

Cucumber and tomato seedling growth effects by biochar supplemental rates

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Abstract

Studies on the effects of biochar on seedling emergence are limited despite the popularity of biochar. The objective of this study was to evaluate the use of biochar as a soilless media supplement for the emergence of cucumber 'Diva' and tomato 'Early Girl F1' seedlings. Eastern red cedar (ERC) biomass was produced at 300–350 °C, 400–450 °C, and 500–550 °C, and was incorporated with soilless media at rates (15%, 30%, 45%, and 60% v/v) plus a soilless media only. ERC bark was also applied at the same rates to the soilless media. Emergence rate and growth characteristics along with nutrient properties of the plant and media were recorded. For tomatoes, a lower rate of biochar (15%) produced at all three temperatures reduced the days for emergence by 12%–16% compared to 100% soilless media. Among biochar treatments, seeds emerged faster with 15% which increased by 7.1% to 16% compared to 30% and greater. Soilless media had the greatest tomato seedling height, width, shoot dry weight, and number of leaves but were similar to lower rates (up to 30%) of biochar produced at different temperatures, which outperforms the 45% and 60% rates. Whereas, for cucumber, the greatest days (10 d) required for emergence was with 60% ERC bark. Among biochar treatments, 15% biochar produced at 300–350 °C required the least days (reduced emergence days by 3 d for emergence while 45% biochar produced at the same temperature required greater days to emerge. The greatest height was with 30% biochar produced at 500–550 °C, while the greatest width was with 30% biochar produced at 300–350 °C. The 60% biochar produced at 300–350 °C had the greatest chlorophyll content. The greatest number of leaves and shoot dry weight were with 100% soilless media. In general, 100% soilless media outperforms the higher biochar mixes, however, the lower rates of biochar showed significantly similar effects as the 100% soilless media. This study found biochar produced at 45% or less at any temperature studied can be used for cucumber seedlings' emergence, while no more than 15% biochar produced at all three temperatures should be used for tomato seedling emergence.

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Introduction

Soilless media is commonly used as an alternative to traditional soil for seedling growth^[1]. Soilless media generally includes peat moss, perlite, vermiculite, fiber, and pumice, and often excludes soil^[2]. Peat moss has long been used in greenhouse production as a major growing medium^[3] because of desirable properties like high water holding capacity, low bulk density, and high air capacity at 100% water holding capacity^[4]. Peat is a finite resource, and over-extraction of peat from peatland can disturb the ecosystem and release greenhouse gases^[5]. Increasing environmental concerns and the rising price of peat moss have led to the search for cheaper and eco-friendly substrates^[2,6].

Biochar is a carbonaceous black matter produced by heating biomass wastes in anaerobic conditions^[7]. The physio-chemical properties of biochar depend upon pyrolysis temperature and the feedstock used^[8]. Biochar produced from wheat (*Triticum aestivum* L.) residue at 500–700 °C was found to be well carbonized with a greater surface area ($> 300 \text{ m}^2\text{-g}^{-1}$) in comparison with those produced at 300–400 °C ($< 200 \text{ m}^2\text{-g}^{-1}$), indicating high absorption capability for water and organic molecules^[9,10]. Similarly, Vithanage et al.^[11] explained that Quickstick (*Gliricidia sepium* Kunth.) biochar produced at greater temperatures immobilized the heavy metals and increased organic carbon and fertility of growing media. On the other hand, heavy metal concentration increased with increasing pyrolysis temperature of sewage sludge biochar from 350 to 450 °C, due to syngas volatilization^[12]. Biochar from crop and wood

feedstock exhibit greater surface area, carbon (C) content, volatile matter, lower cation exchange capacity (CEC), and low pH compared to animal litter feedstock^[13,14]. Biochar properties like high porosity, high surface area, and high pH make it suitable for horticulture uses^[15]. Biochar increases bulk density and water-holding capacity and improves the air space and total porosity of the medium^[16]. However, the effect on biochar in bulk density and water-holding capacity of growing media depends upon the type and properties of biochar and soilless media used^[17].

Peat moss amended with biochar improved the water-holding capacity of media used for container nurseries^[18]. Biochar is one of the potential potting substrates that might increase bulk density and water holding capacity, improve the air space and total porosity of the medium^[16], and improve the nutrients contained in the growing media leading to enhanced biomass production of a crop^[19]. In addition to being used as a substrate, biochar can be used for reducing climate change due to its ability to sequester carbon (C) and reduce greenhouse gas (GHG) emissions^[8]. Plant species used, biochar source, temperature, and properties determine the effect of biochar on plant growth and development^[20]. Biochar added to growing media in small amounts ($< 25\%$) promotes seedling growth and thus can be used as a growth promoter for seedling propagation in the greenhouse^[21]. Additionally, according to Vaughn et al.^[22] incorporation of biochar up to 20% v/v can be used to improve plant growth in container substrate cultivation. However, it is important to understand that the chemical and physical properties vary based on the feedstock used and pyrolysis

temperature, and all biochar may not be suitable for use in the soil-less substrate^[23,24].

