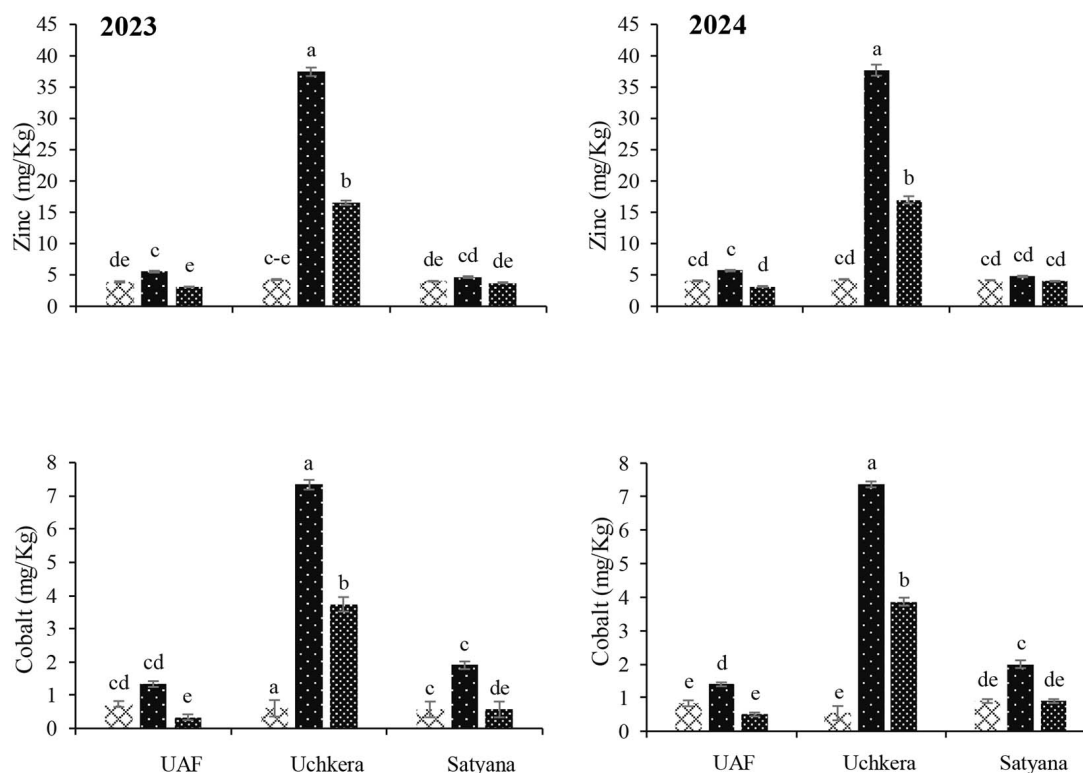


Figure 6. Effect of biochar-amended wastewater irrigation on zinc and cobalt across multiple sites

