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PAGES: 417-424

ORIGINAL PDF URL: <https://dergipark.org.tr/tr/download/article-file/1325362>

Effect of biochar applications on certain quality parameters and lettuce yield (*Lactuca sativa* L.)

Biochar uygulamalarının marul (*Lactuca sativa* L.)'un bazı kalite parametreleri ve verimi üzerine etkisi

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ARTICLE INFO

Received 02 October 2020

Received in revised form 30 November 2020

Accepted 04 December 2020

Keywords:

Biochar
Cropping season
Pyrolysis
Plant height

ABSTRACT

Biochar is increasingly used as an amendment to improve agricultural soil functions and plant growth. However, the effect of biochar application on plant growth can be different due to high variability in the quality of biochar. In this study the effect of different types of biochar application on the growth of lettuce (*Lactuca sativa* L.) was assessed in a pot experiment over two period of cultivation in greenhouse. The biochar were produced from four different feedstocks biomass [Vinyard (*Vitis vinifera* L.), Tomato (*Solanum lycopersicum* L.), Banana (*Musa*) and Carnation (*Dianthus caryophyllus* L.)] by slow pyrolysis at 300°C and 500°C temperatures and were used to amend the soil. The experiment was design in randomized complete block with five replications and 9 treatments. The treatments included the Control, VB300, TB300, BB300, CB300, VB500, TB500, BB500 and CB500. The trial consisted of a total of 45 pots (4 agricultural waste × 2 pyrolysis temperature × 5 replications) + 5 controls). Two (2) tons da⁻¹ (80 g⁻¹ 10 kg⁻¹ pot⁻¹) of biochar was applied with a basic application of fertilizers NPK (18-18-18) and calcium oxide (CaO). The biochar treatments were found to increase plant height and number of leaves in the second cropping cycle in comparison to no biochar treatments. The application of Tomato (TB300) and Banana (BB500) biochar significantly increased plant height by 15.2% and 10.2% respectively. The greatest increase due to biochar additions was found in the soils with tomato biochar treatment and the least increase was found in the soils without biochar application (Control). The second cropping season appeared to be better in terms of yield and quality parameters than the first season. As a result, in this study revealed that a variation in the temperature of pyrolysis does not impact lettuce growth, and recommends a long-term incubation period for further effect of biochar applications on crops.

MAKALE BİLGİSİ

Alınış tarihi 02 Ekim 2020

Düzeltilme tarihi 30 Kasım 2020

Kabul tarihi 04 Aralık 2020

Anahtar Kelimeler:

Biochar
Üretim sezonu
Piroliz
Bitki boyu

ÖZ

Tarım topraklarının fonksiyonlarının ve bitkisel üretimde gelişimin sağlanması bakımından biochar kullanımı artmaktadır. Ancak Biochar'ın bitki gelişimi üzerine etkisi biochar kalitesindeki yüksek çeşitlilikten dolayı farklı olabilir. Bu çalışmada farklı biyokömür uygulamalarının marul gelişimi üzerine etkisi iki farklı yetiştirme döneminde saksı denemesi olarak sera koşullarında belirlenmiştir. Biyokömler dört farklı tarımsal atıktan [Bağ budama atığı (*Vitis vinifera* L.), Domates atığı (*Solanum lycopersicum* L.), Muz Plantasyon atığı (*Musa*) ve Karanfil atığı (*Dianthus caryophyllus* L.)] 300°C ve 500°C sıcaklık değerlerindeki yavaş piroliz işlemiyle elde edilmiş ve toprağa uygulanmıştır. Çalışma, tesadüf blokları deneme desenine göre 5 tekerrürlü olarak 9 uygulama şeklinde gerçekleştirilmiştir. Uygulamalar, kontrol, VB300, TB300, BB300, CB300, VB500, TB500, BB500 ve CB500 uygulamalarını kapsamaktadır. Deneme 4 tarımsal atık × 2 piroliz sıcaklığı × 5 tekerrür + 5 kontrol olmak üzere toplamda 45 uygulama konusundan oluşmaktadır. NPK (18,18,18) ve Kalsiyum Oksit (CaO) temel gübre uygulamaları ile birlikte 10 kg toprak içeren saksılara 2 ton da⁻¹ olacak şekilde (80 g⁻¹ 10kg⁻¹saksı⁻¹) biyokömür uygulanmıştır. Araştırmanın sonunda, Domates (TB300) ve Muz (BB500) biyokömür uygulamaları sırasıyla %15.2 ve %10.2 oranında olmak üzere marul bitkisinin boyunu önemli ölçüde artırmıştır. Biyokömür uygulamalarıyla elde edilen en yüksek marul bitki boyu artışı domates biyokömür uygulaması ile en düşük ise biyokömür uygulanmayan toprakta (kontrol) elde edilmiştir. Verim ve kalite parametreleri açısından biochar uygulamaların etkisi birinci döneme göre ikinci dönemde daha belirgin olduğu ortaya çıkmıştır. Sonuç olarak, bu çalışmada piroliz sıcaklığındaki bir değişikliğin marul gelişimini etkilemediği ortaya çıkmış ve biyokömür uygulamalarının mahsuller üzerindeki daha fazla etkisi için uzun vadeli bir inkubasyon dönemi önerilmiştir.