Biochar rates and sources are found to have different effects on the seedling emergence and growth. For seedling growth, biochar at a rate of less than 25% mixed with soilless media like peat moss is suitable as a container substrate^[25]. With the addition of sugarcane (*Saccharum officinarum* L.) bagasse biochar to peat moss and 30% perlite, the root lengths, root surface, and number of root tips of basil (*Ocimum basilicum* L.) were found to be reduced compared to control (commercial propagation substrate)^[26]. In another seedling growth study, coconut (*Cocos nucifera* L.) shell biochar mixed at a different rate (0% to 40% v/v) with peat moss and vermiculite showed no significant effect on emergence percentage of large-flower tickseed (*Coreopsis grandiflora*), daisy (*Leucanthemum × superbum*), and California poppy (*Eschscholzia californica*)^[27]. However, Nair & Carpenter^[28] reported that in cell trays with sphagnum peatmoss-based substrates, pepper emergence increased with hardwood mixes biochar concentrations up to 60% compared to 0% and 80% biochar. Application of pine biochar produced at 800 °C enhanced seedling growth of soybean (*Glycine max* L. Merr.) compared to that produced at 400 °C and oak biochar produced at 400 and 800 °C.^[29] Thus, there is no single universal biochar feed-stock and temperature which will outperform across all types of plant and growing media. A lower emergence rate of lettuce with a higher rate of biochar (> 50%) when applied to peat moss and vermiculite was reported which was thought to be due to high metal toxicity and inhibitory action of organic compounds^[30].

The emergence of tomato (*Solanum lycopersicum* L.) seedlings decreased with wood chip biochar compared to manure^[20,31]. Huang et al.^[32] demonstrated that a mixture of 70% and 80% biochar with 30% and 20% vermicompost showed the similar emergence rates of tomato seed compared to commercial seed propagation mix, thus having the potential to substitute for commercial propagation mixture for seed propagation. Biochar applied at 15%–20% as gasified rice (*Oryza sativa* L.) hull biochar provided sufficient phosphorus (P) and potassium (K) for tomato 'Megabite' growth but not micronutrients in peat moss and perlite substrate^[33]. Another study found that biochar (woodchips, forest wood, husk, and paper fiber) concentrations greater than 25% negatively affected root and shoot growth, fresh and dry weight^[21], and yield of container-grown tomato plants^[34]. Graber et al.^[35] found that biochar blends with a soilless mixture (coconut fiber and 7:3 v/v ratio tuff) improved the height and leaf area of tomato 'cv. 1402' and setting fruits, fruit weight, and leaf area in pepper (*Capsicum annuum* L.).

Between the different sources of biochar (wood chip and manure), there was no difference in the emergence rate of cucumber 'Wisconsin SMR-58'^[20,31]. Increased fresh weight of the shoots and roots of cucumber seedlings by 10.5% and 5.8% with hydroponics exposure to biochar nanoparticles might be attributed to the release of nutrients like nitrogen (N) and P and hormones like ethylene from biochar^[36]. The greatest plant height, root length, and dry weight of cucumber 'Jinyou 1' seedlings were observed with 5% pine (*Pinus sp.* L.) biochar, and the lowest was observed at 15% biochar with peat moss^[37].

In the 1800s Eastern red cedar (ERC) (*Juniperus virginiana* L.) was considered a valued species for its various uses like oils and scents, fences, wine barrels, and windbreaks in uncultivated lands^[38]. However, in recent years, Eastern red cedar has been considered invasive because of rapid expansion replacing the native plants and animals, resulting in monoculture^[39,40]. The encroachment of ERC increases water usage altering the water yield, potentially leading to a problem of water shortage in the coming future^[41]. Eastern Red Cedar biomass can be converted into biochar through processes

such as gasification and pyrolysis which are considered promising technology^[42]. A recent report by Vaughn et al.^[22] showed that the high porosity, electrical conductivity (EC), and pH of ERC biochar make it a suitable supplement for soilless media.

Therefore, the study was conducted with the objective to evaluate the use of biochar produced by the pyrolysis process of ERC bark under different temperatures in soilless media for tomato and cucumber emergence and growth as transplants in the greenhouse. This study aims to ascertain the viability of ERC biochar as a substitute for soilless media, considering the lack of research focused on its impact on vegetable seedling emergence and growth, and the encroaching spreading growth of ERC across the USA. We hypothesize that biochar produced from Eastern red cedar at a lower rate will improve the emergence and growth traits of tomato and cucumber seedlings in soilless media.

Methods and materials

Preparation of biochar

ERC bark (Custom Wood Fibers & Cedar ERC bark, LLC., Stillwater, OK, USA) was used as a source for biochar production. A double barrel system was made to produce biochar through the pyrolysis process, with a 30-gal (0.114 m³) barrel as the inner (carbonization) barrel with a lid and a 55-gal (0.208 m³) barrel as the outer (burning) barrel^[43,44]. To improve the airflow, 32 small holes (1.27 cm) were drilled around the burning barrel (spaced 10 cm vertically and 50 cm horizontally) with one large 7 cm diameter hole at the base for the aluminum pipe to facilitate the airflow from the fan (motor horsepower 3 hp, high pressure blower, radial base of 0.3 m in wheel diameter, 230/460 voltage) (Chicago Blower Corporation, Glendale Heights, IL, USA). Two outer barrels were connected to the fan through a three-way pipe system. Likewise, the small barrel was drilled with eight equally spaced smaller holes (0.95 cm) at the base so the syngas produced during the burning could pass through. Another hole (2.12 cm) was drilled in the center of the small barrel's lid to insert a thermocouple (SZZJ INC, Shenzhen, Guangdong, China), which was then connected to a PID controller (Ink bird, Shenzhen, Guangdong, China). To start the fire, seasoned woods of size 35 cm × 15 cm from Payne Country Tree Service (Stillwater, OK, USA) were placed in between the inner and outer barrels. Biochar was produced by pyrolysis of the Eastern red cedar biomass at three temperature ranges of 300–350 °C, 400–450 °C, and 500–550 °C for 3 h after the temperature reached the desired temperature.

