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Article

Effect of Biochar Amendment and Organic Fertilization on the Yield and Nutritional Quality of Artichoke (*Cynara cardunculus* L.)

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Abstract: Organic fertilization is gaining importance as an alternative to chemical fertilization. However, the effects of organic products on crop quality need to be characterized, especially in biochar and derived products. Therefore, the effect of enriched-biochar supplied with an organic fertilizer (Biochar) on the yield and nutritional compounds of artichoke (*Cynara cardunculus* var. Scolymus) heads was analyzed and compared to inorganic conventional fertilization (Control). For this purpose, the number of fruits and their weights were determined, along with the nutritional properties of artichoke heads, such as mineral content, fatty acids, phenolic compounds and flavonoids. The results showed that biochar increased crop yield, as registered by the higher number of marketable or commercial artichokes compared to the Control. Additionally, biochar-treated plants exhibited significantly higher contents of Ca, Mg, and S in the artichoke heads, but a decreased content of Co, Cu, and Zn. The fatty acids content in the Biochar-treated artichokes was also significantly higher, primarily due to the presence of the palmitic acid. Conversely, the contents of phenolic compounds (e.g. flavonoids), were negatively and significantly affected by the application of Biochar. In conclusion, the application of Biochar as soil amendment in combination with organic matter for growing artichokes in the Semiarid Mediterranean area (e.g. Murcia Region) favored an increase in yield without compromising its nutritional composition. Further developments to establish protocols for the extended use of Biochar in this agri-food production area are guaranteed.

Keywords: artichoke; biochar; fatty acids; flavonoids; mineral content; phenolic compounds

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1. Introduction

According to global organizations such as United Nations Organization (UNO), the human population is projected to reach 8.5 billions of people in 2030 and 9.7 billions in 2050. To feed this growing population, food production has traditionally relied on the use of chemical fertilizers, especially those containing nitrogen (N), phosphorus (P), and potassium (K). However, the inadequate use of these fertilizers has major drawbacks, including the loss of soil biodiversity, deficiencies in soil microelements, and the alteration of the nutritional profile of vegetable, such as a decreased content of antioxidant compounds [1]. Therefore, the search for new agricultural practices and substrates that mitigate these adverse effects and serve as alternatives to synthetic fertilizers deserves further attention. Among the management practices, biochar has gained attention in recent years as an economical product that can be produced from various waste materials and used as a soil amendment.

Biochar is defined as "solid material, rich in carbon, obtained from the thermochemical decomposition of organic biomasses in an oxygen-limited environment"[2]. The characteristics of biochar depend on the organic source and the condition of pyrolysis [2]. Several studies have reported that the addition of biochar to agricultural soils promotes plant growth and development in crops such as wheat, tomato [1], rice [3], tobacco [4], Chinese cherry [5] and broccoli [6]. The beneficial

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effect of biochar on plant growth is attributed to its nutrient retention capacity, increased cation-exchange capacity (CEC), water-holding capacity, soil microbial and mycorrhizal activity [7]. Improved soil structure may affect mineral availability and absorption by plant roots. For instance, biochar application has been reported to increase the levels of essential cations such as potassium (K) and anions such as nitrate(NO_3^-), phosphate (PO_4^{3-}) and sulphate (SO_4^{2-}) in broccoli plants [6], and improve P availability and content in wheat. In radish roots, biochar derived from macroalgae enhanced the content of macronutrients such as calcium (Ca), magnesium (Mg), K, as well as molybdenum (Mo), suggesting that biochar can be used to accumulate essential nutrients in the edible part of plants [8]. However, contradictory results have been reported by Butnan et al. (2015), who observed a reduction in the content of Ca, Mg, and manganese (Mn) in maize when biochar from eucalyptus was applied [9]. Additionally, the relationship between biochar and minerals is more complex for micronutrients, with greater variability in trace nutrient concentrations found in different crops after biochar application [10].

