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Manure-mixed biochar decreases concentration of Cadmium and arsenic in fruits of tomato grown in coal mine tailing debris-contaminated soil

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Abstract

Tomato is one of the major vegetable crops of the province of Balochistan, Pakistan. This province has been under extensive mining activities for decades for various minerals. Heavy metals from these activities enter into the food chain via particulate dispersion in air, water and soil while eventually getting absorbed into crops. Empirical reports suggest that the use of biochar as an amendment in agricultural lands reduces the absorption of heavy metals by crops from contaminated soil. In this study, the soil was contaminated with coal mine tailing debris. A mixture of air-dried poultry manure with wood biochar or farmyard manure biochar at the ratio of 1:1 w:w was amended in soil. For the treatment of thorough mixing of fertilizer in the soil, biochar-based organic fertilizer was applied at the rates of 5 and 10 t ha^{-1} and the soil was contaminated with 10 t ha^{-1} of debris from coal mine tailings. To assess the influence of fertilizer in heavily-contaminated soil, debris was placed in the root zone of tomato at 166.5 t ha^{-1} rate and fertilizers were added at 33.3 and 66.6 t ha^{-1} rates, followed by their mixing with debris. The fertilizers decreased the concentration of Cadmium (Cd) by 63 – 83% and arsenic (As) by 51 – 73% in tomato fruits than control ($P \leq 0.05$) in low-contaminated soil. In high-contaminated soil, fertilizers decreased the concentration of Cd by 40 - 49% and As by 44 – 56% in tomato fruits ($P \leq 0.05$). A negative correlation was observed between the level of contamination in soil (i.e., root-zone application versus thoroughly mixing of debris in soil) and the potential of fertilizer in reducing the concentration of heavy metals in fruits of tomato ($R^2 = 0.79$; $P < 0.01$ for Cd and $R^2 = 0.27$; $P < 0.01$ for As). The application of poultry manure-mixed biochar in coal mine tailing contaminated soil has the potential for reducing the concentration of these heavy metals in tomato fruits.

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Introduction

The particulate dispersion of metalloids into the air, soil and water from mining activities poses serious threats to human health and the environment (Gil-Loaiza *et al.,* 2016; Ayub and Ahmad, 2020; Chandio *et al.,* 2021). The dumping sites of mine tailings from mining activities usually have acidic pH, high concentration of heavy metals (e.g., Cadmium (Cd), zinc (Zn), copper (Cu), chromium (Cr), arsenic (Ar)) and have lower water and heavy metal retention capacity, which threatens surrounding vegetation (Mendez *et al.,* 2007; Zhang *et al.,* 2014). Agricultural lands in the mining regions have high concentrations of heavy metals in soil from mining activities (Limie *et al.,* 2008; Chandio *et al.,* 2021). Limie *et al.* (2008) reported that agricultural lands that were cultivated with rice in the Chenzhou City, Southern China, at distances from mining activities of as high as 23 km to 63 km, had Cadmium that exceeded many times its critical concentration in soil, vegetables and rice. Their findings show that mining activities cause contamination of soils from high distances.

Balochistan is the largest province of Pakistan that covers more than 40% of the land area of the country. The total land area of this province is 347,190 km^2. This province has large reserves of different minerals such as chromite, coal, copper and fluoride. These minerals have been excavated for decades (Ahmad *et al.,* 2019; Chandio *et al.,* 2021). The mining activities in Balochistan have heavily contaminated soil and water (Geological Survey of Pakistan unpublished data; Ayub and Ahmad, 2020). Chamalang, Deragi, Duki, Sor-Range, Harnai, Sharagh, Khost, Ziarat and Mach are the mining regions of coal in Balochistan (Ayub and Ahmad, 2020). Debris from coal mining have high concentrations of lead, Cd and As (Bhardwaj *et al.,* 2020; Raj, 2019; Ishtiaq *et al.,* 2018). These heavy metals are accumulated from dumping sites of coal mine tailings to the agricultural soils and then to the edible tissues of crops (Cao *et al.,* 2009; Raj, 2019; He *et al.,* 2021).