1. Introduction

Lettuce is a significant dietary vegetable, which is principally eaten raw as salad. Utilization of lettuce has some medical advantages credited to the presence of nutrient C, phenolic compounds, and fiber content (Mulabagal et al. 2010). Thinking about the worldwide market, Spain and Italy are two of the highest level lettuce makers of the world, answerable for about 9% of worldwide lettuce (*Lactuca sativa* L.). Spain, Italy, Turkey and France are the significant lettuce-creating nations in the Mediterranean valley arriving at a yield of very nearly 3 million tons/year. Since 2000 until 2007, 77% of the complete lettuce yields in the European Association were reaped in Spain, Italy, Turkey, and France (Eurostat 2012). Nowadays biochar is commonly used in order to enhance crops yield. Biochar is a generic term whose characteristics depend on the type of biomass from which it is produced and the pyrolysis conditions (Mcbeath et al. 2015; Cha et al. 2016). Biochar is the high carbon materials created from the moderate pyrolysis (warming without oxygen) of biomass comparable in its appearance to charcoal created by regular consuming or by the ignition of biomass under oxygen-limited conditions. It is a fine grained and permeable substance (Lehmann et al. 2006; Chan et al. 2007). Biomass generally contained cellulose, hemicellulose, lignin and small amounts of volatiles mater. Whereas their ratios vary from biomass to biomass, even though biochar produced from various biomass have the same carbon ratio, their physicochemical properties can be different (Lei and Zhang 2013; Xie et al. 2014). In the literature several studies have reported the beneficial effects of biochar application on soil quality and plant productivity (Glaser et al. 2002; Krull 2006; Yamato et al. 2006; Chan et al. 2008; Torun 2018). Due to his nutrient content and release characteristics biochar can directly enhance plant productivity, an indirectly (by improving nutrients retention, improving soil pH, increasing soil cation exchange capacity, soil physical properties (Lehmann et al. 2003; Rondon et al. 2007; Chan et al. 2008; Liu et al. 2016; Kerré et al. 2017; Tian et al. 2018). However, a few examinations show yields decline (comparative with the control) if an amount of biochar is added to soil (Asai et al. 2009; Hammond et al. 2013). The objective of the study reported here was to test the quality parameters and lettuce yield by the application of different type of biochar, alongside pyrolyzed temperature of 300°C and 500°C.

2. Materials and Methods

2.1. Experiment site and soil characteristics

The research was carried out in the modern greenhouse located in Mediterranean climate zone of Akdeniz University Faculty of Agriculture. Summers are hot and dry, winters are warm and rainy. The annual average temperature is around 16.3°C whereas the average annual precipitation is 725.9 mm and most of the precipitation occurs in winter. The average of relative humidity is 63.2%. Soil samples were collected from Aksu site located in the Agriculture Research and Application of Akdeniz University in Antalya, Turkey. In order to determine the characteristics of the experiment soil, the following analysis was performed. Soil texture was determined according to the hydrometer method (Bouyoucos 1955); Walkley-Black was used to determine the organic matter (Nelson and Sommers 1982); 1: 2.5 soil water ratio was used to determine the pH and EC (Jackson 1967); Scheibler calcimeter is used in determination of soil lime content and the percentage of the soil