Plant materials and growth conditions

The research was conducted at the Department of Horticulture and Landscape Architecture Research Greenhouses of Oklahoma State University (Stillwater, OK, USA). Greenhouse temperature was set at 21.9 ± 4.1 °C. The average relative humidity of the greenhouse was 58 ± 9.2% and the daily light integral averaged 19.3 ± 0.7 std mol·m⁻²·d⁻¹ without any additional light facility.

Cucumber (*Cucumis sativus* L.) and tomato (*Solanum lycopersicum* L.) (Johnny's seed, Winslow, ME, USA) seeds were sown into two six-pack inserts (3.8 × 3.8 × 5.7 cm³ per cell) with two seeds per cell on 8 Jan 2023. The trial was repeated four times. The trays were kept in a greenhouse and hand-watered twice a day. Seedlings were irrigated with plain water for a week and then with 200 mg·L⁻¹ N with 20N-4.3P-16.6K fertilizer (EC: 1.29 mmhos) (J.R. Peter's Inc., 6656 Grant Way, Allentown, PA, USA) on a daily basis. Seedling thinning was done 10 d following sowing, to one seedling per pot.

Data collection

Seedling emergences were determined by daily counts for the first 10 d following sowing. At the time of harvest (45 d after

sowing), measurements were taken on seedling height (from top of the tray to top of the seedling), seedling width (perpendicular), chlorophyll concentration (greenness), leaf numbers, and root and shoot dry weight. Roots were washed and both root and shoot were oven-dried at 60 °C for 3 d for dry weight. The greenness was measured using a chlorophyll meter (atLEAF, FT Green LLC, Wilmington, MA, USA) by selecting the midsection of the leaf from the central part of each seedling. One sample of media per treatment per replication was submitted to the Soil, Water, and Forage Analytical Laboratory at OSU for total nutrient analysis, using a LECO TruSpec Carbon and Nitrogen Analyzer (LECO Corporation, St. Joseph, MI, USA)^[45].

Treatment and experimental design

There were a total of 17 treatments for different potting mixes [12 treatments of potting mixes with biochar (biochar produced at three different temperatures and mixed at four different ratios), four treatments of potting mixes with Eastern red cedar (ERC) barks (mixed at four different rates) plus a control (100% soilless media)]. Biochar produced at three different temperature was mixed with soilless media (Environmental Soil Solutions LLC, Stroud, OK, USA) at four ratios by volume (60%:40%, 45%:55%, 30%:70%, and 15%:85%) biochar: soilless media, and a control with only soilless media. The components of soilless media were pumice, rice hulls, compost, earthworm casting, peat, and greensand. The biochar rate for the research was chosen based on our findings from previous studies and our objective of finding the optimal rate of biochar as a supplement for the soilless media. The required volume was calculated by the specified ratio and volume of tray used. The volume of biochar was measured using a graduated cylinder according to specified ratio and the remaining volume of soilless media was also measured. Subsequently, the biochar and soilless media were mixed properly and filled in the tray for the experiment. Additionally, ERC bark at the same volume ratios as biochar was included to observe if the same rate of ERC bark will give a similar result to biochar or not. Experiment 1 was conducted from 8 Jan to 28 Mar 2023. Seedlings did not emerge properly due to the fungal growth infection thus, experiment 2 was conducted using a different soilless media from 25 Mar to 27 Jun, 2023 (BM7 45% pine bark, 5.5–6.5 pH, Berger, Saint-Modeste, Canada) for both tomato and cucumber. The physical properties of biochar and ERC bark are reported in a previous study^[46]. The experiment was conducted as a completely randomized design with 17 treatments. Six-pack inserts were considered as an experimental unit, while six seedlings within the insert as subsamples. The replication consisted of two six-pack inserts per treatment. Both experiments were replicated four times. The data from experiment 1 were excluded due to fungal infection.

Statistical analysis

Statistical analysis was performed using PROC GLIMMIX in SAS/STAT software (Version 9.4; SAS Institute, Cary, NC, USA). Assumptions for normality and homogeneity of variances were tested using the PROC UNIVARIATE in SAS 9.4. Replications over time were considered random elements, whereas different treatments for the potting mix were considered fixed effects. Residual plots were visually examined to check the appropriate designation of the distribution/link function. The distribution was normal, so no transformation was applied. Tests of significance were reported at the 0.05, 0.01, and 0.001 levels. Tukey's Honestly Significant Difference multiple comparison methods were used to determine the significant effects. A 95% confidence level was used to estimate the significant differences between means. The result represent experiment 2 only which used BM7 45% bark (Berger, Saint-Modeste, Canada) as a soilless media.

