



## Article Impact of Biochar Application on Germination Behavior and Early Growth of Maize Seedlings: Insights from a Growth Room Experiment

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Abstract: Reduced germination and early crop maturity due to soil compaction, nutrients stress, and low moisture are major constraints to achieve optimum crop yield, ultimately resulting in significant economic damages and food shortages. Biochar, having the potential to improve physical and chemical properties of soil, can also improve nutrients and moisture access to plants. In the present study, a growth room experiment was conducted to assess biochar influence on maize seed germination, early growth of seedlings, and its physiological attributes. Corn cob biochar (CCB) was mixed with soil at different rates (0.5%, 1%, 1.5%, 2%, 2.5%, and 3% w/w) before seed sowing. Results obtained showed that increasing CCB application rate have neutral to positive effects on seed germination and seedling growth of maize. Biochar addition at the rate of 1.5% (w/w) significantly increased shoot dry biomass (40%), root dry biomass (32%), total chlorophyll content (a and b) (55%), germination percentage (13%), seedling vigor (85%), and relative water content (RWC) (68%), in comparison to un-amended control treatment. In addition to this, it also improved germination rate (GR) by 3% as compared to control treatment, while causing a reduction in mean emergence time (MET). Moreover, application of biochar (3%) also resulted in enhancement of antioxidant enzyme activity, particularly superoxide dismutase (SOD) and catalase (CAT) by 13% and 17%, respectively. Conclusively, biochar application is an attractive approach to improve the initial phase of plant growth and provide better crop stand and essential sustainable high yields.

Keywords: biochar; germination; seedling vigor; chlorophyll; germination rate; maize growth

## 1. Introduction

Better crop yield depends on early growth and developmental phases, namely germination, seedling vigor, and plant development. These phases of plant growth are critically related with soil properties, such as porosity, moisture, and nutrient supply. In this way, germination and seedling quality have vital role on crop growth and yield throughout the cropping season [1]. Poor germination of seeds could be enhanced through scarification (mechanical method to reduce seed dormancy) and temperature treatment to seeds [2,3],



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**Copyright:** © 2021 by the authors. Licensee MDPI, Basel, Switzerland. This article is an open access article distributed under the terms and conditions of the Creative Commons Attribution (CC BY) license (https:// creativecommons.org/licenses/by/ 4.0/). and the decontaminating or screening of seeds using treatment based on magnetic fields and irradiation with light of various wavelength [4,5]. Germination rate could also be improved by soaking seeds in algal or *Cyanobacteria* aqueous culture prior to sowing [6]. Soil properties perform a key role in seed germination and growth because early crop stand is directly influenced by soil characteristics such as organic matter (OM), soil bulk density (BD), soil porosity, and soil moisture content [7]. In addition to this, improvement in soil properties also increases nutrients and moisture availability to plants, resulting in high germination percentage and improved seedling vigor of crop. Along with this, improving soil organic contents positively manipulates soil physical parameters that ultimately leads to better crop growth and enhanced productivity [8].

Biochar is a highly carbon-containing organic material produced through the process of pyrolysis of several kinds of biomasses [9–11]. Due to its complex aromatic structure, biochar is recalcitrant in nature that makes it more difficult to degrade it in soil [12]. This property of biochar is also an excellent approach to fix organic carbon in soil for longer durations by using crop residues which not only improves soil organic carbon, moisture holding capacity, and physical properties, but also reduces environmental carbon footprints [13,14]. Biochar mixing into the soil directly or indirectly affects different soil indicators such as soil structure, soil porosity [15], cation retention capacity [12], moisture retention [16], soil fertility improvement [17], retention of contaminants and xenobiotics in soil [18], and bacterial diversity [19,20]. It also improves soil physico-chemical characteristics by enhancing plant growth and productivity [21]. Although biochar is widely used in agriculture, its effect varies depending on plant variety and soil type [22], as soil type may also affect the function of biochar [23]. Several other researchers also proposed that biochar application into the soil is an attractive approach for better crop growth [24–26].

