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Mitigating the Toxic Effects of Chromium on Wheat (*Triticum aestivum* L.) Seed Germination and Seedling Growth by Using Biochar and Polymer-Modified Biochar in Contaminated Soil

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Abstract: The present study was conducted to investigate the potential influences of biochar in mitigating the phytotoxic effects of hexavalent chromium (Cr^{VI}) on the germination of wheat (*Triticum aestivum* L.). Biochar (JBC) was produced from Jujube (*Ziziphus jujube* L.) wood waste at three different pyrolysis temperatures (300 °C, 500 °C and 700 °C), which was later polymerized (JPBC) via the solution-polymerization method. Phytotoxicity of Cr^{VI} was induced to wheat seeds at variable Cr^{VI} application rates (5, 10, 20, 40 mg L⁻¹). Applied Cr^{VI} concentrations confined the seed germination and seedling growth in order of: 5 < 10 < 20 < 40 mg L⁻¹. The application of JBCs (0.2 g per petri plate) resulted in a 150% increase in shoot length, while dry biomass was increased by 250% with JPBCs application. Uptake of Cr^{VI} was significantly lower in JBC-300 (7.74 µg/seedling) and JPBC-300 (1.13 µg/seedling) treatments, as compared to control (13.24 µg/seedling), at the highest stress level (40 mg L⁻¹). Therefore, the findings of the current study showed that JBCs and JPBCs performed excellently in improving seedling growth while JPBCs performed more efficiently than pristine JBCs in mitigating Cr^{VI} phytotoxicity and availability.

Keywords: biochar; chromium; phytotoxicity; polymer-modified biochar; remediation



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1. Introduction

Several anthropogenic activities, including industrial waste discharge, mining activities, unplanned municipal waste disposal, use of extensive pesticides, and chemical fertilizers, are responsible for heavy metals accumulation in terrestrial environment and natural water resources. Excessive accumulation of these heavy metals leads to deterioration of fauna and flora in the environment and serious threats to human health [1]. Chromium (Cr) is one of these environmental pollutants, mainly employed in the electroplating, dyeing, metallurgical, paint, paper, pulp production, and tannery industries. It has complex chemistry, and its solubility, mobility, and bioavailability depend on its oxidation states. Trivalent (Cr^{III}) and hexavalent (Cr^{VI}) are the naturally occurring states of Cr. Hexavalent Cr forms chromate and dichromate ions, soluble in water, and more toxic than Cr^{III} . Cr^{VI} is carcinogenic for human and toxic for plants and animals [2].

Among several industries responsible for Cr discharge and accumulation in biosphere, tanneries release a higher influx of Cr in the environment, accounting for 40% of the total industrial use [2]. Continuous discharge of huge amount of wastes from these industries, having large number of Cr salt- and Cr ion-containing compounds, cause severe Cr toxicity in soil and ground water. A surplus amount of Cr in soil induces nutrient metal interaction and disturbs plant nutrient uptake [3,4]. Higher Cr concentration in plants causes low protein contents, inhibition of enzyme activity, chlorosis, and necrosis. It also

affects several morphological, biochemical, and metabolic parameters in plants such as reduced germination, stunted plant growth, inhibition of early seedlings, and interferes with photosynthesis and low biomass production [5,6]. Cr enters in ground water by natural weathering of Cr-contaminated rocks, direct discharge from industries, and leaching from soil. The maximum discharge limit for Cr in wastewater is 1.0 mg L^{-1} and maximum permissible limit in drinking water is 0.05 mg L^{-1} [7]. Use of Cr-contaminated water for irrigation and drinking purposes results in low agricultural production and serious health issues in human beings. Therefore, it is very important to restore such contaminated soil and water resources from Cr toxicity. Previous studies reported that application of organic soil amendments such as bio-solids, manure composites, biochar, and poultry waste in soil reduce Cr^{VI} into a less toxic and less mobile state, i.e., Cr^{III} , subsequently decreasing its mobility and bioavailability [8–10].

Among these organic soil amendments, biochar (BC) has gained ample attraction of the scientific community owing to its agricultural and environmental benefits. BC, which is a solid by-product of the controlled combustion of organic waste, has a large surface area, rich porous structure, enormous oxygen containing functional groups, alkaline pH, and high cation exchange capacity (CEC) [11,12]. The porous structure of BC can hold heavy metals and several organic and inorganic pollutants. Previous studies reported positive impacts of BC soil application such as improved nitrogen fixation, reduced nutrient loss by leaching, higher crop biomass, controlled diseases, and microbe friendliness [13,14]. The ability of BC to mitigate heavy metal toxicity in water bodies, mobility in soil, and bio-availability largely depends on the source of feedstock and pyrolysis conditions such as temperature and resident time [15,16]. Generally, BC produced at high temperature ($600\text{--}700 \text{ }^\circ\text{C}$) has higher aromaticity and recalcitrant potential, but due to dehydration and deoxygenation, has a smaller number of O- and H-containing functional groups [17,18]. Meanwhile, BC produced at low temperature ($300\text{--}400 \text{ }^\circ\text{C}$) has more O- and H-containing functional groups and variety of organic characteristics [19,20]. Herath et al. [21] reported a 93–97% decrease in Cr, Ni, and Mn bioaccumulation in tomato plants by the application of BC. Application of maize stalk-derived BC to heavily Cr-contaminated soil showed an increase in soil fertility, higher nutrient uptake, and amelioration of Cr-contaminated soil [22].

Modification of BC with foreign materials such as silica, zeolites, polymers, and nutrient enrichment improves its physio chemical properties, and ameliorates its efficiency and environmental influence [23]. Polymers are extensively studied cross-linked macromolecules which are hydrophilic in nature and have carboxylic groups. Ekebafe et al. [24] reported that polymers enhance soil water holding capacity, promote plant growth and increase plant tolerance in water stress environment. Synthesized polymers have been used extensively with adsorbents for heavy metal removal since the surface properties of the adsorbents can be modified by enhancing available functional groups to improve their adsorption ability for pollutants [25]. One way to decrease heavy metals' availability to plants is by increasing binding sites for heavy metals in soil through amendments application. It is reported earlier that polymers along with BC induce large number of binding sites to hold cationic contaminants and sequester carbon from environment [24]. Polymers also help to deduce BC pH, and improve its performance in alkaline soils as well. Bai et al. [26]. (2010) found 10.9% to 11.2% decrease in pH in sandy soil using two different kind of polymers. A number of polymers have been used for Cr^{VI} removal from water and soil including glycine doped polypyrrole, 1, 2 ethylenediamine-aminated macroporous polystyrene particles [27]. Use of BC amendments along with synthetic (acrylamide, polyurethane, polyvinyl, resins) and natural polymer derivatives of algal polysaccharides has shown promising influence on immobilization of Cr^{VI} in soil [28]. However, polymer-modified jujube wood waste-BC has not yet been studied for Cr^{VI} immobilization in soil. Therefore, in the current study, BC was produced from Jujube (*Ziziphus jujube* L.) at different pyrolysis temperature ($300 \text{ }^\circ\text{C}$, $500 \text{ }^\circ\text{C}$, and $700 \text{ }^\circ\text{C}$) and subsequently polymerized to synthesize modified-BC. The pristine and modified BCs were thereafter evaluated for their performance to mitigate phytotoxic