CaCO₃ content was calculated (Çağlar 1949). 1N ammonium acetate method was used to the cation exchange capacity (Kacar 1995); the nitrogen was determined by Kjeldahl method (Bremner 1965); Phosphorus was determined according to Olsen method (Olsen and Sommers 1982); K, Ca and Mg was extracted using ammonium acetate and read with ICP (Pratt 1965); Available Fe, Zn, Cu and Mn was extracted with a mixed solution of 0.005 M DTPA + 0.01 M CaCl₂ + 0.1 MTEA and read with ICP (Lindsay and Norvell 1978). The physicochemical properties of soil were as showed in Table 1.

Table 1. Physicochemical properties of the soil used in the study.

Soil properties	Results
pH (1:2.5)	7.7
EC dS m ⁻¹ (1:2.5)	1.96
Lime (CaCO ₃) (%)	29.8
Sand (%)	10.88
Clay (%)	46.72
Silt (%)	42.4
Texture	Clay loam
Organic matter (%)	1.94
Total nitrogen (%)	0.119
Available P (kg P ₂ O ₅ da ⁻¹)	2.62
Extractable K (kg K ₂ O da ⁻¹)	35.0
Extractable Ca (kg CaO da ⁻¹)	1407.4
Extractable Mg (kg MgO da ⁻¹)	74.4
Available Fe (ppm)	7.95
Available Mn (ppm)	5.43
Available Zn (ppm)	0.12
Available Cu (ppm)	1.70

2.2. Biochar characteristics

Four different types of feedstocks materials Vineyard (VB), Tomatoes (TB), Banana (BB) and Carnation (CB) were used to produce biochar that used in this study. Biochar were produced by slow pyrolysis system at 300°C and 500°C for 12h. Biochar were ground into a small particle size less than 1 mm using a sieve, and stored in a sealed plastic box until use. A sample of the biochar was taken for characterization. Proximate analysis was performed following ASTM D3173-03, ASTM D3175-07, ASTM D3174-02 method for moisture, volatile matter and ash respectively. Fixed carbon content was determined by difference (ASTM D3172-07a). Elemental analysis was performed using the Elemental Analyzer (CHNS-932 LECO). pH 1: 10 biochar water ratio was used to determine the pH and EC (Jindo et al. 2014). The basic physical and chemical properties of pyrolyzed biochar were showed in Table 2. Prior to planting of lettuce seedlings, biochar was mixed thoroughly into the topsoil (0-20 cm).

2.3. Experimental design

The experiment was design in factorial randomized complete block with five replications and 9 treatments. The treatments included the Control, VB300, TB300, BB300, CB300, VB500, TB500, BB500 and CB500. The pots were - 0.5 m separated from each other and they were arranged in two rows (22 and 23 pots in the first and second row respectively) on the bench inside the greenhouse. The trial consisted of a total of 45 pots (4 agricultural waste × 2 pyrolysis temperature × 5 replications) + 5 controls). Biochar was hand-applied by mixing thoroughly 10 kg of soil in the pots with biochar types at the

recommended rate of 2 tons da⁻¹. Accordingly, 80 g of biochar was applied to each soil in the pot in addition to application basic of fertilizers (NPK: 18.18.18) and calcium oxide (CaO) was performed at the recommended rate of 75 kg da⁻¹ and 25 kg da⁻¹ respectively.

2.4. Plant growth parameters

A pot was planted to one (1) lettuce seedling and therefore, only one plant from each pot was used to determine the yield and quality parameters of lettuce. At the end of the trial (two cultivation periods of 60 days for each cultivation period) plant growth parameters and yield were measured. Measurements were made in these plants and the results were given per pot by averaging them. The number of leaves (NL) per plant was found by counting leaves of the lettuce plants harvested from outside towards the inside of the plant. The plant height (PH) was measured from the point where the cotyledon leaves of lettuce join the stem to the top. The root collar diameter (RCD) was measured with a calliper just below the point where the first leaf of lettuce begins. SPAD meter was used to determine the chlorophyll (C) content of the lettuce plant. The chlorophyll amounts measured were determined separately for each potted plant.