Previous research experiments performed by various scientists were more inclined to assess biochar role in soil health and crop yield when applied to soil [27,28]. Whereas the effect of biochar on early growth stages of plants, viz. seedling emergence and growth is seldom studied. Therefore, the present study was conducted with the special aim to understand the effect of biochar application as organic amendment on the germination of maize seeds and early growth of its seedlings.

#### 2. Materials and Methods

## 2.1. Production and Characterization of Biochar

Corn cobs were collected from field, crushed, and air dried to  $\leq 10\%$  (v/w) moisture level for biochar production. Crushed cobs were then subjected to pyrolysis at 350 °C temperature according to Sanchez et al. [29]. Biochar pH and electrical conductivity (ECe) was determined in distilled water in 1:20 [30], while ash and moisture contents of biochar were measured by placing open crucible in muffle furnace at 107 °C and 750 °C, respectively, till constant weight. Wet digestion method was practiced for nitrogen (N), phosphorus (P), and potassium (K) determination in biochar by using the Kjeldahl method, a UV-VIS spectrophotometer, and a flame photometer, respectively [31]. Various physico-chemical properties of biochar are presented in Table 1.

Parameters	Units	Pyrolysis Temperature (350 $^\circ$ C)
Yield	%	43–46
pH <sub>1:20</sub>	-	7.10
EC <sub>1:20</sub>	$dS m^{-1}$	0.73
Ash content	%	13.2
Moisture content	%	2.49
Cation exchange capacity (CEC)	cmol <sub>c</sub> kg <sup>-1</sup>	43–45
Carbon	%	58.23
Nitrogen	%	1.33
Phosphorus	%	0.43
Potassium	%	1.02
Sulphur	%	0.93

**Table 1.** Physicochemical properties of biochar produced at 350 °C.

#### 2.2. Germination Assay

A germination assay was practiced to evaluate biochar effect on germination of maize seeds and seedling growth. For this purpose, sandy loam soil was filled in plastic trays. Before sowing, developed biochar was manually mixed in sand at the rates of 0%, 0.5%, 1%, 1.5%, 2%, 2.5%, and 3% (w/w) in triplicates. Twelve seeds of maize (*Zea mays* L.) variety Syngenta NK-6621 were seeded in each tray to calculate germination percentage and rate. Thus, a total of 21 experimental units were assorted using completely randomized design (CRD) and the method described by International Seed Testing Association (ISTA) was used seven days after sowing (DAS). Seed germination percentage (GP), germination rate (GR), mean emergence time (MET), and seed vigor (SV) were calculated using following formulas:

Germination Percentage (GP) = 
$$({}^{SNG}/{}_{SN0}) \times 100$$

where, "SNG" represent germinated seeds in total and "SN0" represents total number of viable seeds [32].

Germination Rate = 
$$\frac{\sum N}{\sum (n \times g)}$$

where, "N" is the number of germinated seeds, "n" is the seeds germinated on growth day, "g" is the total number of germinated seeds.

Mean Emergence Time (MET) = 
$$\frac{\sum Dn}{\sum n}$$

where, "n" is the germinated seeds and "D" represents total number of days [33].

Seed Vigour 
$$(SV) = GP \times Seedling length$$

where, "GP" is germination percentage.

An equation by AOSA [34] was used to calculate emergence index (EI):

$$Emergence index = \frac{Germinated seeds}{No of days of first count} + \frac{Germinated seeds}{No of days of final count}$$

Time required for 50% emergence of seeds (E50) was determined using equation given by Farooq et al. [35]:

$$E_{50} = ti + \left[ \left( \frac{N}{2} - ni \right) \left( tj - ti \right) \ \left( nj - ni \right] \right]$$

Here, in the equation, "N" is the final number of germinating seeds, while nj and ni are the cumulative number of seeds germinated by adjacent counts at times tj and ti, respectively, when ni < N/2 < nj.