Biomaterials such as biochars, which have high sorption capacity due to high surface area and porosity, reduce the concentration of heavy metals in crops that are grown in the soils, which are contaminated with trace metals from mining activities (Munir *et al.,* 2020; Khan *et al.,* 2020). The positive impact of these biomaterials on the yield of the crops that are grown in the contaminated soils can be achieved when these are co-applied in soil with other organic or inorganic fertilizers (Gul and Whalen, 2016). Balochistan ranks first in the production of tomato in Pakistan (GOP, 2015). As this region is also under high mining activities, this causes serious problems regarding water and soil pollution (Chandio *et al.,* 2021). Tomato being a major vegetable crop in the regions with high mining activities such as Harnai and Duki, can be grown with improved yield and reduced accumulation of hazardous trace metals, e.g. Cadmium (Cd) and arsenic (As), if biochar is applied in the soil as a mixture with manure.

The objective of this study was to evaluate the influence of mixtures of wood biochar + poultry manure or cow manure biochar + poultry manure as soil amendments on the reduction of Cadmium and arsenic in the fruits of tomato that was grown in the contaminated soil with coal mine tailing debris under field conditions.

Materials and methods

Study site

The study was conducted in Pishin, Balochistan, in private land that was not previously cultivated. The climate of the study area is the Mediterranean. Winter and spring seasons receive rainfall and snowfall; whereas, the summer season usually goes dry. From April 01 till August 31 of the study year (2018), the average monthly rainfall was 0.33, 0.06, 0, 0.02 and 0.01 mm, respectively. The soil was sandy, with 670 g sand, 110 g silt and 50 g clay, pH as 7.84, organic matter as 12 g kg^{-1}, nitrate as 0.06 mg kg^{-1} and Olsen phosphorus as 3 mg kg^{-1} soil.

Coal mine tailing debris was collected from the mining site at Sharagh, Balochistan. A factorial experiment with the factors 1) two methods of organic amendments (root-zone application versus mixing of organic fertilizer in soil), 2) two biochar types produced separately from cow manure and wood and mixed separately with poultry manure and 3) three application rates of biochar-based organic fertilizers as thoroughly mixed in the soil, i.e., 0 (control), 5 and 10 t ha^{-1} rate, two application rates of biochar-based fertilizers in the root-zone of tomato, i.e., 33.3 and 66.6 t ha^{-1} rate and 4) two levels of contamination of soil with coal mine tailing debris, i.e., 10 t ha^{-1} for the treatments of thorough-mixing of organic fertilizer in the soil and 166.5 t ha^{-1} for the treatment of the application of fertilizer in the root-zone of tomato. Each treatment had three replications and the total experimental units were 27.

Preparation of a mixture of poultry manure with biochars

In timber markets of Quetta, Balochistan, Pakistan, wood biochar is available for Bar BQ purposes. The wood biochar used in this study was obtained as small leftover broken pieces from the timber market in Quetta, Balochistan, Pakistan, as these leftovers are inexpensive (~10/- PKR per 1 kg biochar) as compared to unbroken pieces. The feedstock for this wood biochar is *Acacia nilotica* and is produced in mud-made underground kilns (known as Bhatti in the local language). Manure from a farmyard of cows and buffalos in Quetta city was purchased and air-dried. The air-dried poultry manure was obtained (free of charge) from a poultry farm in Quetta city. The air-dried FYM was burned in the domestic kiln. These biochars were slow-pyrolyzed since they were produced in kilns. The production temperature of kilns for biochar is in the range of 350-550 °C (Mia *et al.,* 2015). The properties of manure and biochars are listed in Table 1. The mixture of air-dried poultry manure with these air-dried biochars was made at a 1:1 w:w ratio.