Fatty acids are essential components of plant lipids, playing crucial roles in membrane structure, energy storage, and signaling pathways. Alterations in fatty acid composition can influence plant growth, stress tolerance, and nutritional quality [11]. Recent research suggests that biochar application can impact fatty acid metabolism in plants, offering opportunities to enhance crop productivity and nutritional value. In the aromatic plant *Nigella sativa* L., biochar significantly increased the content of linoleic acid compared to plants grown without biochar under different tillage regimes [12]. Other studies have observed changes in the fatty acid profiles according to the mode of fertilization. For example, the content of palmitic acid, stearic acid, arachidonic acid, and arachidic acid improved the quality of garden cress seed oil obtained under different fertilization treatments, including nitrogen from chicken manure and inorganic sources [13]. In maize (*Zea mays*), biochar amendment increased the content of unsaturated fatty acids (UFAs), such as linoleic acid (C18:2) and α-linolenic acid (C18:3), while decreasing saturated fatty acids (SFA) like palmitic acid (C16:0). Similarly, in soybean (*Glycine max*), biochar treatment resulted in a higher proportion of UFAs, contributing to improved membrane fluidity and stress tolerance. However, the specific changes in fatty acid composition may vary depending on factors such as biochar type, application rate, soil conditions, and plant species.

The effect of biochar, alone or combined with other substances, has also been evaluated on the content of important phytochemicals such as flavonoids and phenolic compounds [4,6,14,15]. These molecules are crucial for human health due to their antioxidant properties. However, the mechanism by which biochar affects the amount of these compounds depends on plant species and environmental factors [16]. For example, in *Viola cornuta* cultivars, the content of phytochemicals was greatly influenced by both biochar and the applied fertilizer. It seems that biochar exerted a negative effect on plant phytochemicals, which fertilization could counteract [14]. A similar trend was observed in tobacco, where the content of total phenolic acid decreased with biochar application alongside a reduction in fertilizers compared to control conditions [4]. However, biochar derived from tobacco wastes was reported to upregulate the expression of genes involved in flavonoid synthesis in Chinese cherries [5]. Similarly, the flavonoid content in the roots and leaves of cowpeas was significantly higher in soils fertilized with biochar compared to non-fertilized soils [15].

Artichoke (*Cynara cardunculus* L. var. Scolymus) is a plant of significant agronomic interest, widely cultivated for its large edible parts, such as the tender inner leaves (bracts) and the receptacle, commonly known as "heart" or "head". It has played an important role in human nutrition since ancient times, especially in the Mediterranean area, and has been used in traditional medicine for its beneficial effects in treating diseases such as scurvy, anemia, and digestive pathologies [17]. These beneficial effects are attributed to the content of phenolic compounds such as cynarin, luteolin, and chlorogenic acid [18]. The levels of these compounds in artichokes are influenced by soil fertilization, with reported changes in phenol compounds and antioxidant activities depending on whether conventional or organic fertilization is applied [19]. However, there is limited information about the effect of biochar application, alone or in combination with organic fertilization, on the nutritional properties of the edible parts of artichoke cultivars.

Thus, the aim of this study was to compare the effects of biochar application as a soil amendment in combination with organic fertilization, compared to conventional inorganic fertilization, on crop performance and the nutritional composition of artichoke plants. To achieve this, plant yield and the contents of minerals, fatty acids, and phenolic compounds were analyzed in the artichoke heads to understand the effects of biochar soil amendment on the plant and soil characteristics of this crop.

2. Materials and Methods

2.1. Field Experimental Design and Treatments

Artichoke plants of the cultivar GREEN QUEEN F1 were planted in a field trial (CDTA-El Mirador station, Murcia, Spain) (37°50'51.9"N 0°53'00.1"W). The climate is semi-arid Mediterranean with a total annual precipitation of 275 mm and a mean annual temperature of 18 °C. Annual potential evapotranspiration exceeds 900 mm. Transplanting was carried out on July 15, 2022, with a planting frame of 0.8 m between plants and 2m between rows, resulting in a density of 0.62 plants/m^2.

The test was carried out in a clayey soil; (sand 26.6%, silt 27.5% and clay 45.8%), with an apparent density of 1.35g cc^{-1}, a pH of 7.8, a CEC in the aqueous extract of 0.515 and a percentage of organic matter of 1.9%

The experimental field was divided into randomized blocks of four rows with two repetitions per treatment (Figure Supp.1), totaling eight sub-parcels for each treatment. Artichokes treated with conventional inorganic fertilization (Suppl. Table 1; Suppl. Table 2) were used as the control (Control) and were compared with Biochar which consisted of the biochar addition as amendment (1300 kg ha^{-1}) and organic fertilization (Suppl. Table 4; Suppl. Table 5). Biochar longevity of in the soil is approximately 8-10 years. Over the first two years, we did not observe any effects on crop productivity or significant alterations in soil properties (data not shown).