## 2.3. Determination of Agronomic Parameters of Maize Seedlings

After 20 days of seedling growth, the experiment was terminated and data regarding agronomic parameters, such as seedling length, fresh and dry weight, etc., were recorded considering equal number of germinated seeds (six seeds) for all the treatments with and without biochar application.

## 2.4. Physiological and Biochemical Attributes

Freshly harvested maize seedlings were subjected to study various physiological attributes, viz. chlorophyll a and b contents, relative water contents (RWC), superoxide dismutase, and catalase activity.

The Arnon [36] method was used for chlorophyll a and b determination in leaf samples. For this purpose, 0.5 g fresh leaf sample was taken from every treatment and homogenized with 80% acetone, and left for 24 h at 0.4 °C, filtered, and stored for analysis. Chlorophyll a and b contents in leaf extract was determined using a UV-visible spectrometer (Shimadzu UV-1201) at a wavelength of 645 and 663 nm, respectively, and were calculated in mg g<sup>-1</sup> by using the following formula [37].

$$\text{Chlorophyll}_{a} = (0.999 \text{ OD}_{663} - 0.099 \text{ OD}_{645})$$

$$\text{Chlorophyll}_{h} = (-0.328 \text{ OD}_{663} + 1.770 \text{ OD}_{645})$$

Relative water contents (RWC) were also measured by using the formula given below:

Relative water content % = 
$$\frac{\text{fresh wt} - \text{dry wt}}{\text{turgid wt} - \text{dry wt}} \times 100$$

The Elavarthi and Martin [38] method was used to determine antioxidant (catalase and superoxide dismutase) activity in leaf samples.

#### 2.5. Statistical Analysis of Data

Results of this experiment were statistically analysed using statistical software Statistix 8.1<sup>®</sup> (Analytical Software, Tallahassee, FL, USA). Tukey's multiple comparison test ( $p \le 0.05$ ) was used for comparing treatment means at 95% confidence interval. Principal component analysis was performed by using R software packages.

## 3. Results

## 3.1. Impact of Biochar on the Germination of Maize Seeds

Significant ( $p \le 0.05$ ) improvement in mean emergence time, germination energy, and emergence index were noticed with biochar application, at the rate of 1.5 and 2.5% (Figure 1). Germination of maize seeds started on third day, and the highest number of seeds (eight seeds) were germinated under treatment where biochar was applied at 1 and 1.5% rate, compared to the control, where six seeds germinated without biochar. Seed germination percentage was maximum (97%) with 1%, 1.5%, 2% and 2.5% biochar application (Figure 2). Seedling vigor significantly ( $p \le 0.05$ ) increased (85%) with 1.5% biochar compared to the control (Figure 3).



**Figure 1.** Effect of biochar rates on cumulative utilization energy (CUE) (**a**), emergence index (**b**), mean emergence time (**c**), and germination energy (**d**) of maize seedlings. Data presented is average of three repeats. Means sharing the similar letters do not differ significantly ( $p \ge 0.05$ ).



**Figure 2.** Effect of different biochar rates on (**A**) seedling vigor and (**B**)  $E_{50}$ . Data is the average of three repeats. Error bars showing significance of treatment ( $p \le 0.05$ ).



Figure 3. (A) Effect of different biochar rates on number of germination seeds per day. (B) Effect of different biochar rates on germination percentage of maize seeds. Here, the data presented is the average of three replicates of each treatment.