2.5. Statistical Analysis

The multi-variate analysis was conducted on the data collected using SPSS V.17.0 (SPSS 2008). Duncan Means of treatments were separated using Duncan Multiple Range Test (DMRT) where their significant differences ($p < 0.05$) observed.

3. Results

3.1. Quality parameters of lettuce

Results of the effects of biochar applications on the number of leaves (LN), plant height (PH), root collar diameter (RCD), head length (HL) and chlorophyll (C) in two cultivation periods are presented in Table 3. The results revealed significant differences ($p < 0.05$) between the first and the second season with respect to number of leaves and plant height. However, no significant differences ($p < 0.05$) were observed between the two seasons in terms of root collar diameter, head length and chlorophyll. All the parameters except plant height had no significant effect ($p < 0.05$) on lettuce growth or quality parameters after biochar application in both seasons. Biochar application significantly affected plant height in selected

Table 2. Physical and chemical properties of biochar used in the experiment.

Parameters	Treatments							
	VB300	VB500	TB300	TB500	BB300	BB500	CB300	CB500
pH (1:10)	8.13	9.19	8.92	9.67	8.72	10.01	9.56	9.82
EC (dS m ⁻¹)	0.29	0.56	4.41	4.44	3.41	3.64	4.92	6.23
C (%)	63.57	54.75	60.00	51.62	51.75	47.69	54.00	49.93
H (%)	4.53	2.62	3.77	2.29	3.97	1.92	4.55	1.88
N (%)	1.37	0.86	1.78	1.39	0.67	1.04	3.22	2.49
O (%)	30.53	41.77	34.15	44.27	43.48	49.18	38.11	45.42
FC	58.01	73.03	40.42	46.51	41.42	54.35	33.05	40.82
Ash	5.39	8.93	23.82	30.62	17.65	25.55	23.92	32.48
VM	33.26	14.01	30.87	19.86	37.13	17.38	38.66	22.55

FC: Fix Carbon; VM: Volatil Mater. Vineyard (VB), Tomatoes (TB), Banana (BB), Carnation (CB).

Table 3. The effects of biochar applications on lettuce leaf number (LN), plant height (PH), root collar diameter (RCD), head length (HL) and chlorophyll (C).

Quality parameters										
Treatment	LN (1 plant)		PH (cm)		RCD (mm)		HL (cm)		C	
	Growth period									
	I	II	I	II	I	II	I	II	I	II
Control	44.0A	29.60B	13.72B	19.70a ¹ A ²	14.39	12.30	19.52	23.20	30.82	21.80
VB300	40.8A	31.20B	15.62B	20.60abA	12.51	12.62	18.58	24.80	33.90	27.06
TB300	41.4A	34.60B	15.32B	22.70cA	14.04	14.17	17.56	22.60	35.10	22.70
BB300	39.2A	30.80B	13.70B	21.20abcA	14.46	13.01	18.52	25.00	30.74	23.40
CB300	40.8A	36.40B	14.08B	20.80abA	14.87	15.12	19.02	26.00	27.24	24.18
VB500	40.0A	33.00B	14.14B	21.30abcA	14.28	13.61	17.68	25.40	33.54	25.38
TB500	46.2A	34.60B	11.76B	20.54abA	14.43	13.64	19.76	26.06	33.82	26.62
BB500	42.0A	32.80B	16.16B	21.70bcA	14.41	13.69	17.92	24.58	31.58	26.08
CB500	39.4A	32.40B	14.25B	20.36abA	13.97	13.87	18.68	23.70	31.42	24.76
ANOVA (One-way; LSD 5%)	NS ⁵	NS	NS	* ³	NS	NS	NS	NS	NS	NS
Season	*** ⁴		*		NS		NS		NS	

¹Means with different small letters in a column are significantly different at the 5 % level. ²Means with different capital letters in the row are significantly different at the 5 % level. ³*, significantly different at the 5 % level. ⁴***: significantly different at the 0.01 % level. ⁵NS: Not significantly different at 5 % level. Vineyard (VB), Tomatoes (TB), Banana (BB), Carnation (CB).

treatments with respect to the control in the second season. Compared to the control, plant height in TB300 and BB500 treatment increase by 15.2% and 10.2% respectively. Plant height in TB300 and BB500 treatments were significantly higher ($p < 0.05$) than other treatments. Plant height for the second cultivation period varied between 19.70-22.70 cm, and the highest value was obtained by TB300 treatment while the lowest plant height was obtained by the control.