effects of hexavalent chromium (Cr^{VI}) on wheat seed germination and plant growth. This study will focus on application of jujube wood waste derived biochar and its polymer derivatives to mitigate Cr^{VI} toxicity in tannery waste contaminated soil and to restore it for agricultural activities. Moreover, it will help to decrease Cr^{VI} mobility and bioavailability in soil, and enhance retention and plant availability of essential nutrients, which will ultimately increase plant growth and agriculture production.

2. Materials and Methods

2.1. Biochar Production, Polymerization and Characterization

The BC was produced and polymerized by following the procedure already described in our previous research work [10]. Briefly, jujube wood waste was used as a biomass (BM) to produce BC. Raw BM was collected and washed with distilled water to remove dirt particles and other impurities followed by air drying and crushing into small pieces. Afterward, BM was subjected to thermal combustion in controlled supply of oxygen at three different pyrolysis temperatures (300 °C, 500 °C, 700 °C) by using Digital Muffle furnace (Wisetherm FH14, Germany) for 4 h at a heating rate of 10 °C min^{-1} . Produced BCs were collected, cooled in a desiccator, sieved by 0.5 mm mesh, stored in an air tight container and tagged as JBC with numeric numbers to show production temperature, such as JBC-300 (BC produced at 300 °C pyrolysis temperature), JBC-500 (BC produced at 500 °C pyrolysis temperature), and JBC-700 (BC produced at 700 °C pyrolysis temperature). Later on, these freshly produced JBCs were polymerized by following Shigetomi et al. [29] method. Briefly, a specific amount of JBC was added in aqueous solution of N, N1 methylenebisacrylamide (cross-linker) and acrylamide (monomer), mixture was stirred vigorously at 70 °C for 5 h, followed by addition of potassium peroxodisulfate (initiator). Later on, the solution was filtered, and solid material was collected from the filter paper, oven dried, passed through a 0.5 mm size sieve, stored in air-tight containers, and samples were tagged as JPBC-300, JPBC-500, and JPBC-700 with respect to their production pyrolysis temperature. Produced and polymerized JBCs were characterized for physical and chemical characteristics by following standard procedures [30]. Later, all produced materials along with raw BM were subjected to proximate analysis to calculate moisture percentage, volatiles, ash contents, and residual carbon contents by following the ASTM D1762-84 [31] method. Produced and polymerized JBCs were analyzed by scanning electron microscope (SEM, EFI S50 Inspect, Netherlands) for their morphology and structure, while mineral composition and presence of surface functional groups were determined by using X-Ray diffractometer (MAXima X XRD-7000, Shimadzu, Japan), and Fourier transformation infrared (Bruker Alpha-Eco ATR-FTIR, Bruker Optics, Inc), respectively.

2.2. Soil Collection and Characterization

Soil was collected from Derab agriculture research center. Soil was collected in patches and stored in plastic bags. Later, soil was air dried and sieved by a 2 mm size sieve. Soil samples were analyzed for their physio-chemical properties by following standard procedure [30]. Bouyoucos [32] and Walkley and Black [33] methods were followed to determine soil texture and organic matter respectively. Soil samples were digested by Hossner [34] method to determine heavy metal contents by using ICP-OES (Perkin Elmer Optima 4300 DV Inductively coupled plasma-optical emission spectrometry).

2.3. Germination Experiment

A germination experiment was conducted to investigate the potential of produced and polymerized BCs in mitigating Cr^{VI} toxic effects on wheat seed germination. One hundred grams of soil was taken in petri plates and plates were irrigated by a Cr^{VI} solution of different concentrations (0, 5, 10, 20, and 40 mg L^{-1}) separately. Potassium dichromate (Labogens LG3683 Extra Pure, $\text{K}_2\text{Cr}_2\text{O}_7$) was used a source compound to prepare Cr^{VI} solutions of different concentration. Wheat seeds were provided by Plant Production Department, King Saud University, Riyadh. Subsequently 10 seeds of wheat were added

in each petri plate. Amendments (JBCs and JPBCs) were added at 1% (*w/w*) in each petri plates. The petri plates were covered and placed in growth chamber for 48 h at 25 °C, followed by cycles of 16 h light and 8 h dark for the next 72 h. Afterwards, the number of germinated seeds in each petri plate were counted and germination percentage rate was calculated. Seedlings were harvested after four weeks, fresh weight and root and shoot lengths were recorded followed by oven drying of plant samples. Later, the dry weight of oven-dried seedling samples was measured, and Cr uptake by seedlings was measured by the dry digestion method [35]. Briefly, a specific amount of plant dry biomass was heated in a muffle furnace at 550 °C to obtain ash. Later, these plant biomass ash samples were treated and extracted with diluted HNO₃ and collected filtrate was analyzed by ICP-OES to determine Cr concentrations. Then, the uptake of Cr to the seedlings was calculated based on dry biomass of seedlings. All the treatments were triplicated and a control treatment with no added amendments was also included.

2.4. Statistical Analysis

All analyses were performed at least in triplicate. One-way analysis of variance (ANOVA) and Tukey's honestly significance difference (HSD) studentized range test were applied on mean values of all treatments, using Statistix software [36].

3. Results and Discussion

3.1. Biochar and Polymer MODIFIED Biochar Characteristics

Physiochemical characters of JBCs and JPBCs have already been explained in our previous work [10]. Morphological analysis of feedstock, produced and polymerized materials showed amorphous surface of BM, while more porosity and crystallinity was found in JBC and JPBC, specifically in BCs produced at higher pyrolysis temperature (Figure 1). Amorphous surface of BM could be due to presence of volatiles and clogged impurities while presence of broad channels and more porous surface in JBCs and JPBCs specifically produced at higher thermal treatment (700 °C) indicated more degradation and decomposition of organic compounds and a higher release of moisture and volatiles clogged in the pores [37,38]. Figure 1(i–viii) indicates the penetration of the polymer matrix in the porous structure of JBC, which resulted in the formation of an extended interlocking network between the polymer matrix and the BC particles, leading to enhanced tensile strength and stability in polymerized BC [39].