Biochar was composed by a mix of untreated log charcoal from sustainable forestry (FSC certified), vinasse and sugar cane molasses (Suppl. Table 6), obtained by pyrolysis at low temperature (around 500ºC), rending a size of particle of 2-5mm.

Prior to our experimental setup, the field had different crop rotations. Both treatments (Control and Biochar) were normalized for N fertilization during crop phenology, considering the N soil composition. A total of 211.1, 150.49 and 49.0 fertilizer units (FU) of N, P and K, was applied respectively in the inorganic form, while 149.5, 0.20 and 38.7 FU of N, P and K was applied respectively in the organic form.

Planting took place on July 15, 2022, with the last artichoke cut on 8 May 2023, resulting in a total crop cycle of 297 days. The first harvest of artichokes occurred from November 19th to January 20th and the second harvest took place from April 14 to May 8. Climatic parameters, such as temperatures, were recorded by an experimental meteorological station located in the field (SiAR Network, Murcia, Spain).

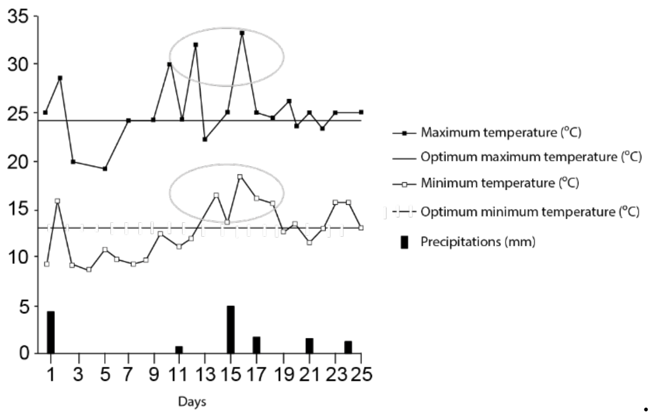

Figure 1. Climatological conditions registered at the moment of head artichokes harvest. Grey opal represents anomaly high temperatures at harvest.

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2.2. Crop Yield and Production

The heads were harvested when they reached commercial size, with firm and tight buds, a consistent head length (average 200 mm), head diameter (average 150-180 mm), and stem diameter (30-50 mm). The total fresh weight (FW) was directly measured using portable scales (PCE-EP 1500, PCE-Iberica, Spain) at the Farms Association to determine the fresh weight of the heads. The number of fruits per plant and production (Kg ha^{-1}) were also calculated. A total of 200 plants from the eight sub-parcels were selected for production measurements, with two artichoke head harvests per plant. For nutritional analysis, the two harvests were combined per treatment, as both are commercially viable.

2.3. Cations Determination in the Plant and Soils

Macronutrients (sodium (Na), K, Ca, Mg and sulfur (S)) and micronutrients (Mn, iron (Fe), copper (Cu), zinc (Zn) and Mo) were determined from 16 sample of artichoke heads mixed for each treatment and harvest date, with five technical replicates of each mixture. Additionally, K, Ca, Mg and S were also determined in the roots, leaves and soil to detect differences in cation translocation along the continuous soil-plant system.

For this analysis, 0.5g of leaf and root tissue were digested after the addition ofHNO_3/HCl (3:1 v/v) in a microwave oven (Milestone UltraWAVE SRC Microwave Digestion System (Milestone Inc., Shelton, Connecticut, USA).

For soils, homogenized aliquots of soil were taken from each replicate sub-parcel (yielding four repetitions per treatment). A 1:5 soil:water ratio (g:ml) using deionised water as a reagent was applied for the extraction procedure [20]. The tubes were then shaken in an orbital shaker (Stuart SSL1, Stone, UK) for 2h at 110 rpm at 50 ºC and filtered through 0.45 µm nylon membrane filters (CHM).

The contents of cations in plant and soils were then analysed by inductively coupled plasma (ICP) spectrometry (PerkinElmer® Optima™ 8300). Cation-exchange capacity (CEC) was determined according to FAO, 2022.