From the Figure 1, 2, 3, 4 and 5 considering the same feedstock material but different temperature rate, the effects of biochar produced from the same feedstock material at different temperature degree (300°C and 500°C) on plant growth parameters (LN, PH, RCD, HL and C) were not significant ($p < 0.05$) between biochar produced at 300°C and 500°C during the two cultivation period.

3.2. Lettuce yield

The effect of biochar applications on the head weight (HW) and yield of lettuce in two cultivation periods are presented in Table 4. The results revealed significant differences ($p < 0.001$) between the first and the second season with respect to plant head weight. Thus, lettuce head weight was significantly higher in the second season compared to the first season. The applications of biochar on lettuce growth and yield in both cultivation periods were not found to be significant ($p < 0.05$). Noticeably, the head weight in biochar applications was higher than the head weight of the control but this was not significant ($p < 0.05$).

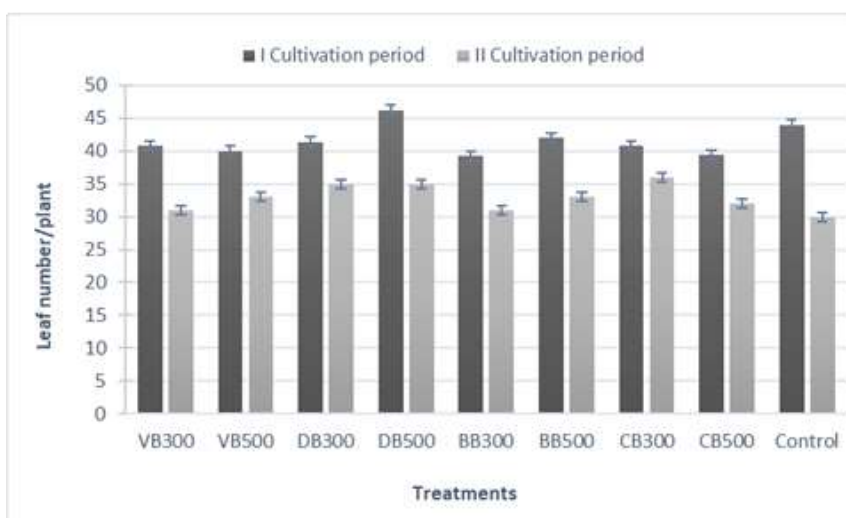


Figure 1. The effect of biochar produced from the same feedstock material at different temperature (300 and 500°C) on leaf number of lettuce during first and second cultivation period. Vineyard (VB), Tomatoes (TB), Banana (BB), Carnation (CB).

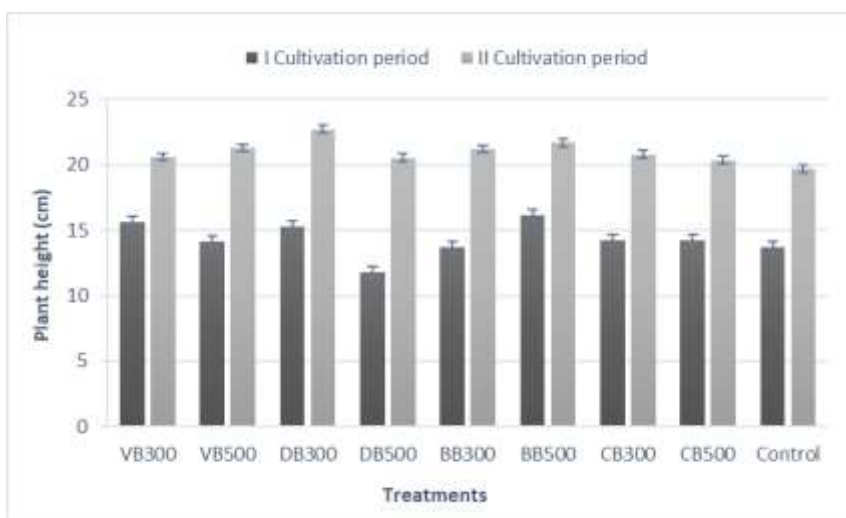


Figure 2. The effect of biochar produced from the same feedstock material at different temperature (300 and 500°C) on lettuce plant height during first and second cultivation period. Vineyard (VB), Tomatoes (TB), Banana (BB), Carnation (CB).