The difference in the mineralogical composition of raw BM and all BCs is described in Figure 2. In BM, broad peaks indicating calcium oxalate and cellulose were found, which later shifted and diminished in JBCs due to thermal degradation of organic compounds and release of volatiles [40,41]. A similar trend of appearance of organic compounds and calcium oxalate peaks in BM and BC produced at lower thermal treatment was found in study conducted by Al-Wabel et al. [42], which later diminished by increasing pyrolysis temperature from 400 °C to 800 °C. Broad peaks at 3.58–2.93 Å range in JBC-500, JBC-700, JPBC-300, JPBC-500, and JPBC-700 indicated a crystal index of carbon which showed higher aromaticity and recalcitrance in the produced BCs [43]. Such peaks appeared again at 2.10–1.92 Å in BCs depicting condensed carbonized matrix due to higher degree of thermal treatment of raw BM. Some additional peaks were found in the JPBCs at 1.45–1.23 Å values indicating presence of quartz (SiO₂) and calcite (CaCO₃) [44,45]. FTIR spectra of BM and its derived JBCs showed band of water molecules at 3328.5 cm⁻¹, which later on faded in JBCs produced at higher pyrolysis temperature (Figure 3). C-H group stretching in BM and JBC-300 and JPBC-300 were found indicating the presence of cellulose and hemicellulose compounds. Additionally, the appearance of a band in the range of 1601–1083 cm⁻¹ in JPBCs showed presence of a C=C, C–C, and amine group (C–N) [46].

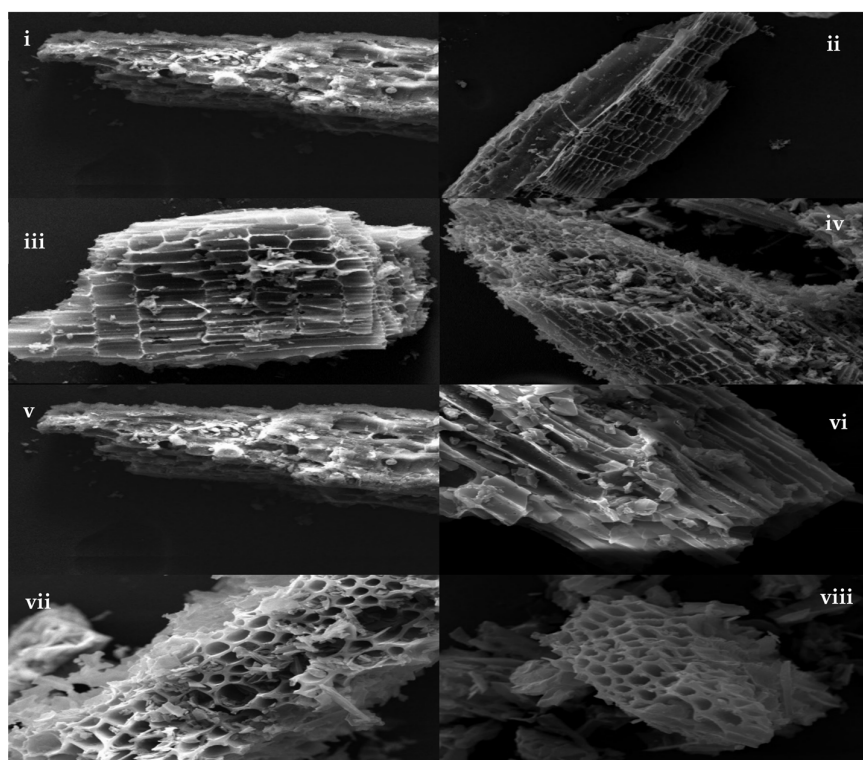


Figure 1. SEM images of the biomass (BM), biochars (JBCs) and polymerized biochars (JPBCs). (i): (BM), (ii–iv): (JBCs produced at 300, 500 and 700 °C), (v–vii): (JPBCs produced at 300, 500 and 700 °C).

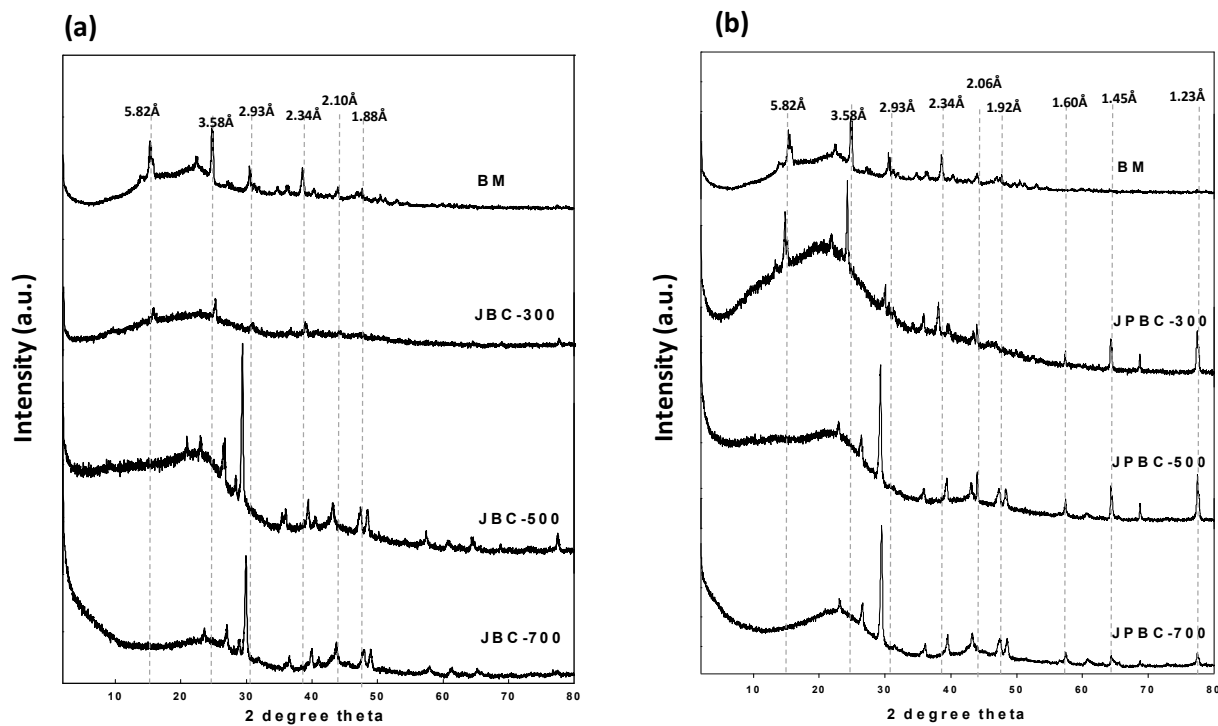


Figure 2. The X-ray diffraction analyses of the biomass (BM), biochars (JBCs) and polymerized biochars (JPBCs). (a): (BM and JBCs produced at 300, 500 and 700 °C), (b): (BM and JPBCs produced at 300, 500 and 700 °C).