2.4. Organic Matter (OM) and Total Organic Carbon (TOC) in the Soils

Organic matter (OM) and Total Organic Carbon (TOC) were determined in the soil solid samples (1gr) using an elemental analyzer CNHS-O (EA-1108, Carlo Erba).

2.5. Extraction and Determination of Fatty Acids

Fatty acids were obtained following the instructions described in O'Fallon et al [21] with some modifications. Briefly, 50-100 mg of ground material was placed in 10 mL glass tubes, and 5.3 mL of tridecanoic acid (0.5 mg mL^{-1} in methanol) was added as an internal standard. Then, 0.7 mL of 10N KOH was added to each sample, and the samples were incubated at 55°C for 90 minutes, with manual shaking every 20 minutes. The samples were cooled and then 0.58 mL of 24N H_2SO_4 was added. The samples were incubated again at 55°C for 90 minutes, with shaking every 20 minutes. After that, 1.5 mL of hexane was added to each tube and vortexed for 5 seconds. The samples were then centrifuged at 3000 rpm for 10 minutes. One milliliter of the organic phase was collected and used for fatty acid analyses.

A gas chromatography system (Agilent 6890N) coupled to an autosampler system (Gerstel MPS2) and a mass spectrometry detector (Agilent 5975) was used to analyze the fatty acid composition of the FAMEs (Fatty Acid Methyl Esters). A Supelco SP-2560 (100 m x 0.25 mm x 0.2 µm) capillary column with a Flame Ionization Detector (FID) was used. Helium was used as the carrier gas (20 cm s^{-1}), and the temperature program was as follows: 140°C for 5 minutes, 140–240°C at 4°C min^{-1}, 240°C for 30 minutes, and finally, 140–240°C at 4°C/min. Both the injector and detector temperatures were set to 260°C. Methyl ester standards (Sigma-Aldrich 47885-U, Merck KGaA, Darmstadt, Germany) were used for identification. The polyunsaturated (PUFA) to saturated (SFA) fatty acid ratio was determined based on the types and concentrations of fatty acids.

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2.6. Extraction and Determination of Phenolic Compounds and Flavonoids.

Phenolic compounds were analyzed by HPLC-DAD-ESI/MSn following the methodology previously described [22,23] with slight modifications. The hydromethanolic extracts were analyzed by HPLC-DAD (Agilent, Santa Clara, California, USA). The identification and quantification of phenolic compounds were based on their absorbance recorded at 320 nm for hydroxycinnamic acids and 350 nm for flavonols, using chlorogenic acid and rutin hydrate as external standards, respectively (Sigma, St Louis, MO, USA).

2.7. Statistics

Parameters that conformed to a normal distribution were subjected to one-way ANOVA. A multiple range Tukey test was used to separate means, and statistical significance was assessed at the level of $P \leq 0.05$, using the SPSS 20.0 software package (IBM, Armonk, New York, USA).

3. Results

3.1. Crop Yield and Production

Total crop yield and plant production were determined by analyzing the total number of heads per m² and the number of heads per plant (Figure 1). The data indicated that there were no significant differences in the mean weight of heads between the control and biochar treatments. However, the Biochar treatment resulted in higher total numbers of heads and heads per plant compared to the Control, with increases of 5.56% and 5.30%, respectively, for both parameters.

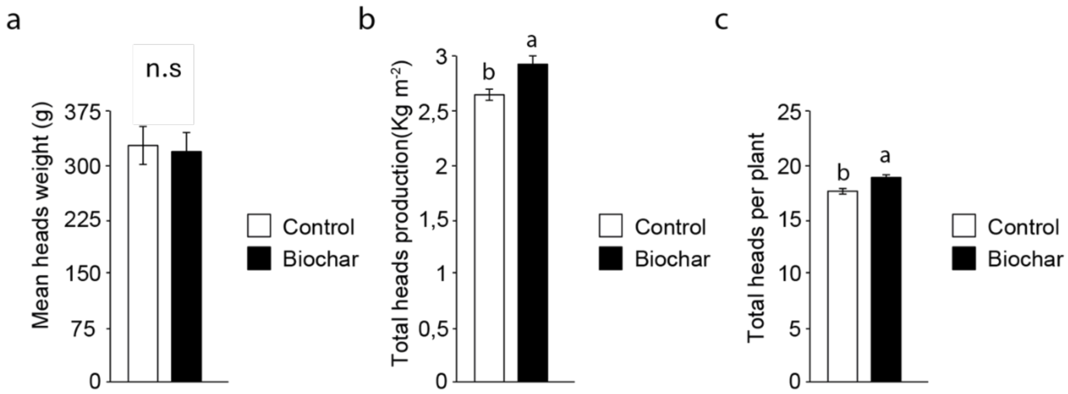

Figure 2. Effect of fertilization on the weight (a), total number of heads per m² (b) and number of heads per plant (c) of plants grown with conventional fertilization (Control) or biochar treatment (Biochar). Data represented in a are the mean value (n=8) +/- the standard deviation.