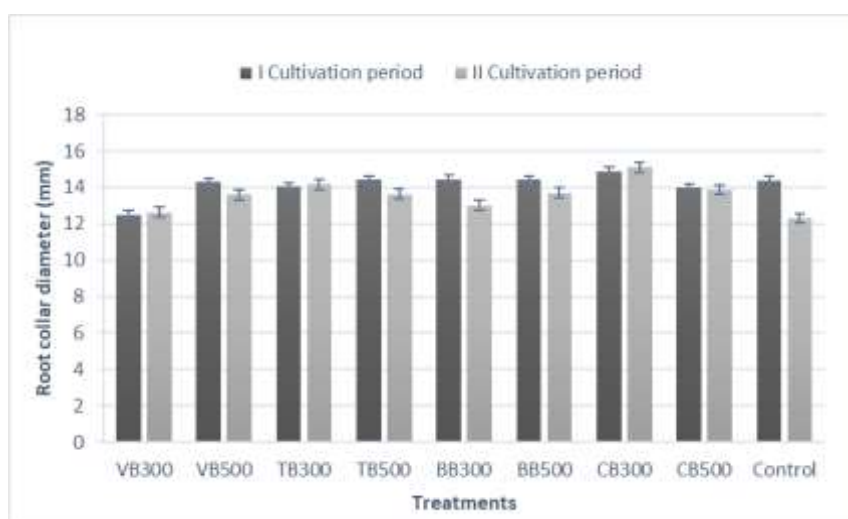


Figure 3. The effect of biochar produced from the same feedstock material at different temperature (300 and 500°C) on root collar diameter of lettuce during first and second cultivation period. Vineyard (VB), Tomatoes (TB), Banana (BB), Carnation (CB).

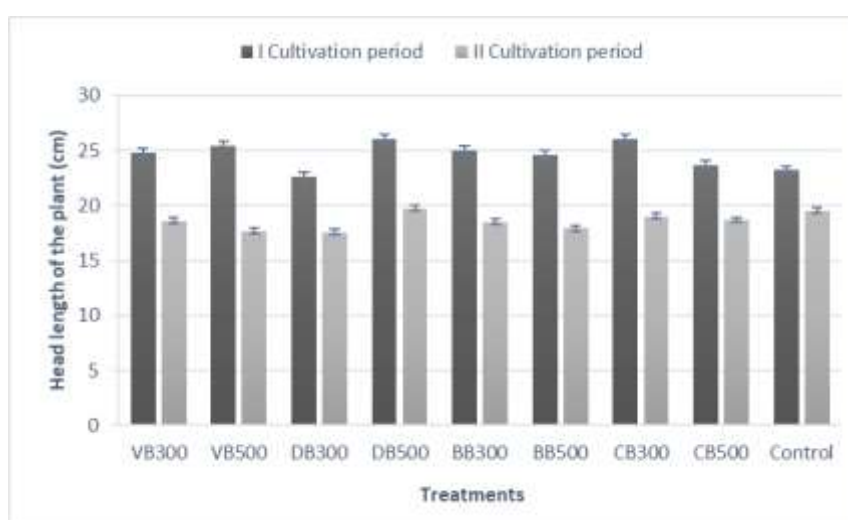


Figure 4. The effect of biochar produced from the same feedstock material at different temperature (300 and 500°C) on head length of lettuce during first and second cultivation period. Vineyard (VB), Tomatoes (TB), Banana (BB), Carnation (CB).