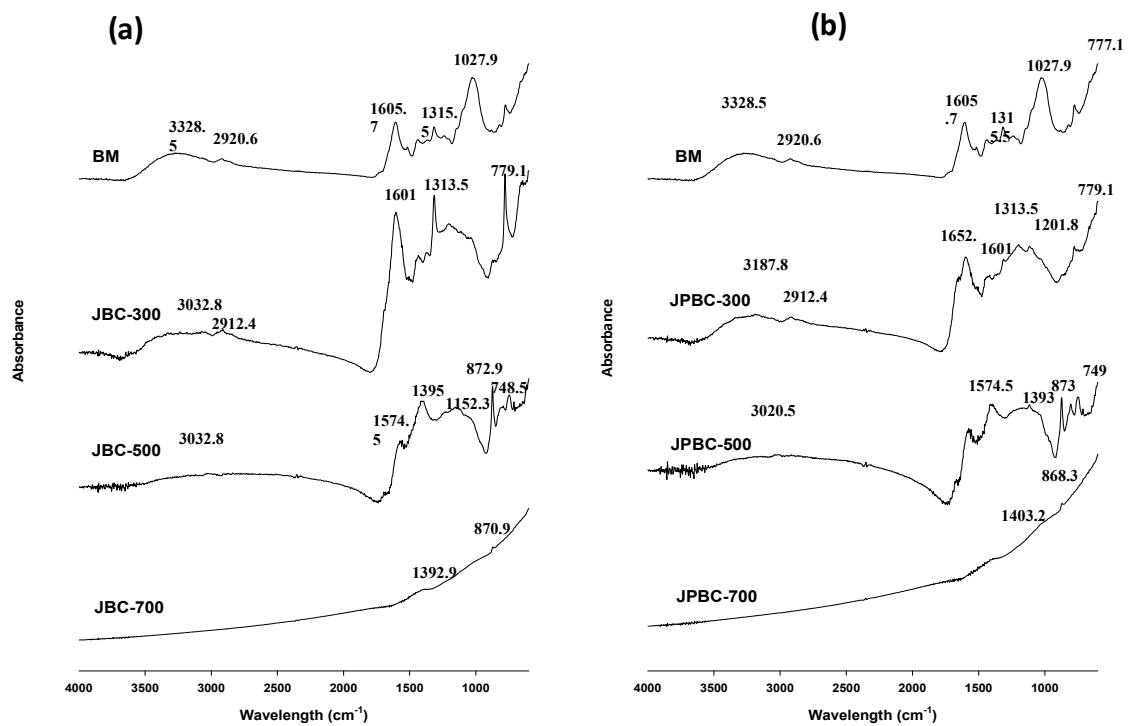


Figure 3. The FTIR analyses the biomass (BM), biochars (JBCs) and polymerized biochars (JPBCs). (a): (BM and JBCs produced at 300, 500, and 700 °C), (b): (BM and JPBCs produced at 300, 500, and 700 °C).

3.2. Germination Experiment

3.2.1. Soil Characterization

Calculated soil parameters are mentioned in Table 1. Soil was found slightly alkaline (pH 7.41) with 1.05 dS m⁻¹ electrical conductivity value. Soil texture was found sandy loam with 2.5% of mean organic matter, and 9.95 cmol kg⁻¹ of average cation exchange capacity. Soil heavy metals were found in permissible range except for Cr which was indicated high toxicity of Cr in soil (160, 270, 129, 119, and 94 mg kg⁻¹ of Cr, Fe, Mn, Zn, and Cu, respectively).

Table 1. Calculated physiochemical characteristics of soil.

Electrical Conductivity (dSm ⁻¹)	pH (1:2.5)	CEC (cmol kg ⁻¹)	Organic Matter (%)	Total Metal Concentration (mg kg ⁻¹)				
				Cr	Fe	Mn	Zn	Cu
1.05 ± 0.03	7.41 ± 1.1	9.95 ± 1.3	2.5.0 ± 0.15	160 ± 16	270 ± 14	129 ± 30	119 ± 13	94 ± 11

3.2.2. Effect of Cr^{VI} Concentration on Germination Percentage

Germination percentage was calculated by using Equation (1). With the increase in the concentration of applied Cr^{VI}, there was an obvious decrease in the germination % of seeds (Figure 4). At 0 mg L⁻¹ Cr^{VI} concentration, there was 100% germination, and no significant drop in germination was noted up to 10 mg L⁻¹ Cr^{VI} concentration. Germination decreased significantly at 20 and 40 mg L⁻¹ of Cr^{VI} concentration (Table 1). Germination dropped below 50%, indicating a 40 mg L⁻¹ Cr^{VI} concentration as a lethal dose. Toxic effects of Cr^{VI} on seed germination were recorded in the following order: 5 > 10 > 20 > 40 mg L⁻¹. These results are in agreement with earlier studies conducted to observe the inhibitory effects of Cr toxicity on seed germination [47–49]. Barcelo and Poschenrieder [50] and Panda [49] found that higher application rate of Cr^{VI} decreased the germination % by degradation of pigment, higher oxidative stress in the plant, and reducing seeds' tendency to grow. In separate studies, Rout et al. [51] and Peralta et al. [48] found 75% and 67% decreases in the

germination rate of bush bean and lucerne seed by application of Cr^{VI} at a rate of $200 \mu\text{M}$ and 40 mg L^{-1} , respectively. The curtailed ability of seed to germinate under the influence of Cr^{VI} stress might be due to the suppressive effects of Cr^{VI} on amylases activity and sugar transport to the embryo [52]. Over all, in control treatments with no added amendments, the germination % was decreased significantly as compared to other treatment with JBCs and JPBCs at same Cr^{VI} application rates (Table 2).