Results

Tomato

In tomato seedlings, there was a significant effect of potting mix for days to emergence, seedling height, width, number of leaves, shoot and root dry weight, and leaf greenness (Table 1). The 60% ERC bark required the greatest number of days (11) to emerge. Among biochar 45% biochar produced at 300–350 °C required greater days to emerge while 15% biochar produced at the same temperature required the least days to emerge. Increasing biochar rate increased the days required for emergence. The greatest seedling height was with 100% soilless media but was not different from 15% ERC bark, 15% and 45% biochar produced at 300–350 °C and 400–450 °C, 15% biochar produced at 500–550 °C, and 30% biochar produced at 300–350 °C. Similarly, the greatest width was with 100% soilless media and reduced by 34.14% with 60% ERC bark. The greatest number of leaves was with the 100% soilless media and biochar treatment reduced the leaves number up to 27.1% compared to 100% soilless media. Moreover, the addition of biochar and ERC bark reduced the shoot dry weight up to 63.2%, and 342.9% respectively compared to 100% soilless media. Greenness was greatest with 45% biochar produced at 500–550 °C but was only different from 30%, 45%, and 60% ERC bark. Root dry weight was greatest with 100% soilless media and was similar with 15% and 30% ERC bark, 15% biochar produced at 300–350 °C and 400–450 °C, and 15% and 30% biochar produced at 500–550 °C. Additionally, 60% ERC bark increased root-to-shoot ratio by three times compared to 100% soilless media.

Tomato media nutrients

A significant effect was seen with pH, ammonium (NH₄), phosphorous (P), potassium (K), calcium (Ca), magnesium (Mg), sulfur (S), and boron (B) (Table 2). The greatest pH (8.0) was with 60% biochar produced at 500–550 °C and 45% biochar produced at 400–450 °C and was only different from 100% soilless media, 15% ERC bark, and 30% ERC bark. The greatest NH₄, K, and Ca were with 60% ERC bark. Magnesium was greatest with 45% ERC bark and was not different from 100% soilless media and 60% ERC bark. Sulfur was greatest with 15% biochar produced at 500–550 °C and was only different from 60% ERC bark. Phosphorus content was greatest with 100% soilless media and reduced up to 67.3% with the biochar addition. Boron was greatest with 100% soilless media but was not different from 15% ERC bark, 15% and 30% biochar produced at 300–350 °C, and 15% biochar produced at 400–450 °C.

Cucumber

In cucumber seedlings, there was a significant effect for days to emergence, seedling height, width, number of leaves, shoot and root dry weight, and leaf greenness (Table 3). The 60% ERC bark showed the greatest days (10) to emergence. Among biochar 45% biochar produced at 300–350 °C required greater days for emergence while 15% biochar produced at the same temperature required the least days to emerge. 30% biochar produced at 500–550 °C increased the seedling height by 8.0% compared to 100% soilless media. The greatest width was with 30% biochar produced at 300–350 °C and was not different from 15% and 45% biochar produced at 400–450 °C, and 15% and 30% biochar produced at 500–550 °C. Greenness was greatest with 60% biochar produced at 300–350 °C and was only different from 30%, 45%, and 60% ERC bark, 15% biochar produced at 400–450 °C, and 30% and 60% biochar produced at 500–550 °C. The greatest number of leaves occurred with 100% soilless media and was similar to 15% biochar produced at all three temperatures. The addition of biochar and ERC bark reduced the shoot dry weight up to 33.3% and 37.5%

Table 1. Effect of 17 different potting mix ratios on growth and plant quality of tomato 'Early girl F1' seedlings grown in a greenhouse during spring 2023 in Stillwater, OK, USA.