3.2. Mineral Content in Plant and Soils. Organic Matter and Total Organic Carbon in the Soil

Levels of macronutrients (K, Ca, Mg, and S) (Figure 1) and micronutrients (Mn, Fe, Cu, Zn, and Mo) (Table 2) were analyzed in the heads of Control and Biochar-treated plants. The results showed that heads from Biochar-treated plants had higher levels of Ca, Mg, and S compared to those from the Control plants. However, there were no significant differences in the K content between the Control and Biochar-treated plants.

Regarding micronutrients, Biochar treatment increased the levels of Mn but decreased the levels of Co, Zn, and Cu in the artichoke heads compared to the Control. Levels of Fe did not show significant differences between the treatments.

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Table 1. Effect of fertilization on the micronutrient concentration of heads of control and biochar treated plants. Data represented are the mean values of different measures (n=16) +/- standard deviation. Data with different letters mean that there are significantly different.

Micronutrients (mg kg^{-1} DW)	Control	Biochar
Mn	91.3±2,11a	104.8 ±4,60b
Co	1.3±0.15a	0.90±0.02b
Zn	305.0±7.30a	225.5±7.75b
Fe	314.5±17.61 n.s	310.2±6.27 n.s
Cu	42.9±1.30a	30.5±0.60b

Available cations were determined in the soil, plant roots, leaves and heads to analyze nutrient translocation along the plant. Ca increased in the heads of Biochar-treated plants compared to Control, correlating with the higher soil Ca content in the Biochar treatment. However, in Biochar-treated plants, Ca was predominantly accumulated in the roots rather than in the leaves, whereas in Control plants, Ca content was similar between roots and leaves. Mg soil content was also higher in the Biochar treatment compared to Control, but significant differences in Mg content were observed only in the heads of the plants. There were no significant differences found in the accumulated S in the soil; however, in addition to the heads, S was higher in the roots of Biochar-treated plants compared to Control. Significant differences between treatments in K concentration were observed only in the leaves, with lower levels in Biochar-treated plants compared to Control.