$$\text{Germination (\%)} = \frac{\text{No of seeds (sown)}}{\text{No of seeds (germinated)}} \times 100 \quad (1)$$

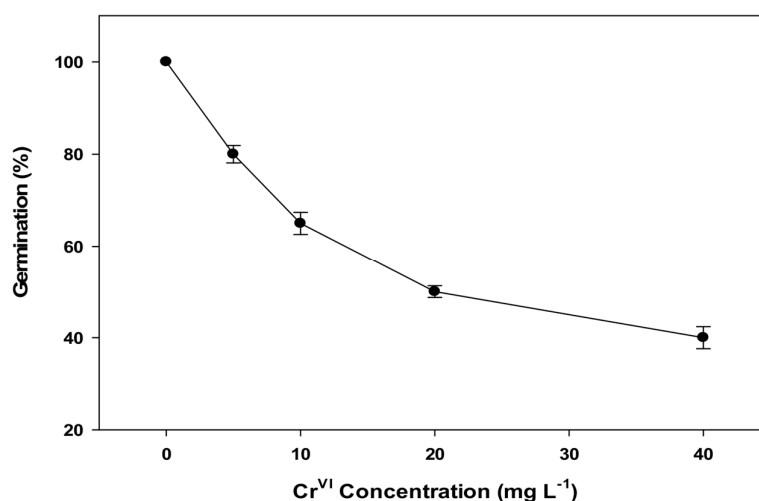


Figure 4. Germination of wheat (*Triticum aestivum* L.) seeds affected by applied concentrations of Cr^{VI} . The bars represent standard deviation.

Table 2. Effects of applied Cr^{VI} concentration on germination and seedlings growth of wheat (*Triticum aestivum* L.).

Cr^{VI} Concentration (mg L ⁻¹)	Germination %	Fresh Weight (g)	Dry Weight (g)	Shoot Length (cm)	Root Length (cm)	Cr Uptake (μg/Seedling)
0	100 ± 1.5 ^A	0.18 ± 0.08 ^A	0.13 ± 0.05 ^A	7.39 ± 1.5 ^A	7.29 ± 1.2 ^A	0.5 ± 0.02 ^C
5	80 ± 2.8 ^{AB}	0.14 ± 0.07 ^B	0.09 ± 0.02 ^B	6.15 ± 1.4 ^B	5.67 ± 0.9 ^B	7.66 ± 1.1 ^B
10	65 ± 2.2 ^{BC}	0.11 ± 0.02 ^B	0.08 ± 0.01 ^B	4.90 ± 0.9 ^C	3.94 ± 0.6 ^C	8.26 ± 1.8 ^B
20	50 ± 3.8 ^C	0.07 ± 0.01 ^C	0.04 ± 0.01 ^C	2.66 ± 0.4 ^D	1.54 ± 0.8 ^D	9.98 ± 1.5 ^B
40	40 ± 4.2 ^C	0.03 ± 0.01 ^D	0.02 ± 0.01 ^C	1.37 ± 0.6 ^E	0.84 ± 0.2 ^D	13.24 ± 1.3 ^A
Tuckey HCD	28.377	0.0311	0.0347	0.8238	0.8979	2.6961

Alpha = 0.05. Different letter show significance.

3.2.3. Effect of Cr^{VI} Concentration on Fresh and Dry Weight of Seedlings (g)

Seedlings were harvested after 4 weeks of germination and weighed instantly for their respective fresh weight followed by oven drying until constant weight and then dry weight was calculated (Figure 5). A gradual drop in fresh and dry weights of seedlings was found with increasing Cr^{VI} concentration, which indicated the toxic effects of Cr^{VI} on seedlings growth. Fresh weight was dropped considerably to 0.14, 0.11, 0.07, and 0.03 g at 5, 10, 20, and 40 mg L⁻¹ Cr^{VI} concentration, respectively, which was 0.18 g in control condition with no induced Cr^{VI} stress. A similar trend of decline in dry weight was found with the highest dry weight (0.13 g). Earlier, Fozia et al. [53] also reported a gradual decrease in fresh and dry weight of *Helianthus annuus* L. under different levels of Cr toxicity. Previous studies reported harmful effects of Cr on plants' development, including wilted aerial portions, deficient photosynthesis, and scarce mineral and fluid adsorption, resulting in stunted growth of plants and reduced plant BM [54–56]. In another study, Kumar [57] reported

reduced biomass production and leaf growth in sorghum (*sorghum bicolor* L.) due to the accumulation of Cr^{VI} which generated competition between essential nutrient uptake and oxidative injury to the plant adsorption system.

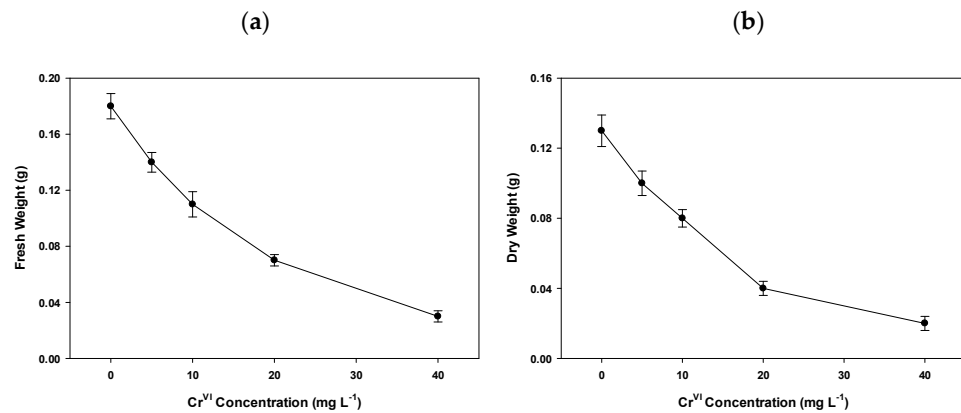


Figure 5. Fresh (a) and dry weights (b) of wheat (*Triticum aestivum* L.) seedlings affected by different concentration of Cr^{VI} . The bars represent standard deviation.