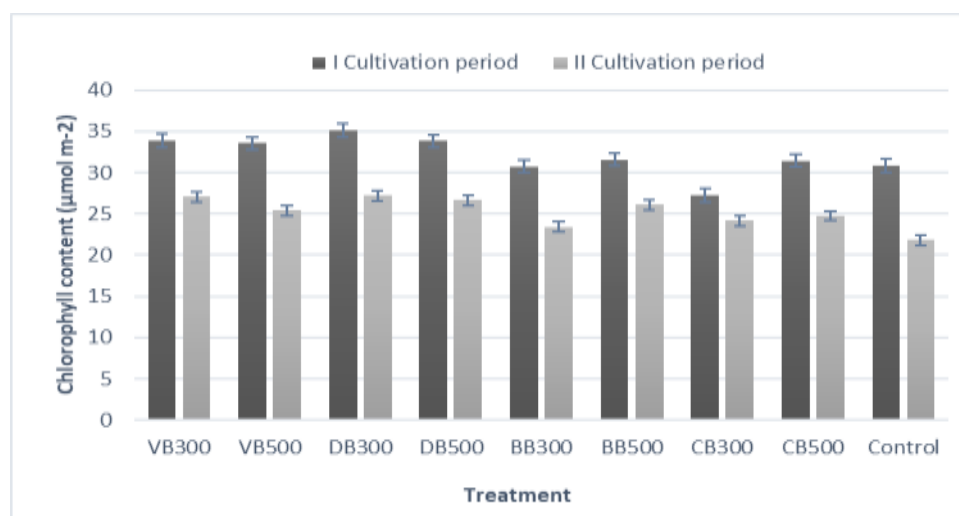


Figure 5. The effect of biochar produced from the same feedstock material at different temperature (300 and 500°C) on chlorophyll content of lettuce during first and second cultivation period. Vineyard (VB), Tomatoes (TB), Banana (BB), Carnation (CB).

Table 4. The effects of biochar applications on head weight (HW) and yield at the end of lettuce growing periods.

Treatment	HW (g plant ⁻¹)		Yield (t da ⁻¹)	
	Growth period			
	I	II	I	II
Control	94.49	173.97	0.19	0.35
VB300	92.32	187.34	0.18	0.37
TB300	81.71	221.03	0.16	0.44
BB300	89.51	205.99	0.18	0.41
CB300	92.32	221.73	0.18	0.44
VB500	67.92	190.49	0.14	0.38
TB500	101.16	201.02	0.20	0.40
BB500	93.58	217.01	0.19	0.43
CB500	82.12	188.49	0.16	0.38
ANOVA (One-way; LSD 5%)	NS	NS	NS	NS

NS: Not significant. Vineyard (VB), Tomatoes (TB), Banana (BB), Carnation (CB).

4. Discussion and Conclusion

Although biochar acts as the sink and the source of most available nutrients for the plant growth and yield, there was no significant difference in growth parameters and yield of lettuce. Our findings are consistent with the results of [Awad et al. \(2017\)](#) and [Wang et al. \(2016\)](#). From Table 3 and 4, this study observed that the application of biochar to the soil slightly increased growth and yield parameters but the effect was not significant ($p < 0.05$). These findings are in line with the results of [Hagner et al. \(2016\)](#) that revealed all tested biochar types had parallel effects on plant growth and the differences were mostly slight and short-term. The impacts of biochar application on plant development and yield boundaries rely upon the biomass type, biochar qualities and biochar application rates. It likewise relies upon the conditions and the creation condition of the plants, edaphic factors, the substance composts utilized and the assessment year ([Jeffery et al. 2011](#); [Zhang et al. 2012](#)).

The crop yield can comprehensively reflect the soil fertility. Many reports showed that biochar had a good effect on growth parameters and crop yield ([Han et al. 2012](#); [Haider et al. 2017](#)). [Chan et al. \(2007\)](#), [Asai et al. \(2009\)](#), [Lin et al. \(2015\)](#), and [Liu et al. \(2017\)](#) revealed in their examinations the beneficial outcome of biochar applications on plant creation and the exhibition of items relying upon the environment of the soil wherein the experiment is established. In any case, conflicting impacts were likewise found in past examinations or studies ([Shang et al. 2011](#); [Nelissen et al. 2015](#); [Singh et al. 2015](#); [Subedi et al. 2016](#)).