# 3.2. Impact of Biochar on Morpho-Physiological Attributes and Enzyme Activity of Maize Seedlings 3.2.1. Morphological Attributes

Results regarding maize seedlings growth (Table 2) revealed that biochar application to soil influenced maize seedlings positively. Seedling length was increased significantly ( $p \le 0.05$ ) at a lower rate of biochar, and maximum shoot length was 34 cm at 1.5% biochar, which was 46% more than control; conversely, higher rates of biochar (>1.5%) showed non-significant effects on shoot length, as compared to the control (Table 2). Fresh (1.09 g) and dry weight (0.089 g) of seedlings shoots was the highest with 1.5 and 2% biochar application, while it was 0.67 g and 0.063 g, in control. However, biochar application at higher rates (>2%) inhibited seedlings root length, while lower rates (<2%) increased root length. The maximum root length (27.33 cm) was observed with biochar application at the rate of 1.5%. Moreover, this increased root length was statistically at par where 1% and 2% biochar was applied (Table 2). Significant ( $p \le 0.05$ ) increase in root fresh and dry mass of maize seedlings was observed with the application of biochar when applied at the rate of 1.5 and 2%, respectively, although higher rate of biochar addition showed no-significant difference between fresh and dry mass of seedling roots, as compared to the control treatment (without biochar).

Table 2. Biochar effects on maize seedling growth parameters.

Biochar Level – (%)		Seedlings Shoot		Seedlings Root			
	Length (cm)	Fresh Weight (g)	Dry Weight (g)	Length (cm)	Fresh Weight (g)	Dry Weight (g)	
Control	$23.33 \pm 1.45 d$	$0.67\pm0.02d$	$0.063\pm0.002e$	$13.33 \pm 1.45 d$	$0.37\pm0.02d$	$0.090\pm0.004d$	
0.5	$28.33 \pm 1.67 \text{bc}$	$0.85\pm0.05c$	$0.074\pm0.004cd$	$21.00\pm1.53 bc$	$0.47\pm0.02 bc$	$0.098\pm0.005cd$	
1.0	$32.33\pm0.88ab$	$1.01\pm0.02ab$	$0.085\pm0.002ab$	$26.67 \pm 1.67 a$	$0.52\pm0.02ab$	$0.114\pm0.003ab$	
1.5	$34.00\pm0.58a$	$1.09\pm0.05a$	$0.088\pm0.004 ab$	$27.33 \pm 1.20 a$	$0.57\pm0.02a$	$0.119\pm0.002ab$	
2.0	$28.67 \pm 1.76 \text{bc}$	$1.08\pm0.07 \mathrm{ab}$	$0.089\pm0.003a$	$25.00 \pm 1.15 ab$	$0.52\pm0.05 \mathrm{ab}$	$0.122\pm0.003a$	
2.5	$26.33 \pm 1.20 cd$	$1.03\pm0.06ab$	$0.078\pm0.003 bc$	$20.33 \pm 1.33 c$	$0.42 \pm 0.03$ cd	$0.107\pm0.004 bc$	
3.0	$25.67\pm2.03cd$	$0.94\pm0.03 bc$	$0.063\pm0.005 de$	$18.00 \pm 1.53 \mathrm{c}$	$0.40\pm0.03cd$	$0.100\pm0.005cd$	

Values are the mean of three replicates. Means sharing similar letters do not differ significantly (at  $p \le 0.05$ ).

## 3.2.2. Physiological and Biochemical Attributes

Data regarding RWC showed that biochar application significantly ( $p \le 0.05$ ) increased RWC, and the highest (83%) was observed with 3% biochar treatment, as compared to the control treatment (Table 3). Chlorophyll a and b contents were 74% and 43% higher than control with biochar application at the rate 1.5% and 2%, respectively (Table 3). Moreover, a significant increase in total chlorophyll (chlorophyll a + b) was also observed with biochar application over the treatment set as the control.

**Table 3.** Biochar effect on relative water content (RWC), chlorophyll (a and b), and enzyme (catalase (CAT), super oxide dismutase (SOD)) activity of maize seedlings.