Treatment (°C)	Media ratio ^z (v/v)	Emergence rate (d)	Height (cm)	Width (cm)	Greenness (unitless)	Number of leaves	Shoot dry wt (g)	Root dry wt (g)	Root-to-shoot ratio
Control	Soilless media	9.2cde ^y	27.2a	27.5a	43.6abc	9.6a	3.1a	0.4a	0.1b
ERC bark	15 ERC bark : 85 soilless media	9.1def	25.7abc	25.0h	43.8abc	7.8ab	2.3fg	0.4abc	0.2b
ERC bark	30 ERC bark : 70 soilless media	9.7bc	25.1bc	26.0e–h	43.5bc	7.0bc	1.8i	0.4a–d	0.2abc
ERC bark	45 ERC bark : 55 soilless media	9.7bcd	17.2d	23.3i	43.0c	5.6cd	1.2j	0.3d–g	0.3ab
ERC bark	60 ERC bark : 40 soilless media	11.1a	9.6e	20.5j	41.5d	4.6d	0.7k	0.3h	0.4a
300–350	15 biochar : 85 soilless media	8.4g	26.0abc	26.7b–e	44.5abc	7.7abc	2.7bcd	0.4ab	0.1b
300–350	30 biochar : 70 soilless media	9.0efg	25.3bc	25.7e–h	44.8ab	7.6abc	2.4efg	0.3e–h	0.1b
300–350	45 biochar : 55 soilless media	10.0b	25.5abc	27.3abc	44.6ab	7.4bc	2.2fg	0.3b–f	0.1b
300–350	60 biochar : 40 soilless media	9.7bc	24.5c	25.4fgh	44.9ab	7.0bc	1.9i	0.3fgh	0.2b
400–450	15 biochar : 85 soilless media	8.6fg	26.4ab	27.2abc	44.8ab	8.0ab	2.9b	0.4a	0.1b
400–450	30 biochar : 70 soilless media	9.1def	24.8bc	25.7e–h	44.7ab	7.6abc	2.5def	0.3d–g	0.1b
400–450	45 biochar : 55 soilless media	9.9b	25.5abc	27.0a–d	44.4abc	7.6abc	2.1gh	0.3c–g	0.1b
400–450	60 biochar : 40 soilless media	9.8b	24.4c	25.5fgh	44.5ab	7.0bc	1.7i	0.3fgh	0.2b
500–550	15 biochar : 85 soilless media	8.6fg	26.4ab	27.5ab	44.7ab	7.8ab	2.8bc	0.4a–d	0.1c
500–550	30 biochar : 70 soilless media	9.1ef	25.1bc	26.1d–g	44.2abc	7.5abc	2.6cde	0.4a–e	0.2bc
500–550	45 biochar : 55 soilless media	9.9b	25.3bc	26.4c–f	45.0a	7.6abc	2.2gh	0.3gh	0.1c
500–550	60 biochar : 40 soilless media	9.7b	24.8bc	25.4gh	44.1abc	7.5abc	1.9hi	0.3h	0.2bc
Significance		< 0.0001	< 0.0001	< 0.0001	< 0.0001	< 0.001	< 0.0001	< 0.0001	< 0.0001

^zERC bark and biochar were from Eastern red cedar and biochar was made using the double barrel pyrolysis system; Soilless media (BM7 45% bark, Berger, Saint-Modeste, Canada). ^yMeans (Tukey's Honestly Significant Difference) (n = 4) within a column followed by the same lowercase letter are not significantly different ($p \leq 0.05$).

Table 2. Effect of 17 different potting mix ratios on tomato 'Early girl F1' substrate nutrients grown in a greenhouse during spring 2023 in Stillwater, OK, USA.

Treatment (°C)	Media ratio ^z (v/v)	pH	NH ₄ (mg·L ⁻¹)	P (mg·L ⁻¹)	K (mg·L ⁻¹)	Ca (mg·L ⁻¹)	Mg (mg·L ⁻¹)	B (mg·L ⁻¹)	S (mg·L ⁻¹)
Control	Soilless media	7.3d ^y	1.5ab	26.8a	18.5b	37.0bc	30.5abc	0.3a	404.7ab
ERC bark	15 ERC bark : 85 soilless media	7.4cd	1.2ab	22.5ab	12.0b	38.3bc	28.3bcd	0.3ab	275.0abc
ERC bark	30 ERC bark : 70 soilless media	7.4bcd	1.1ab	14.8ab	24.3b	46.9b	28.6bcd	0.2b–g	239.1abc
ERC bark	45 ERC bark : 55 soilless media	7.6a–d	1.4ab	17.4ab	92.5ab	78.2a	44.3a	0.1c–g	300.6abc
ERC bark	60 ERC bark : 40 soilless media	7.8abc	2.1a	11.0b	119.3a	81.4a	36.6ab	0.1efg	188.9bc
300–350	15 biochar : 85 soilless media	7.7abc	1.8ab	20.8ab	15.0b	40.6bc	26.8b–e	0.3ab	367.3abc
300–350	30 biochar : 70 soilless media	7.7a–d	0.9ab	16.2ab	16.0b	34.6bc	22.7b–e	0.2a–d	334.5abc
300–350	45 biochar : 55 soilless media	7.9a	0.8ab	13.4ab	24.0b	31.1bc	18.7cde	0.2bcd	304.2abc
300–350	60 biochar : 40 soilless media	7.9ab	0.6ab	10.4b	19.8b	31.3bc	18.1cde	0.2bcd	231.9abc
400–450	15 biochar : 85 soilless media	7.7a–d	1.1ab	21.6ab	13.0b	30.8bc	20.5cde	0.2abc	301.6abc
400–450	30 biochar : 70 soilless media	7.7a–d	0.7ab	17.7ab	20.0b	29.2bc	19.7cde	0.2b–e	273.9abc
400–450	45 biochar : 55 soilless media	8.0a	0.9ab	12.4ab	19.5b	24.7bc	16.7cde	0.2b–g	283.2abc
400–450	60 biochar : 40 soilless media	7.9a	0.5ab	10.6b	44.0ab	21.2bc	15.1de	0.1d–g	163.5c
500–550	15 biochar : 85 soilless media	7.8ab	2.1a	19.2ab	14.2b	32.9bc	23.1b–e	0.2b–f	431.6a
500–550	30 biochar : 70 soilless media	7.9a	1.1ab	14.6ab	12.5b	24.5bc	15.9cde	0.1d–g	310.7abc
500–550	45 biochar : 55 soilless media	7.9ab	1.0ab	10.1b	22.8b	19.7bc	14.0de	0.1fg	291.0abc
500–550	60 biochar : 40 soilless media	8.0a	0.6ab	12.8ab	27.8b	17.1c	12.3e	0.1g	243.6abc
Significance		< 0.001	0.03	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001	0.02

^zERC bark and biochar were from Eastern red cedar and biochar was made using the double barrel pyrolysis system; Soilless media (BM7 45% bark, Berger, Saint-Modeste, Canada). ^yMeans (Tukey's Tukey's Honestly Significant Difference) (n = 4) within a column followed by the same lowercase letter are not significantly different ($p \leq 0.05$).

respectively compared to 100% soilless media. The greatest root dry weight was with 30% ERC bark and was 25% greater compared to 100% soilless media. Whereas, the greatest root-to-shoot ratio was with 60% ERC bark.