3.2.4. Effect of Cr^{VI} Concentration on Root and Shoot Lengths (cm)

Root and shoot lengths of tested crop were also affected by applied Cr^{VI} stress (Figure 6). A similar trend of decreasing lengths was found in both roots and shoots. Maximum root length (7.29 cm) was found at 0 mg L^{-1} (control) application rate of Cr^{VI} , while a gradual decrease in seedlings root length was found at 10 mg L^{-1} (3.94 cm) and 20 mg L^{-1} (1.54 cm) application rates (Table 2). Seedlings' shoot length was also decreased significantly at each application rate of Cr^{VI} , which was 7.39 cm at 0 mg L^{-1} , 4.90 cm at 10 mg L^{-1} , 2.66 cm at 20 mg L^{-1} , and 1.37 cm at 40 mg L^{-1} application rate. Jiang et al. [58] and Liu et al. [59] stated that inhibition of root and shoot elongation under heavy metal stress could be due to the metal intervention with cell division, chromosomal aberration, and asymmetrical mitosis. Samantaray et al. [60] also reported similar findings of decreased root and shoot elongation of mung bean cultivars cultivated in chromite mine-polluted soil. A number of studies have stated the negative impact of excessive Cr accumulation in plants, including decreased efficiency of chloroplast, inhibited photosynthesis, and destructive stem and roots [61–64].

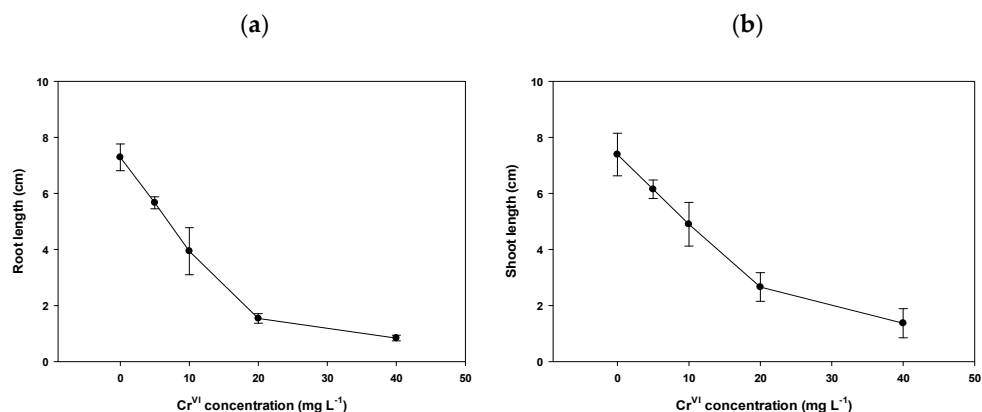


Figure 6. Root (a) and shoot (b) lengths (cm) of wheat (*Triticum aestivum* L.) seedlings affected by different concentration of Cr^{VI} . The bars represent standard deviation.

3.2.5. Effect of Charred Materials on Germination (%)

Significant increase in germination % was found in treatments with JBCs and JPBCs as compared to control treatments with no JBCs (Table 2). JPBC produced at 300 °C pyrolysis temperature (JPBC-300) showed minimum drop in germination % with each increasing rate of Cr^{VI} (Figure 7). JPBC-300 showed highest germination % (95%, 80%, 75%, and 65%)

at each applied Cr^{VI} (5, 10, 20 and 40 mg L^{-1} , respectively) stress followed by JBC-300 (90%, 80%, 70%, and 55%) and JPBC-700 (90%, 75%, 70%, and 55%, respectively). In a study to observe the effects of maize straw-derived BC on germination and growth of wheat in saline soil, Wang and Xu [63] found that wheat seed germination and overall plant growth was enhanced by BC application, while seeds germination and plant growth were found consistently low in control conditions. Addition of JBC as soil amendment immobilized toxic Cr^{VI} contents and enhanced bioavailability of essential nutrients in soil and ultimately higher plant uptake [9,10]. Bashir et al. [64] found positive effects of application of sugarcane bagasse derived BC in mitigating germination inhibition of maize cultivars grown in Cr contaminated soil. Chopala et al. [8] found delayed germination of mustard (*Brassica juncea* L.) seeds in artificially Cr-contaminated soil, while application of chicken manure derived BC avoided delayed germination and improved overall plant growth. Ruqin et al. [38] reported similar result of higher germination of spinach by application of BC with super adsorbent polymer against unmodified BC and no BC (control) treatment.

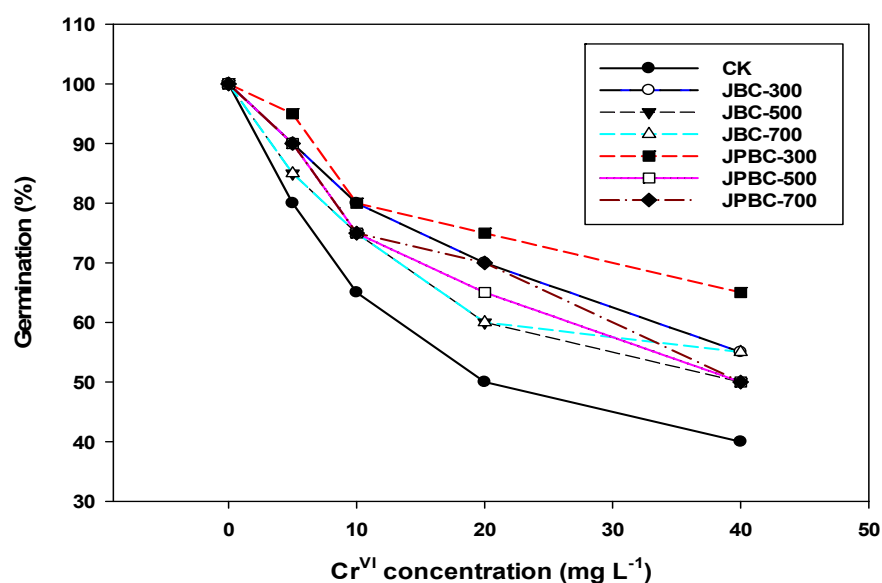


Figure 7. Effect of the biochars and polymerized biochars on germination of wheat (*Triticum aestivum* L.) seeds under the influence of applied concentrations of Cr^{VI} , JBC-300, JBC-500, and JBC-700, JPBC-300, JPBC-500, and JPBC-700 (Jujube wood waste derived and polymerized biochars produced at 300, 500, and 700 °C).