Biochar Level (%)	RWC (%)	Chlorophyll a mg g <sup>-1</sup> Fresh Weight	Chlorophyll b mg g <sup>-1</sup> Fresh Weight	Chlorophyll a + b	$\begin{array}{c} \text{SOD} \\ \text{U}\text{g}^{-1}\text{fw} \\ \text{min}^{-1} \end{array}$	$\begin{array}{c} \text{CAT} \\ \text{U}\ \text{g}^{-1}\ \text{fw}\ \text{h}^{-1} \end{array}$
Control	$61 \pm 2.08 d$	$1.40\pm0.11\mathrm{c}$	$0.82\pm0.02e$	$2.20\pm0.13c$	$11.93\pm0.73a$	$15.20\pm1.17 \mathrm{ab}$
0.5	$63 \pm 1.86$ cd	$1.55\pm0.07\mathrm{c}$	$0.87\pm0.03$ de	$2.40\pm0.09c$	$11.80\pm0.98\mathrm{a}$	$14.68\pm0.86\mathrm{ab}$
1.0	$67 \pm 2.31$ cd	$1.98\pm0.08\mathrm{b}$	$0.97\pm0.04$ cd	$2.95\pm0.13b$	$11.68\pm0.88a$	$13.68\pm0.88b$
1.5	$68 \pm 3.53$ bd	$2.43\pm0.11$ a	$0.99\pm0.05 \mathrm{bc}$	$3.41\pm0.16a$	$11.31\pm0.65a$	$14.33\pm0.34ab$
2.0	$71 \pm 2.96 bc$	$2.31\pm0.06a$	$1.17\pm0.04a$	$3.47\pm0.10a$	$11.66 \pm 0.32a$	$15.68\pm0.88 \mathrm{ab}$
2.5	$76 \pm 1.53$ ab	$2.30\pm0.13a$	$1.10\pm0.05 \mathrm{ab}$	$3.39\pm0.18a$	$12.34\pm0.33a$	$16.01\pm0.58 \mathrm{ab}$
3.0	$84\pm2.91a$	$2.20\pm0.08ab$	$0.92\pm0.04ce$	$3.11\pm0.07 ab$	$12.68\pm0.88a$	$16.68 \pm 1.33 a$

Data is the average of three repeats. Columns sharing similar letters do not differ significantly (at  $p \le 0.05$ ).

Data regarding measurement of antioxidant enzyme activity showed that biochar application also increased superoxide dismutase (SOD) and catalase (CAT) activity non-significantly ( $p \ge 0.05$ ), although all the treatment results were statistically significant ( $p \le 0.05$ ) at par with each other (Table 3).

#### 3.3. Principal Component and Correlation Analyses

The principal component analysis (PCA) showed the distribution of different treatments in maize crop as presented in the PCA-biplot (Figure 4). Significant results were obtained from the biplot of PCA performed for two factors (cumulative variance 83.9%), the first factor represents 56.3% variation, while 27.6% of the difference is explained by the second factor. Hence, great variation was found among all the treatments in maize plants. Here, visual impact (Figure 4) highlights the relationship and variation observed among all studied parameters of maize plant. Moreover, significant positive and negative correlations were observed between various studied parameters (Table 4).



**Figure 4.** Principal component analysis showing the correlation between the impact of biochar application on the germination of maize seeds and early growth of its seedlings, with the, treatments: T1: Control; T2: 0.5% Biochar; T3: 1.0% Biochar; T4: 1.5% Biochar; T5: 2.0% Biochar; T6: 2.5% Biochar; T7: 3.0% Biochar. The abbreviations are: GP, germination percentage; GE, germination energy; MET, mean emergence time; EI, emergence index; SV, seed vigor; RWC, relative water content; CHL, chlorophyll a and b; CAT, catalase; SOD, superoxide dismutase; SSDW, seedling shoot dry weight; SRDW, seedling root length; SSL, seedling shoot length; CUE, cumulative utilization energy.