Cucumber media nutrients

The pH, ammonium (NH₄), potassium (K), and calcium (Ca) had a significant main effect (Table 4). The greatest pH was with 60% biochar produced at 500–550 °C and was only different from 100% soilless media, all ERC bark treatment, and 15% biochar produced at 400–450 °C. The greatest NH₄ was with 60% ERC bark. Potassium was greatest with 45% biochar produced at 400–450 °C and was only different from 15% and 30% ERC bark and 15% biochar produced at 400–450 °C and 500–550 °C. The greatest Ca was with 60% ERC bark which was 109.1% greater compared to 100% soilless media.

Discussion

The higher rates (45% and 60%) of biochar increased the days to emerge for tomato and cucumber seedlings. Similar to our result, Kafle et al.^[47] reported that 12.5% v/v softwood biochar increases the emergence of cucumber seedlings compared to 25% softwood biochar. 0 g/L birch (*Betula* spp. L.) wood biochar rate inhibited lettuce seed emergence compared to 20 g·L⁻¹. Similarly, Solaiman et al.^[48] reported seed emergence, growth, and emergence percentage of wheat seed was reduced at 5 g rice husk biochar per Petri dish compared to 1 g and 2.5 g, while for the same seed, days to emerge were reduced at 2.5 g and 5 g with jarrah (*Eucalyptus marginata* Donn ex Sm) biochar. This shows that the same biochar source will impact different seed emergence differently. Furthermore, Gasco et al.^[20] reported 100% cucumber seed emergence and

Table 3. Effect of 17 different potting mix ratios on growth and plant quality of cucumber 'Diva' seedling grown in a greenhouse during spring 2023 in Stillwater, OK, USA.

Treatment (°C)	Media ratio ^z (v/v)	Emergence rate (d)	Height (cm)	Width (cm)	Greenness (unitless)	Number of leaves	Shoot dry wt (g)	Root dry wt (g)	Root-to-shoot ratio
Control	Soilless media	8.0d ^y	28.9cd	20.1c–f	44.3ab	8.7a	2.4a	0.3fg	0.1d
ERC bark	15 ERC bark : 85 soilless media	8.0d	29.0cd	19.7d–g	42.4a–d	7.9bc	2.3ab	0.3c	0.1cd
ERC bark	30 ERC bark : 70 soilless media	8.8b	28.2cd	18.2h	40.5de	7.6cd	2.0c–f	0.4a	0.2bc
ERC bark	45 ERC bark : 55 soilless media	8.7bc	28.1d	14.9i	39.4e	7.0d	1.8fg	0.3ab	0.2b
ERC bark	60 ERC bark : 40 soilless media	10.0a	26.5e	15.2i	37.3f	6.4e	1.5h	0.3cd	0.2a
300–350	15 biochar : 85 soilless media	7.5d	29.9abc	20.2cde	42.8abc	8.3ab	2.1b–e	0.3c	0.1cd
300–350	30 biochar : 70 soilless media	8.1cd	30.9ab	21.8a	43.5abc	7.8bc	2.1b–e	0.3def	0.1cd
300–350	45 biochar : 55 soilless media	9.0b	29.8abc	20.1c–f	43.0abc	7.6cd	1.9c–f	0.3def	0.2cd
300–350	60 biochar : 40 soilless media	8.7bc	30.3abc	18.8gh	44.4a	7.5cd	1.6gh	0.3cd	0.2b
400–450	15 biochar : 85 soilless media	7.6d	30.1abc	21.2abc	41.9cd	8.3ab	2.3ab	0.3b	0.1cd
400–450	30 biochar : 70 soilless media	8.1cd	31.0ab	20.7bcd	42.9abc	7.9bc	2.1b–e	0.3efg	0.1cd
400–450	45 biochar : 55 soilless media	8.9b	29.8bc	20.9abc	43.1abc	7.4cd	1.9d–g	0.2g	0.1cd
400–450	60 biochar : 40 soilless media	8.8b	29.7abc	19.1e–h	43.7abc	7.4cd	2.0fg	0.3fg	0.2cd
500–550	15 biochar : 85 soilless media	7.6d	30.9ab	21.4ab	42.5a–d	8.3ab	2.2abc	0.3ab	0.1cd
500–550	30 biochar : 70 soilless media	8.0d	31.2a	20.8abc	42.3bcd	7.9bc	2.1bcd	0.3cde	0.1cd
500–550	45 biochar : 55 soilless media	8.8b	29.3abc	19.1e–g	43.5abc	7.9bc	2.2abc	0.3fg	0.1cd
500–550	60 biochar : 40 soilless media	8.7b	29.6bc	18.9fgh	42.1cd	7.4cd	1.9efg	0.3fg	0.2cd
Significance		< 0.0001	< 0.0001	< 0.0001	< 0.0001	< 0.0001	< 0.0001	< 0.0001	< 0.0001

^zERC bark and biochar were from Eastern red cedar and biochar was made using the double barrel pyrolysis system; Soilless media (BM7 45% bark, Berger, Saint-Modeste, Canada). ^yMeans (Tukey's Tukey's Honestly Significant Difference) (n = 4) within a column followed by the same lowercase letter are not significantly different ($p \leq 0.05$).