Preparation of plots, soil contamination, amendment of fertilizers and plantation of tomato seedling

Twenty-seven 1.5 X 0.5 m plots were prepared. The edges of plots were made thick and elevated for tomato seedling implantation (Supplementary Figure). In 15 plots, 1 kg of coal mine tailing debris was amended thoroughly in soil. This amount equals to 10 t ha^{-1}. For the 12 of these plots, on the sides of edges of each plot, poultry manure + biochar mixtures were applied at 5 or 10 t ha^{-1} (Supplementary Figure) and were subsequently mixed in the soil. For the rest of 12 plots, in each plot, five holes of 10 cm height and 3 cm diameter were made (Supplementary Figure). In each hole, 0.5 kg coal mine tailing debris was added. This amount equals to 166 t ha^{-1} amendment. In these holes, mixtures of poultry manure with biochars were added as 0.1 kg or 0.2 kg. This amendment rate equals 33.3 and 66.6 t ha^{-1}, respectively. The fertilizer and debris were thoroughly mixed in these holes after amendment. Thereafter, the hole was filled with clean soil (soil with no contamination). Tomato seedlings of Roma variety were purchased from the local nursery. Seedlings were planted on April 3, 2018. These seedlings were planted at the edges of plots facing the plot center (Supplementary Figure). This is the common practice of tomato cultivation in Balochistan. For the root-zone fertilizer treatment, seedlings were planted in filled holes. In each plot, five seedlings were left for growth and others were removed after the establishment of seedlings.

Harvest of biomass, fruits, and assessment of arsenic (As) in fruits of tomato

Fruits started to develop from the last week of May. Ripened fruits were regularly harvested, followed by their air-drying, weighing and grinding, till mid of August. On August 21, aboveground plant biomass was harvested with roots, air-dried and weighed (Supplementary Figure).

Analysis of tomato fruits for As and Cd

The quantitative analysis of plant samples for As and Cd was performed on Atomic absorption spectrophotometer (Fast Sequential AA240FS) at the Department of Microbiology, Quaid-i-Azam University, Islamabad, Pakistan. Each sample was analyzed three times and the average value of absorbance was considered as the concentration of a given trace metal. The digestion of plant samples was carried out by grinding 1 g of plant sample followed by its mixture in 15 mL aqua regia (1HCL+3HNO_3). Thereafter, the aliquot was heated at 150°C, left overnight, added 5 mL of $HClO_4$ again and heated again at 150 °C, until the solution became dry and brown fumes came out. 50 mL double deionized water was added and the solution was passed through Whatman filter paper (No. 42) and the volume was again raised to 50 mL (FAO/SIDA, 1983). As a control, soil obtained from the garden was run in parallel (proof of analysis of Cadmium and Arsenic metals found in the processed samples is given in Supplementary File).

The nutrient use efficiency as "nutrient efficiency ratio" of tomato fruits for As and Cd was assessed with the following formula (Baligar *et al.,* 2001);

$$\text{NUE (for As or Cd)} = \frac{\text{Tomato yield}}{\text{Concentration of heavy metal in tomato fruit}} \quad (1)$$

Statistical analysis

The data of tomato fruit yield, stover biomass and heavy metals were assessed for normal distribution via D/Agostino-Pearson K^2 test. Thereafter, data of all parameters were analyzed with analysis of variance (ANOVA) and least significant difference (LSD) test. Data analysis was carried out using Microsoft Excel and CoStat (version 6.311).

Results and discussion

With an exception, the amendment of the mixture of poultry manure with biochars did not increase yield than control treatment, as is evident from Fig. 1. The only acceptance was observed for the poultry manure + wood biochar mixture that was applied at 33.3 t ha^{-1} in heavily contaminated soil ($P \leq 0.05$; Fig. 1).

This is not uncommon that the application of organic fertilizer-mixed biochar does not increase the yield of crops in mining-affected contaminated soils (Novak *et al.,* 2016). However, in heavily contaminated soil, the application of poultry manure + wood biochar mixture enhanced yield significantly ($P < 0.05$; Fig. 1).