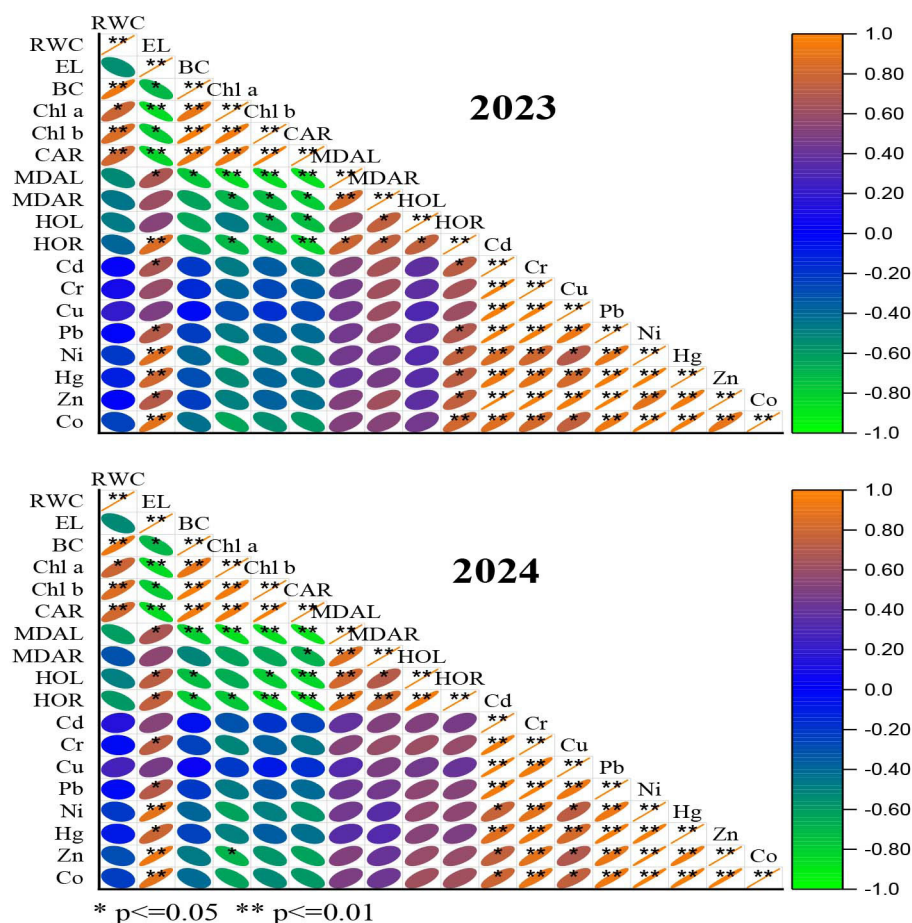


Figure 7. Correlation among beetroot growth, physiological, biochemical, and heavy metal traits under wastewater and biochar treatments. RWC– relative water content, EL– electrolyte leakage, BC– brix content, Chl a – chlorophyll a, Chl b – chlorophyll b, Car – carotenoid, MDAL – malondihyde leaf, MDAR – malondihyde root, HOL – hydrogen peroxidase leaf, HOL – hydrogen peroxidase root, Cd – cadmium, Cr – chromium, Cu – copper, Pb – lead, Ni – nickle, Hg – mercury, Zn – zinc, and Co – cobalt

As toxicity decreases the photosynthesis and minimizes the chlorophyll contents in *Boehmeria nivea* L. Likewise, Cd has a negative effect on the chlorophyll contents and gas exchange attributes of the plant (Shahid et al., 2017). The use of sulfur-iron coated biochar in Cd-polluted soils increased the chlorophyll contents of rice plants (Rajendran et al., 2019).

Biotic as well as abiotic stresses in plants cause oxidative stress through the production of reactive oxygen species (ROS), which in turn causes the degradation of biomolecules, structural changes, etc. (Noman et al., 2018). Higher concentrations of H_2O_2 , EL, and MDA in the plants grown in the absence of BC treatments. The reason for this is the lipid peroxidation due to the toxicity of heavy metals in the plants, which in turn increases the content of MDA in the rice plants. The obtained results are in accordance with the previous results that the application of

BC reduced the activity of MDA, H_2O_2 in plants (Farhangi-Abriz and Torabian, 2018). Plants have a strong defense mechanism to combat the harmful effects of ROS. The increased activity of antioxidant enzymes scavenges the overproduction of ROS (Noman and Aqeel, 2017).

Previous research demonstrated that biochar application significantly reduces heavy metal uptake by plants. Similarly, the present findings show marked reductions in the accumulation of Cd, Cu, Cr, As, Pb, Zn, Ni, and Mn under biochar-amended conditions, indicating its effectiveness in limiting metal phytoavailability and plant uptake (Natasha et al., 2022).

The amendments with biochar have been reported to increase the biosynthesis of secondary metabolites and osmo-protectants in the plants grown with both domestic and industrial wastewater irrigation by improving soil fertility, enhancing microbial diversity, and reducing

contaminant stress (Khaliq et al., 2023). These aspects have been well supported by recent studies and case reports, which confirm the utility of biochar as a sustainable soil amendment for crop growth and productivity under diverse conditions (Ahmed et al., 2023; Jiang et al., 2020). Biochar has been reported to improve soil fertility, water retention, and crop productivity, and to decrease the transfer of heavy metals and organic contaminants from wastewater to crops, thereby improving food safety (Shakeel et al., 2021). Biochar has been reported to increase soil organic carbon, beneficial microbe populations, and root-microbe interactions, which together increase the biosynthesis of secondary metabolites and osmo-protectants in crops (Yang et al., 2025). These effects are consistent with literature showing the multifunctional role of biochar in promoting sustainable and safe crop production under wastewater irrigation (Murtaza et al., 2025).

CONCLUSIONS

Wastewater irrigation resulted in improved beetroot growth and yield, but it also increased heavy metal levels in the beetroot. Biochar amendment significantly reduced heavy metals in beetroot, increased antioxidant activity and improved growth and soil conditions in wastewater-irrigated beetroot cultivation. It is concluded that the biochar-treated wastewater is a safer and more environmentally friendly method of beetroot cultivation, and its effect is more pronounced in 2024 than in 2023.

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