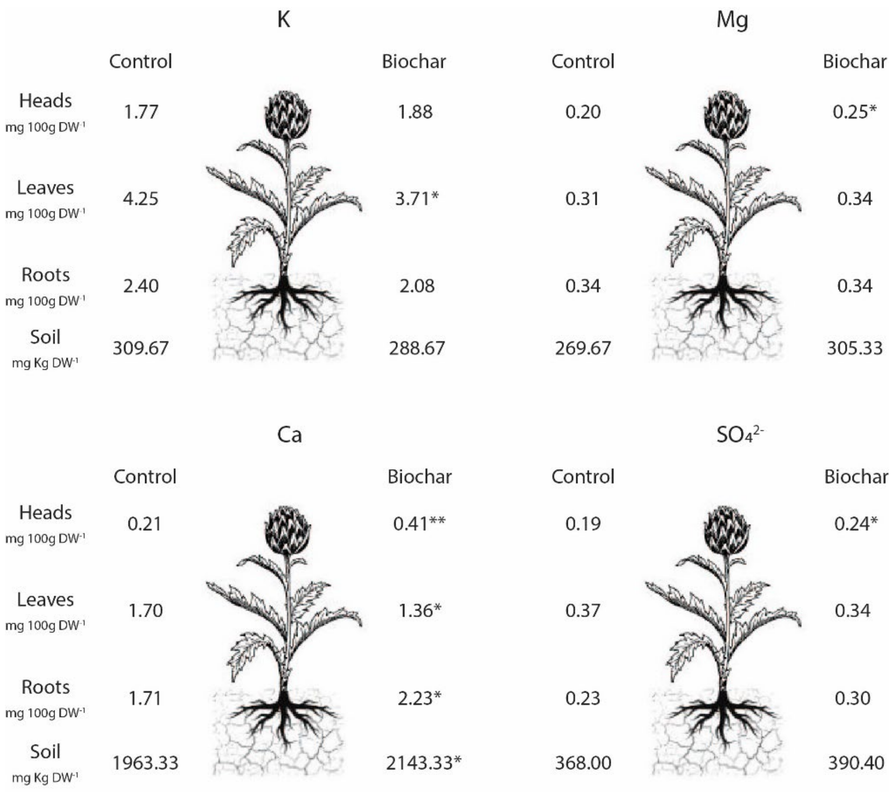

Figure 3. Effect of fertilization on the content of K, Mg, Ca and S in soil, roots, leaves and heads of control plants and plants treated with biochar. Data represented with * are statistically different between control and biochar treatments. Statistical analyses were performed for each mineral and for each part of the plant individually.

Biochar increased cation-exchange capacity according to the increment in Ca, Mg, in the soil organic matter and total organic carbon were also enhanced in the soil compared to Control (Table 2)

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Table 2. Effect of fertilization on the cation-exchange capacity (CEC), Soil Organic matter (OM) and total organic carbon (TOC) fertility parameters of Control and Biochar treated plants. Data represented are the mean values of different measures (n=4) +/- standard deviation. Data with different letters mean that there are significantly different.

	Control	Biochar
CEC (meq $100g^{-1}$)	9,99 b	11,6a
OM (%)	1,88 b	1,98a
TOC (%)	1,0 b	1,2a

3.3. Relationship between Soil Fertility and Plant Parameters

A heatmap correlation analysis revealed significant positive correlations between soil parameters and the mineral content in artichoke heads, which influenced yield (Figure 3). Specifically, yield was positively correlated with organic matter and total carbon content, and there was a slight positive correlation with ion content in the soil. In contrast, a negative correlation was observed between yield and ion levels in the plant leaves.

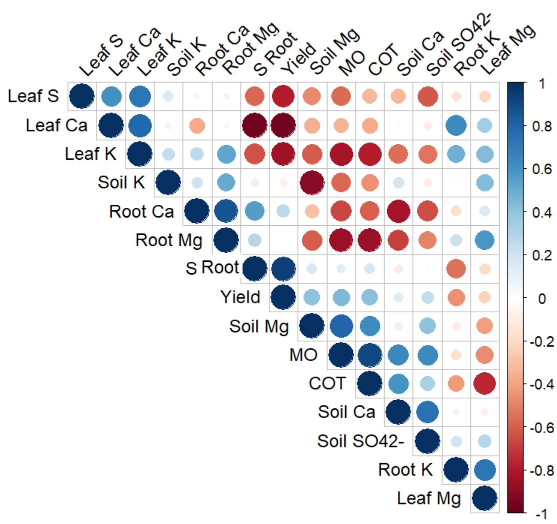

Figure 4. Relationship between soil and plant parameters under both fertilizations. The blue nodes indicate positive correlations, and the pink nodes indicate negative correlations. The size of nodes is proportional to the degree of connectivity. The larger the circle, the stronger the relationships. MO (organic matter). COT (total organic carbon).

3.4. Fatty acid Content

Levels of total fatty acids, palmitic acid, and linoleic acid in the heads were analyzed (Figure 4). The data showed that heads from plants grown with biochar application had higher levels of total fatty acids primarily due to increased palmitic acid content, while there were no significant differences in the linoleic acid content between treatments. Additionally, the PUFA/SFA (Polyunsaturated Fatty Acids/Saturated Fatty Acids) ratio was calculated for the heads of Biochar-treated and Control plants, and it was higher in Biochar-treated plants (0.95) compared to Control plants (0.83).