Table 1. *Properties of manure and biochars.*

Properties	Wood biochar	Cow manure	Poultry manure
pH	8.15	8.55	8.21
EC (μs cm^{-1})	14.6	9.47	11.2
Ash wt. %	4.5	36.8	44.9

Data is obtained from Ghani *et al.* (unpublished submitted data); they used the same biochar and manure.

The increase in the yield of tomato in response to the amendment of wood biochar has also been observed in our previous study (Hameeda *et al.,* 2019; Manzoor *et al.,* 2019). Likewise, improved yield of crops after the amendment of biochar in coal-mine-contaminated soil has also been reported frequently (Liu *et al.,* 2018; Munir *et al.,* 2020). For example, in a pot-based study, the amendment of slow-pyrolyzed biochar (purchased from Henan Sanli New Energy Co., Ltd.), at 1, 2 and 4% application rates with synthetic nitrogen fertilizer, in contaminated soil from coal-mining activities, Liu *et al.* (2018) observed ~2 to ~4 times increase in the grain yield of wheat than the control treatment. They attributed the positive influence of inorganic nitrogen fertilizer-mixed biochar on the yield of wheat to their positive influence on soil quality and soil microbial functions. In our study, at a high application rate (i.e., 66.6 t ha^{-1}), however, a significant decrease in yield than control treatment in heavily contaminated soil maybe because of possible high absorption of nutrients from poultry manure, which might have reduced yield.

The amendment of poultry manure mixture with both biochar types significantly reduced the concentration of heavy metals in fruits by ~2 to ~4 times than control treatment ($P \leq 0.05$; Fig. 2). These fertilizers decreased the concentration of Cd by 63 – 83% and As by 51 – 73% in tomato fruits as compared to control ($P \leq 0.05$) in low contaminated soil.

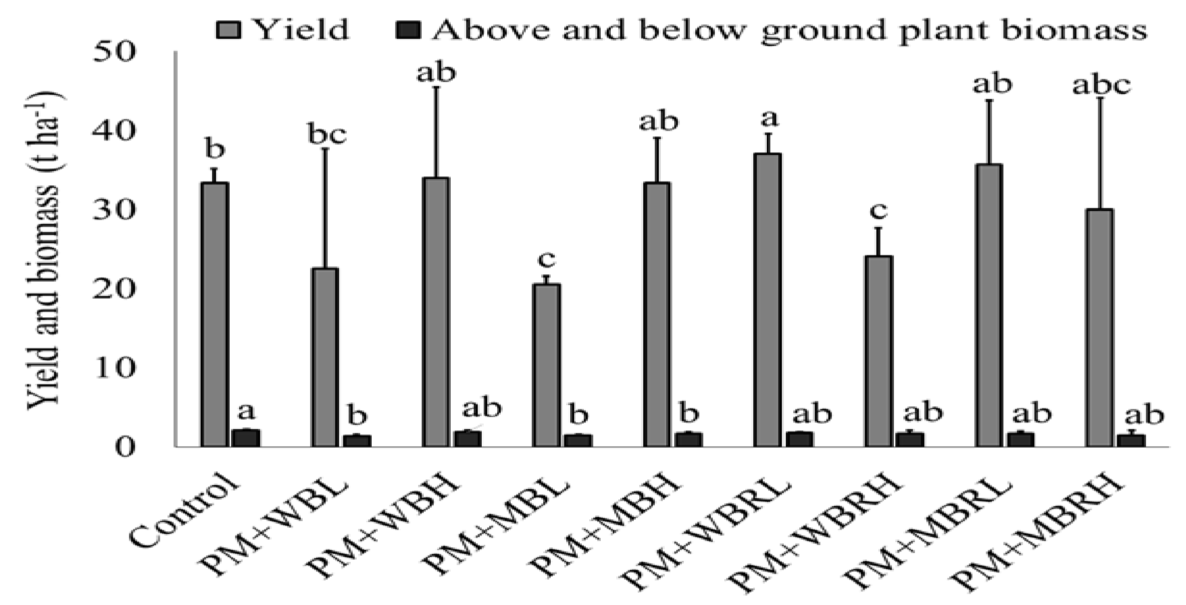

Fig. 1. Yield and cumulative of above and belowground plant biomass of tomato (mean ±SD). Bars of a given parameter (yield or above and belowground plant biomass) followed by different letters are significantly different (P,0.05). PM – poultry manure, WB – wood biochar, MB – cow dung biochar, L – low application rate of manure-mixed biochar (5 t ha^{-1}), H – high application rate of manure-mixed biochar *(10 t ha^{-1}), R – application of manure-mixed biochar in the root zone of the plant*.