Table 4. Effect of 17 different potting mix ratios on cucumber 'Diva' substrate nutrients grown in a greenhouse during spring 2023 in Stillwater, OK, USA.

Treatment (°C)	Media ratio ^z (v/v)	pH	NH ₄ (mg·L ⁻¹)	K (mg·L ⁻¹)	Ca (mg·L ⁻¹)
Control	Soilless media	7.1e ^y	4.2b	49.5abc	28.6bc
ERC bark	15 ERC bark : 85 soilless media	7.4de	26.5ab	42.0bc	31.9abc
ERC bark	30 ERC bark : 70 soilless media	7.4cde	51.7ab	37.0c	39.2abc
ERC bark	45 ERC bark : 55 soilless media	7.4b–e	120.8ab	48.5abc	49.3ab
ERC bark	60 ERC bark : 40 soilless media	7.7a–d	155.4a	58.5abc	59.8a
300–350	15 biochar : 85 soilless media	7.6a–d	2.3ab	48.0abc	32.0abc
300–350	30 biochar : 70 soilless media	7.6a–d	35.5ab	51.0abc	35.5abc
300–350	45 biochar : 55 soilless media	7.7a–d	1.0b	63.2abc	32.5abc
300–350	60 biochar : 40 soilless media	7.8a	0.8b	94.5abc	41.6abc
400–450	15 biochar : 85 soilless media	7.4cde	1.5b	44.5bc	27.6bc
400–450	30 biochar : 70 soilless media	7.7a–d	1.6b	56.5abc	27.6bc
400–450	45 biochar : 55 soilless media	7.8a	0.9b	106.0a	34.6abc
400–450	60 biochar : 40 soilless media	7.9a	0.4b	102.3ab	22.7bc
500–550	15 biochar : 85 soilless media	7.7a–d	24.3ab	44.5bc	25.0bc
500–550	30 biochar : 70 soilless media	7.8ab	1.3b	46.5abc	21.6bc
500–550	45 biochar : 55 soilless media	7.8abc	0.5b	60.5abc	22.1bc
500–550	60 biochar : 40 soilless media	8.0a	0.5b	64.7abc	18.4c
Significance		< 0.001	0.04	0.001	< 0.001

^zERC bark and biochar were from Eastern red cedar and biochar was made using the double barrel pyrolysis system; Soilless media (BM7 45% bark, Berger, Saint-Modeste, Canada). ^yMeans (Tukey's Tukey's Honestly Significant Difference) (n = 4) within a column followed by the same lowercase letter are not significantly different ($p \leq 0.05$).

80% tomato seed emergence with sewage sludge biochar produced at 600 °C. The detrimental effects of polycyclic aromatic compounds (PAHs) present in biochar could be the reason behind the decreased emergence rate of seedlings^[26]. Guo et al.^[49] suggested that the nutrient ion imbalance due to the higher pH of biochar could affect the absorption of the minerals by seed delaying emergence.

Sun et al.^[50] reported a decrease in the emergence rate of brown top millet (*Brachiaria ramosa* might L.) attributed to the inhibition effect of the high pH of the biochar. The shoot dry weight of mung bean (*Vigna radiata* (L.) R. Wilczak) seedling was reduced at 100 t·ha⁻¹ of rice husk biochar applied to soil, whereas jarrah biochar showed no significant effect^[48]. Similarly, Vaughn et al.^[22] found reduced seedling dry weight of tomato 'Red Robin' at 20% nutrient pre-conditioned Eastern red cedar biochar application with peat-based substrate. The addition of 2 gm dry spent peat and wood forestry biochar (650 °C) per liter of peat substrate showed a

negative impact on the strawberry (*Fragaria × ananassa*) growth^[51]. Phytotoxicity with pruning residue may be caused by phenolic compounds; however, when it comes to biochar's effect, phenolic compounds alone do not explain the reduced emergence rate^[52].