3.2.6. Effect of Charred Materials on Fresh and Dry Weight of Seedlings (g)

Application of JBCs and JPBCs showed positive effects on overall seedling growth, and minimum drop in seedling fresh and dry weights was observed under induced Cr^{VI} stress (Figure 8). Fresh biomass was increased by 30%, 40%, and 60% in treatments receiving JBC-500, JBC-700, and JBC-300 application, respectively, while further increases of 48%, 52%, and 79% were recorded in fresh weight as effected by application of JPBC-500, JPBC-700, and JPBC-300, respectively, against control treatments with no added JBCs. Comparatively higher dry weight of seedlings was found in treatment with JPBC-300 (0.07 g) followed by JBC-300, JPBC-700 (0.05 g), and JPBC-500 (0.04 g) at highest applied Cr^{VI} stress (40 mg L^{-1}). Rise in fresh and dry weights of seedlings in JBCs and JPBCs treatments against control condition showed potential of applied JBCs in improving overall plant growth. Soil immobilization of Cr^{VI} contents by JBCs alleviates competition between Cr contents and essential nutrients, which consequently enhances plant availability of nutrients. In a study, Rafique et al. [9] found 76% increase in plant fresh weight by polymer modified BC at 1.5% and 3% (*w/w*) application rate. Rajkovich et al. [65] recorded 30–40% increase in corn plant BM by application of different kind of BCs. Similar results have been reported by Wang and Xu [63] and Ruqin et al. [38] by using BC and BC modified with SAPs.

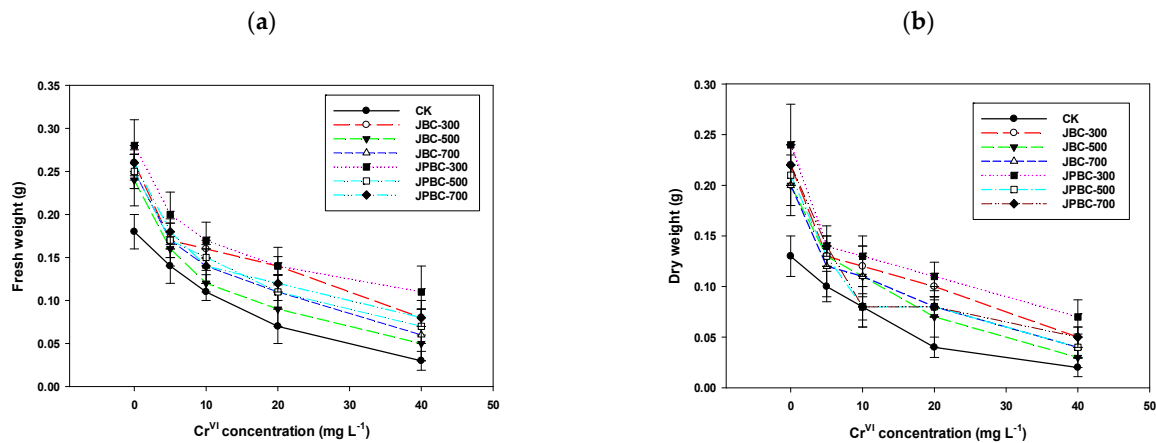


Figure 8. Effect of the biochars and polymerized biochars on (a) fresh and (b) dry weights of wheat (*Triticum aestivum* L.) seedlings under the influence of applied concentrations of Cr^{VI} . JBC-300, JBC-500, and JBC-700, JPBC-300, JPBC-500, and JPBC-700 (Jujube wood waste derived and polymerized biochars produced at 300, 500, and 700 °C).

3.2.7. Effect of Charred Materials on Root and Shoot Lengths (cm)

Alike seedling fresh and dry weights, similar trend of gradual decrease in root and shoot lengths was observed as affected Cr^{VI} induced stress (Figure 9). In comparison with control conditions, application of JBCs and JPBCs showed minimum drop in seedling root and shoot lengths (Table 3). Minimum drop in seedling growth and rise in seedling root and shoot lengths by application of JBCs and JPBCs under sever Cr^{VI} might be attributed to sorption of Cr^{VI} on charged surface of applied BCs via electrostatic interaction [22,66]. Likewise, the mineral components of BC might be involved in Cr^{VI} precipitation and reduced its bioavailability, which consequently improved essential nutrients supply and better growth of plants [67,68]. Presence of oxygenated and hydrogen functional groups on BCs surface (Figure 3) might have formed complexation with Cr^{VI} and reduced its availability [69]. Additionally, BC has ability to reduce Cr^{VI} into harmless Cr^{III} , which consequently reduced its toxic effects on plant growth [70,71].

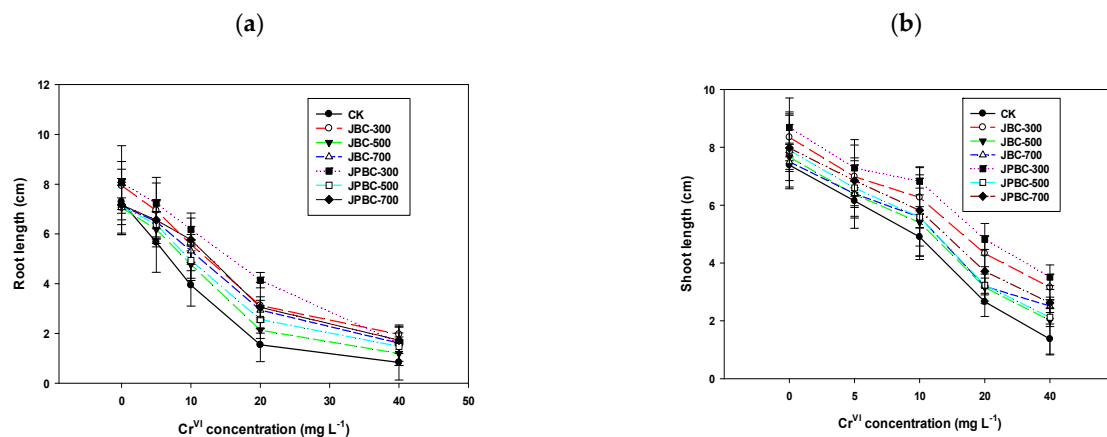


Figure 9. Effect of the biochars and polymerized biochars on (a) root and (b) shoot lengths of wheat (*Triticum aestivum* L.) seedlings under the influence of applied concentrations of Cr^{VI} . JBC-300, JBC-500, and JBC-700, JPBC-300, JPBC-500, and JPBC-700 (Jujube wood waste derived and polymerized biochars produced at 300, 500, and 700 °C).

Table 3. Effects of Jujube (*Ziziphus jujube* L.) wood waste derived biochar (JBC) and polymer modified biochar (JPBC) produced at 300, 500, and 700 °C. on germination, seedling growth and Cr uptake of wheat (*Triticum aestivum* L) under the influence of cumulative effect of applied concentration of Cr^{VI}.