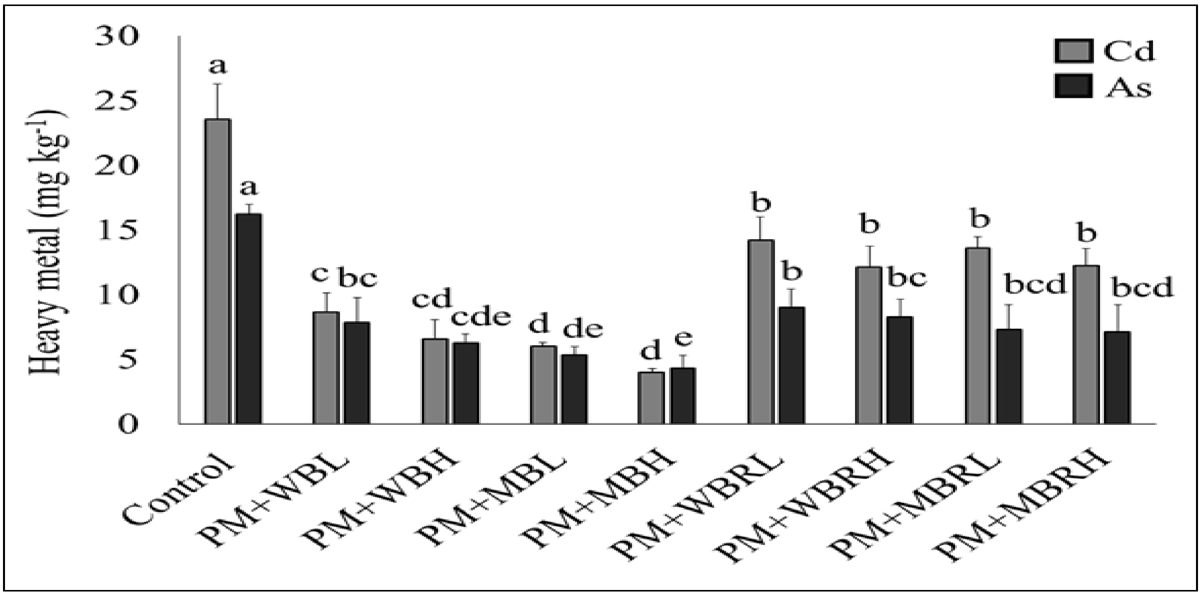

Fig. 2. The concentration of Cd and As in fruits of tomato (mean ±SD). Bars of a given parameter (yield or above and belowground plant biomass) followed by different letters are significantly different (P0.05). PM – poultry manure, WB – wood biochar, MB – cow dung biochar, L – low application rate of biochar + manure mixture (5 t ha^{-1}), H – high application rate of biochar + manure mixture (10 t ha^{-1}), R – application of biochar + manure mixture in the root zone of the plant.

In high contaminated soil, these fertilizers decreased the concentration of Cd by 40 - 49% and As by 44 – 56% in tomato fruits ($P \leq 0.05$). Furthermore, reduction in the concentration of Cd and As in fruits in response to the amendment of manure + biochar mixture is seen in all treatments, suggesting that these organic fertilizers have the potential for reducing the absorption of these hazardous heavy metals by tomato fruits. Our findings are in agreement with previous empirical results, which demonstrated that the amendment of biochar-based fertilizers reduces the uptake of heavy metals in crops (Ali *et al.,* 2017; Munir *et al.,* 2020). Interestingly, a trend can be seen that poultry manure + FYM biochar mixture reduced concentration of Cd and As in tomato fruits more than poultry manure + wood biochar mixture (P ≤ 0.05; Fig. 2).