able 4. Correlation	matrix	among	different	parameters	of maize.
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Parameter	GP	GE	MET	EI	SV	RWC	Chl (a + b)	CAT	SOD	SDW
GE	0.632 ***									
MET	-0.137 <sup>ns</sup>	$-0.121^{ns}$								
EI	0.598 ***	0.249 <sup>ns</sup>	-0.173 <sup>ns</sup>							
SV	0.756 ***	0.563 **	-0.432 **	0.509**						
RWC	0.261 <sup>ns</sup>	0.302 <sup>ns</sup>	0.287 <sup>ns</sup>	0.329 <sup>ns</sup>	0.022 <sup>ns</sup>					
Chl (a + b)	0.600 ***	0.636 ***	-0.214 ns	0.372 <sup>ns</sup>	0.583 **	0.598 ***				
CAT	-0.105 <sup>ns</sup>	0.099 <sup>ns</sup>	-0.170 ns	-0.132 <sup>ns</sup>	-0.238 ns	0.257 <sup>ns</sup>	0.178 <sup>ns</sup>			
SOD	-0.099 ns	0.079 <sup>ns</sup>	-0.166 <sup>ns</sup>	-0.236 ns	-0.148 ns	0.068 ns	-0.050 ns	0.567 **		
SDW	0.480 **	0.648 ***	-0.407 ns	0.216 <sup>ns</sup>	0.800 ***	-0.179 <sup>ns</sup>	0.502 **	-0.144 ns	-0.215 <sup>ns</sup>	
RDW	0.604 ***	0.599 ***	-0.101 <sup>ns</sup>	0.257 <sup>ns</sup>	0.711 ***	0.160 <sup>ns</sup>	0.647 ***	-0.253 <sup>ns</sup>	-0.410 ns	0.787 ***

\*\*\* shows highly significant at  $p \le 0.01$ , \*\* shows significant at  $p \le 0.05$ , ns shows non-significant at  $p \ge 0.05$ . The abbreviations are: GP, germination percentage; GE, germination energy; MET, mean emergence time; EI, emergence index; SV, seed vigor; RWC, relative water content; Chl (a + b), chlorophyll a and b; CAT, catalase; SOD, superoxide dismutase; SDW, shoot dry weight; and RDW, root dry weight.

## 4. Discussion

Biochar feedstock type, rate, and application method could play an important role in better plant response [17,39–41], due to its specific characteristics which are favorable to the soil health and could improve soil properties along with crop productivity [42,43]. However, low or high nutrient content present in biochar that could influence germination of seeds depends upon properties of biochar feedstock [44]. As biochar has high nutrient retention and water holding capacity [45–47], we supposed that soil amendment with biochar may play a potential role in germination and early seedling development. The present study showed that biochar at a lower rate (Table 2) enhanced the early growth of maize seedlings that is evident from earlier studies [15,41,46], and improved plant macro- (N, P, K) and micronutrients (Zn, Mn, Fe, B, Cu, etc.), as well as moisture retention in the soil [48]. Our findings are consistent with [49] who reported an improvement in the initial growth of shoot and root length in soil amended with biochar at a lower rate. Increase in seedling growth with biochar application is also documented by Yang et al. [50]. Similarly, rice seedling lengths of roots and shoots increased with biochar nanoparticles [51]. Marzouk [52] documented that olive pomace wastes biochar improved the root growth of date palm seedlings. Shoot and root length increment in *Coreopsis, Eschscholzia,* and *Leucanthemum* was also reported by Benjamin [53]. This is because of a positive plant growth response to applied biochar [41,54]; although, the increase in plant growth could be due to the synergistic effect of biochar and fertilizer [55], more nutrient availability [56], balanced microbes [57], and substrate with better properties [58].