Cr ^{VI} Concentration	Germination (%)	Fresh Weight (g)	Dry Weight (g)	Shoot Length (cm)	Root Length (cm)	Cr Uptake (µg/Seedling)
Control	67 ± 3.3 ^B	0.18 ± 0.04 ^E	0.075 ± 0.01 ^C	4.492 ± 0.5 ^E	3.854 ± 0.3 ^E	7.928 ± 1.1 ^A
JBC-300	79 ± 5.9 ^{AB}	0.164 ± 0.05 ^B	0.123 ± 0.01 ^{AB}	5.722 ± 0.3 ^B	5.107 ± 0.4 ^{AB}	4.275 ± 0.8 ^B
JBC-500	76 ± 5.1 ^{AB}	0.134 ± 0.02 ^D	0.108 ± 0.01 ^B	4.922 ± 0.3 ^D	4.266 ± 0.4 ^{DE}	5.184 ± 1.1 ^B
JBC-700	74 ± 2.8 ^{AB}	0.145 ± 0.01 ^{CD}	0.111 ± 0.01 ^B	5.044 ± 0.4 ^{CD}	4.502 ± 0.5 ^{CD}	4.923 ± 0.9 ^B
JPBC-300	83 ± 3.4 ^A	0.179 ± 0.03 ^A	0.139 ± 0.01 ^A	6.226 ± 0.6 ^A	5.442 ± 0.9 ^A	0.226 ± 0.03 ^C
JPBC-500	79 ± 3.6 ^{AB}	0.148 ± 0.04 ^{CD}	0.106 ± 0.02 ^B	5.087 ± 0.7 ^{CD}	4.490 ± 0.7 ^{CD}	0.396 ± 0.03 ^C
JPBC-700	77 ± 7.8 ^{AB}	0.152 ± 0.01 ^{BC}	0.111 ± 0.01 ^B	5.376 ± 0.7 ^{BC}	4.853 ± 0.8 ^{BC}	0.328 ± 0.01 ^C
Tuckey HCD	13.235	0.0148	0.0187	0.3650	0.5649	2.4014

Alpha = 0.05, Different alphabetical letters show significance level, JBC-300, JBC-500, and JBC-700 = Biochar produced at 300, 500, and 700 °C temperature, JPBC-300, JPBC-500, and JPBC-700 = Polymer modified biochar produced at 300, 500, and 700 °C temperature.

3.2.8. Effect of Charred Materials on Cr Uptake (µg/Seedling)

Effects of JBCs and JPBCs on Cr uptake at applied Cr^{VI} concentration are mentioned in Figure 10. All applied charred materials considerably decreased Cr^{VI} contents in seedlings, while in control conditions, a gradual increase in Cr^{VI} uptake was noted, which was 0.5 µg/seedling at 0 mg L⁻¹ Cr^{VI} application and increased to 13.25 µg/seedling at highest application rate of Cr^{VI} (40 mg L⁻¹) (Table 1). On the other hand, zero to very low Cr^{VI} contents were found in plants grown in JPBCs and JBCs amended treatments. Especially, in presence of JPBCs zero contents of Cr^{VI} were found in seedlings, while a minimal increase in Cr^{VI} contents was found in seedlings at 40 mg L⁻¹ application rate. Lower Cr^{VI} availability and plant accessibility showed excellent sorption ability of JBCs and JPBCs [22]. In a review study about ameliorating effects of BC on physio-chemical properties of weather soil, Glaser et al. [19] found positive effects of BC in immobilizing Cr in soil and lowering plant accessibility. Moreover, lower plant availability of Cr^{VI} as affected by applied JPBCs could be due to the presence of more active and binding sites on JPBCs' surface, which strengthens its ability in fixing heavy metals [24,25].

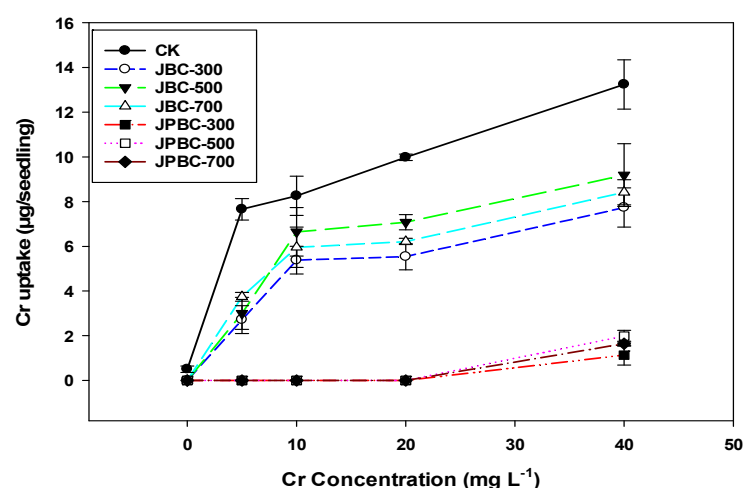


Figure 10. Effect of the biochars and polymerized biochars on Cr^{VI} uptake of wheat (*Triticum aestivum* L.) seedlings (µg/seedling) under the influence of applied concentrations of Cr^{VI}. JBC-300, JBC-500, and JBC-700, JPBC-300, JPBC-500, and JPBC-700 (Jujube wood waste derived and polymerized biochars produced at 300, 500 and 700 °C).

4. Conclusions

Jujube wood waste derived biochar (JBC) and polymer-modified JBCs (JPBCs) showed characteristic variations in their physio-chemical properties. More porosity and a higher number of functional groups were found in JPBCs, while higher pH and fixed carbon contents were recorded in unmodified JBCs. The findings of this study showed that wheat seed germination and overall seedling growth were adversely affected by induced Cr^{VI} stress, while on contrary application of charred materials (JBCs and JPBCs) showed promising effects in lowering Cr^{VI} phytotoxicity by restraining its mobility and bioavailability. In comparison to control conditions, the application of unmodified JBCs enhanced shoot length by 150%, while JPBCs increased seedling dry matter by 250%. On the other hand, JBCs and JPBCs decreased Cr^{VI} uptake in seedlings, which was 13.24 µg/seedling in the control treatment, and decreased to 7.74 µg/seedling and 1.13 µg/seedling in JBCs and JPBCs, respectively. JPBC application showed higher germination percentages, improved plant growth, and exhibited significantly lower Cr uptake in plants as compared to JBC. In general, the application of JBC and polymer-modified JBC could be a fitting strategy in decreasing phytotoxic effects of Cr^{VI} and can potentially improve overall plant growth as well.

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