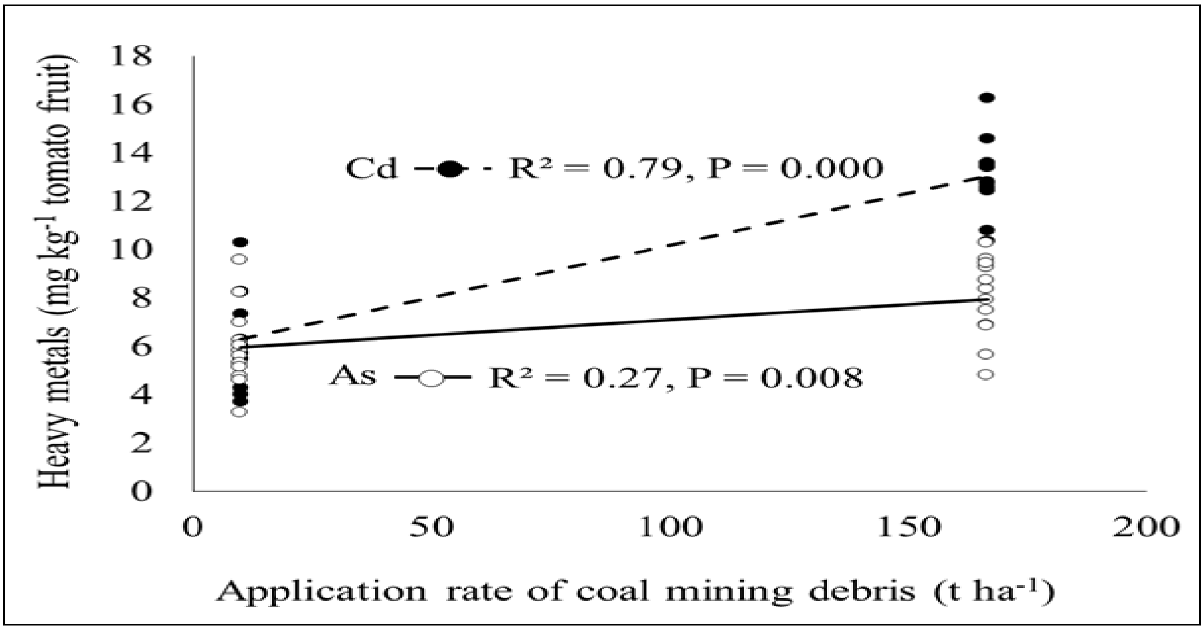

Fig. 3. Regression analysis between Cd and As in fruits of tomato with an application rate of coal mining debris in the soil.