In the present study, the biomass of root and shoot seedlings of maize increased under the influence of biochar addition (Table 2), which is in accordance with the previous findings of Lehmann et al. [59]. They reviewed that biochar application increased shoot and root biomass in biochar amended soil. Similarly, significant enhancement in root and shoot biomass has also been reported previously by Bu et al. [60] in *Robinia pseudoacacia* L. seedlings. However, studies conducted by [59,60] also reported reduced root to shoot ratio due to biochar addition into the soil. Conversely, here in our study, an increase in root to shoot ratio was observed, as reported by Samuel et al. [61].

Significant increase in total chlorophyll, and a and b chlorophyll, was observed in this experiment with biochar amendment as compared to the control treatment (Table 3). Reduced chlorophyll content in control treatment might be due to the reduction in specific enzyme suppression necessary for chlorophyll synthesis and reduced mineral uptake, e.g., Mg required for pigment synthesis [62]. Chlorophyll content of *Malus hupehensis Rehd*. seedlings was improved with biochar application into the soil [63]. Afsharipoor and Roosta [64] also reported improved chlorophyll content (chlorophyll a, b and total chlorophyll) in vegetables grown in organics-amended soil [65], and in *Amaranthus* as well [66].

Biochar application also played a very important role in increasing the enzyme activity; a non-significant increase in superoxide dismutase (SOD) and catalase (CAT) expression in leaves is shown in Table 3. Enhanced antioxidant activity (SOD and CAT) in seedling leaves after biochar addition to soil is also reported previously by Wang et al. [63], which could be due to the improved uptake of essential minerals or biochar trigger activities of some specific enzymes [67].

Mean emergence time (MET) also decreased in the current experiment as evident by (Figure 1). A similar finding, i.e., reduced MET, was also observed by Qayyum et al. [68], due to the utilization of vegetable waste biochar as soil amendment. Low moisture level in soil reduces the seed germination as well [69], and biochar application in our study helped to overcome this shortcoming because biochar enhances the water holding capacity of the soil and maintains the soil moisture level which promotes germination rate of seeds.

In the present study, biochar application increased seed germination and germination rate or mean emergence time (MET) in days (Figures 3 and 4). The possible reason for better germination is more water content held by biochar, as reported by Kammann et al. [46] and Basso et al. [70]. Use of organic amendments put into the soil has greater influence on moisture in growing medium that ultimately results in improved seed germination [71]; although, soluble plant growth regulators and native bacteria present in the amendment also lead to healthier germination of seeds. Here, better seed germination of maize plants could also be due to the mineral nutrients present in biochar which are released slowly, thus maintaining the fertility level of the soil to promote growth attributes [50]. Another factor that may also be the reason of better germination is the adsorption capacity of biochar. This study resembled with [72,73], who reported that biochar addition into the soil increased germination parameters. Seed germination rate was enhanced by biochar addition as organic amendment, specifically when applied at a lower rate [74]. Improved germination of Robinia pseudoacacia L. with biochar addition into the calcareous soil has also been reported [60]. Germination rate was increased significantly with biochar and other treatments compared to the control [52] at a lower rate of applied biochar because a higher rate of biochar may have some unwanted substances in olive pomace-derived

biochar, leading to a reduced germination rate [75]. Overall, biochar addition as organic amendment in soil showed a positive effect on the growth of maize seedlings by making a continuous supply of required nutrients [76–78]. Along with this, biochar also reduces the level of contaminants present in soil that otherwise hamper plant growth by causing negative impacts on both soil and plant health [78].

## 5. Conclusions

The present investigation concluded that application of biochar has a positive to neutral effect on maize seed germination and growth of its seedling. Increased seedling growth and biomass through biochar application reflected that biochar has the capacity to hold more water, and also has necessary minerals in it which are released slowly to promote plant growth or crop yield. Moreover, chlorophyll (a and b) and relative water content were also increased significantly in maize seedlings, but antioxidant activity (SOD and CAT) increased non-significantly with biochar application. Hence, biochar addition to soil as an organic amendment could be a better, easier, and more affordable option to enhance germination and early growth of maize seedlings to improve crop development and production in an environmentally friendly "green" way.

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