Seedling height, the number of leaves, shoot dry weight, and leaf greenness decreased with the increasing rate of ERC bark, while the number of leaves, shoot dry weight, and an average width of cucumber, and tomato seedlings decreased with higher biochar rates. Chrysargyris et al.^[53] reported a decrease in lettuce height with the addition of commercial biochar at both 7.5% and 15% rates with peat moss might be related to the particle size of biochar used instead of nutrient uptake by seedlings. Increased ERC bark rate decreased the growth of weeds like eclipta (*Eclipta* L.), spotted spurge (*Euphorbia maculate* L.), and long stalked phyllanthus (*Phyllanthus tenellus* R.)^[54]. Reduced seedling growth of both tomato and cucumber seedlings with the greater ERC bark rate might be

attributed to the rapid drying of media due to the coarseness of ERC bark^[55]. In contrast to our results, Simiele et al.^[56] found an increased number of leaves and leaf area of cherry tomato with 20% orchard pruning biochar (Romagna Carbone, Bagnacavallo, Italy) produced at 500 °C to commercial soilless substrate. The increase in plant growth rate and dry weight with the greater number of leaves was associated with new cell growth due to assimilated formation. Bu et al.^[57] reported reduced rhododendron (*Rhododendron delavayi* Franch.) seedling growth in greenhouse production with wood chip biochar due to more volatile matter and PAHs (Polycyclic aromatic hydrocarbons) compared to rice husk biochar. Our results with ERC bark were similar to Khan et al.^[58], who reported that wheat straw increased the activity of dehydrogenase and β -glucosidase, breaking down the toxin present in the straw, which inhibits plant growth.

There was not much difference in biochar production temperature effect on cucumber and tomato seedling growth. Similarly, Zhang et al.^[59] found that rice straw and sawdust biochar produced at 300 °C and 500 °C showed no significant effect on tomato seedling growth. Similar to our result, Sun et al.^[50] reported that different temperature biochar (300, 450, and 600 °C) showed not much difference in brown top millet seedling height. However, a study showed that a 1% application of biochar poultry litter biochar produced at 500 °C increased the emergence rate of carrots (*Daucus carota* L.) while mixing of poultry litter biochar produced at 350 °C and pine chips biochar produced at 350 °C increased the emergence rate of lettuce^[49], suggesting different temperature biochar showed a different effect on seed emergence. The disparity in results between our study and Olszyk et al.^[31] could potentially be due to differences in feedstock and pyrolysis process used and the properties of growing media used for plant growth^[11]. This finding demonstrates that the same biochar treatment can have variable effects on different plant species.

Biochar and ERC bark treatments did not affect NO₃ concentration but affected the NH₄ concentration, while pH was similar in all biochar treatments and was only different from 100% soilless media and a lower rate of ERC bark. Our result was similar to that of Nair & Carpenter^[28], who found no significant effect of different rates of oak (*Quercus* sp. L.), elm (*Ulmus* sp. L.), and hickory (*Carya* sp. Nutt.) mixed biochar on EC until 8 weeks of sowing of pepper 'Paladin' plant. Naeem et al.^[60] reported that the effect of biochar in media pH depends upon the pyrolysis temperature of biochar and the initial pH of the growing media.

Phosphorus concentration was the greatest with 100% soilless media and decreased with a higher biochar rate, while K concentration increased with a higher biochar rate. Similar to our results, Sabatino et al.^[61] reported a decrease in P concentration and an increase in K concentration with an increase in poplar biochar rate from 20%–100% and pyrolysis temperature from 450–700 °C with Curley endive (*Cichorium endivia* L., var. *crispum* Hegi) study. Prasad et al.^[21] found that the greater rate of woodchip biochar reduced P concentration, but husk, paper fiber, forest wood, beech (*Fagus* L.), and spruce (*Picea abies* L.) mix biochar increased P availability in growing media and suggested that the difference in P availability might be attributed to the feedstock used. Khan et al.^[58] further reported that an increase in K concentration with an increase in wheat straw biochar rate from 1%–6% might be due to higher ash content and its release in media. Ca and Mg content reduced with a higher rate and temperature of biochar, in contrast with Zhang et al.^[62], which might be due to the volatilized loss of minerals during pyrolysis. Another reason associated with a decrease in Mg and Ca might be due to the antagonistic behavior of Ca, Mg, and K^[63].

Conclusions

Greater rates of biochar increase the number of days required for seed emergence as a result of phytotoxicity and high pH of growing media. The shoot dry weight of cucumber was greatest with the control and was only different from 45% and 60% ERC bark, 60% biochar produced at 300–350 °C and 500–550 °C, and 45% biochar produced at 400–450 °C. For tomatoes, greater shoot dry weight was observed with the control and 15% biochar produced at 400–450 °C. Incorporation of ERC bark reduces seedling growth compared to the same rate of biochar applied. In general, temperature did not show much significant differences in seedling growth so, lower temperature can be used for seedling growth and development. Our study suggests that a lower rate of biochar (< 15%) can be used as a supplement to soilless media for tomato seedlings, whereas any rate less than 60% can be used for cucumber seedlings. Future studies should investigate other species, the effect on transplants in the field, or if leaching of biochar before use could improve emergence and growth.

Author contributions

The authors confirm contribution to the paper as follows: conceptualization: Singh H, Dunn BL; methodology: Dunn BL, Lamichhane B; formal analysis: Singh H; investigation: Lamichhane B; data curation: Lamichhane B; writing—original draft preparation: Lamichhane B; writing—review and editing: Dunn BL, Singh H, Kumar A, Norwood B; funding acquisition: Singh H, Dunn BL. All authors reviewed the results and approved the final version of the manuscript.

Data availability

The datasets generated during and analyzed during the current study are available from the corresponding author on reasonable request.

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Conflict of interest

The authors declare that they have no conflict of interest.

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