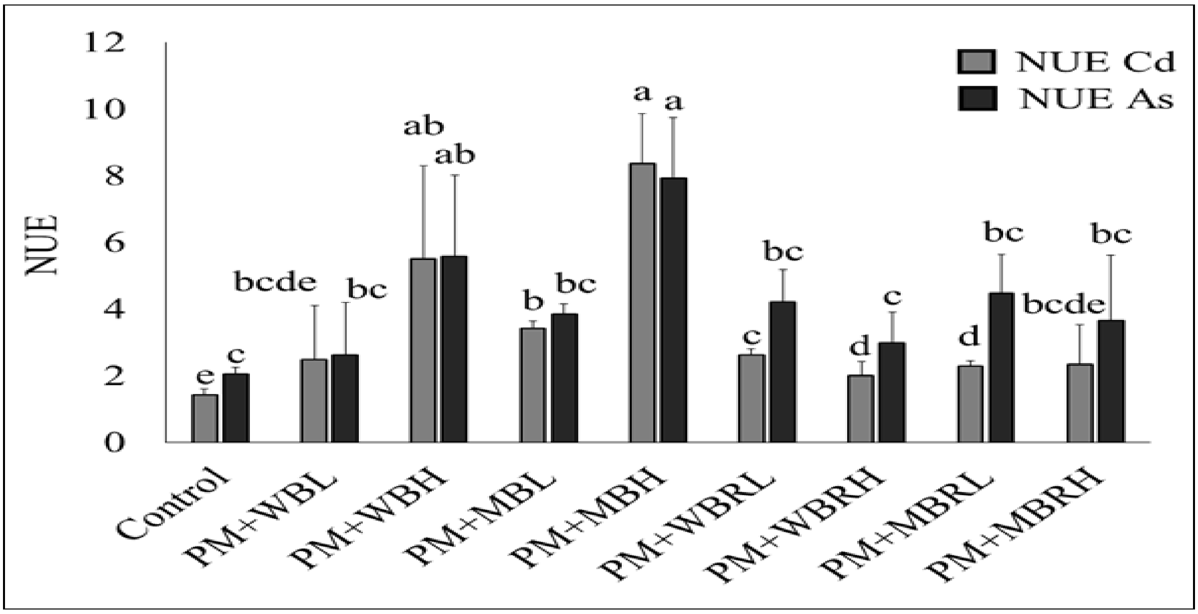

Fig. 4. NUE of tomato fruits for Cd and As (mean ±SD). Bars of a given parameter (yield or above and belowground plant biomass) followed by different letters are significantly different (P,0.05). PM – poultry manure, WB – wood biochar, MB – cow dung biochar, L – low application rate of manure-mixed biochar (5 t ha^{-1} for thoroughly mixed and 33.3 t ha^{-1} for root zone fertilizer placement), H – high application rate of biochar + manure mixture (10 t ha^{-1} for thoroughly mixed and 66.6 t ha^{-1} for root zone fertilizer placement), R – application of biochar + manure mixture in the root zone of the plant.

This effect may be because manure biochars are more porous and have higher binding capacity than wood biochars (Wang *et al.,* 2016; Domingues *et al.,* 2017); therefore, their amendment in contaminated soil may cause more reduction in crop-availability of heavy metals than wood biochar-based organic fertilizers.

However, in heavily contaminated soil, this trend was not observed. Moreover, a negative correlation was found between the level of contamination in soil (i.e., heavily-contaminated soil through the root-zone application of debris versus low-contaminated soil by thoroughly mixing of debris in soil) and the potential of fertilizer in reducing the concentration of heavy metals in fruits of tomato (R^2 = 0.79; P < 0.01 for Cd and R^2 = 0.27; P < 0.01 for As). This suggests that the capacity of biochar for reducing heavy metal absorption in the edible part of tomato (e.g., fruit) declines as the concentration of contamination in soil increases. Another observation is that amendment of organic fertilizers significantly increased NUE for As and Cd in tomato fruits (P ≤ 0.05; Fig. 4). This effect was more profound for Cd than for As.

Conclusion

The amendment of poultry manure mixture with wood-based and FYM-based biochars did not significantly increase yield than control treatment (which had a lower level of contamination i.e., 10 t ha^{-1} mine-tailing debris), except for poultry manure + wood biochar mixture that was applied at 33.3 t ha^{-1} in heavily contaminated soil (166.5 t ha^{-1} mine-tailing debris). Heavy contamination of soil did not reduce the yield of tomato, which may be because fertilizers were also amended at higher rates. The exception was seen for amendment of the mixture of poultry manure with wood biochar at 66.6 t ha^{-1} rate, which significantly reduced yield than the control treatment. The amendment of the mixture of poultry manure with wood biochar and FYM biochar in coal mine tailing-contaminated soil resulted in 2-5 times reduction in the concentration of Cd and ~2 times reduction in the concentration of As in the fruits of tomato. Poultry manure and FYM are inexpensive biowastes. Their use as biochar-based fertilizers can be promising in reducing the concentration of Cd and As in crops that are grown in agricultural soils in the vicinity of mining activities.

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