Some studies discovered that biochar application decreased yield due to different application methods and the time of application ([Singh et al. 2015](#); [Cui et al. 2017](#)). In several recent studies, it has also been reported that biochar had no significant effect on plant growth or yield ([Borchard et al. 2014](#); [Tammeorg et al. 2014](#); [Nguyen et al. 2016](#); [Al-Wabel et al. 2017](#)). Moreover, other exploration reports have likewise been distributed announcing that biochar negatively affects plant development ([Gaskin et al. 2010](#); [Lin et al. 2015](#); [Nelissen et al. 2015](#)). [Joseph et al. \(2010\)](#) reported that the interaction of biochar with environmental conditions is important to determine the contrasting effects of biochar on plant growth. Moreover, the interaction also depends on the physicochemical properties of biochar. For example, plant growth was inhibited when sandy Ultisol was amended with biochar produced at 800°C, whereas plant growth was significantly enhanced by biochar produced at 350°C ([Butnan et al. 2015](#)). Also, [Pan et al. \(2009\)](#) and

[Trupiano et al. \(2017\)](#) observed in maize and lettuce and found that paddy husk biochar increases the plant biomass of cabbage. [Ensarioğlu \(2016\)](#) examined the impact of biochar material on *Terra rosa* (Mediterranean red soil: A soil) at various portion rates (5%, 10% and 20%) and the stream-soil (B soil) gathered from the banks of the Namnam Stream bed was researched on the wheat plant. Results demonstrated the use of biochar has no impact on the vegetative (shoot length, shoot wet weight and dry weight) advancement of wheat. [Hagner et al. \(2016\)](#), after utilization of birch biochar obtained from various temperatures (300°C, 375°C, 475°C) found that the impacts on plant development of lettuce (*Lactuca sativa*), radish (*Raphanus sativus*), grain (*Hordeum vulgare*), and ryegrass (*Lolium perenne*) were similar and the difference was small. [Carter et al. \(2013\)](#), the impact on the advancement of lettuce (*Lactuca sativa*) and Chinese cabbage (*Brassica Chinensis*) with the use of biochar delivered from rice husk is very certain. [Artiola et al. \(2012\)](#) announced that essentially higher lettuce yield can be acquired contrasted with the control by applying biochar got from pine woodland squanders through moderate pyrolysis (450-500°C).

The influence of biochar obtained at different temperatures on plant growth was significant in the second cropping season. Biochar amendment significantly increased plant height but this was significant in the second season. Two out of the eight biochar treatment (i.e. TB300 and BB500) significantly increase ($p < 0.05$) plant height over the control was observed in the second season. Different biochar may have varying influences on crop performance depending on their nutrient content and the charred temperature. The stated difference can be partly attributed to the nutrient content of the original feedstock and pre-existing soil nutrient status. Nutrient-rich biochar like those produced from manure may directly supply nutrients to crops ([Rajkovich et al. 2012](#)). Rice-husk biochar tested in lettuce-cabbage-lettuce cycle increased final biomass, root biomass, plant height and the number of leaves in comparison to no biochar treatments ([Carter et al. 2013](#)). These results were also in agreement with [Riziq et al. \(2017\)](#) where it reported that biochar and bio-compost treatments showed much better growth performance compared to non-added organic amendments.

In this study, the effect different types of biochar derived from different feedstock and produced at various temperatures were investigated on lettuce growth parameters and yield of lettuce during two cultivation period. The results revealed all tested parameters (Leaf Number, Root collar diameter, Head length, Chlorophyll) except plant height had no significant

effect on lettuce growth or quality parameters after biochar application in both cultivation periods. But the biochar applications were found to increase plant height and number of leaves in the second cultivation period in comparison to no biochar treatments (control). Depending on the intrinsic properties of biochar, the differences between the treatments were mostly slight. So far, most studies have focused on the direct or indirect effects of biochar application on plants. Nevertheless, further investigations must be conducted to figure out the optimum pyrolysis temperature at which the best yield would be obtained and the optimum rate of biochar application which provide to the soil-plant system better performance.

Acknowledgement

This study has been financially supported by the Scientific Research Project Commission of Akdeniz University/Turkey with the project number of FDK-2019-4864.

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