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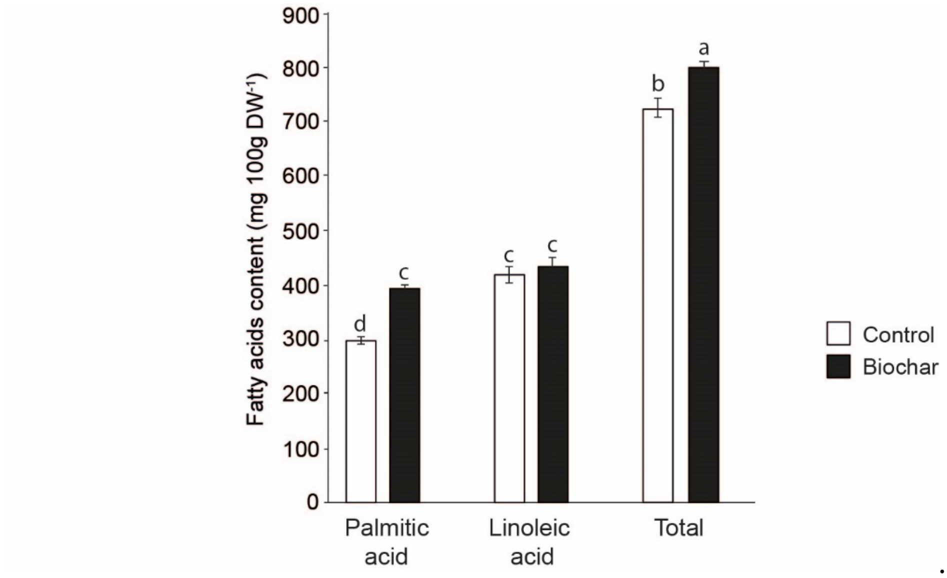

Figure 5. Effect of fertilization on the fatty acid content in heads grown with conventional fertilization (Control) or biochar treament (Biochar). Data represented are the mean values (n=16) +/- the standard deviation. Data with different letters mean that there are significantly different.

3.5. Phenolic Compounds Content.

Individual phenolic compounds in artichoke heads were determined using hydromethanolic extracts, revealing a rich composition that included chlorogenic acid (5-caffeoylquinic acid, 5-CQA), cryptochlorogenic acid (4-caffeoylquinic acid, 4-CQA), 5-p-coumaroylquinic acid (5-p-coumaroylQA), and 3,5-dicaffeoylquinic acid (3,5-di-CQA) as presented in Figure 5. The data showed that Biochar-treated heads had lower contents of most phenolic compounds, except for 5-CQA and 5-p-coumaroylQA, which remained unaffected.

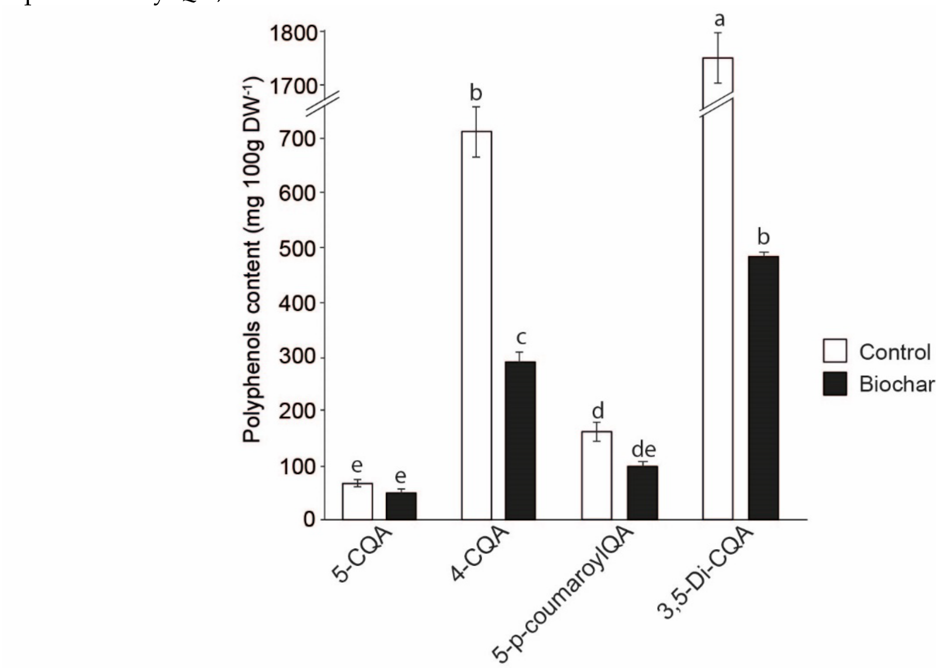

Figure 6. Effect of fertilization on the polyphenols content in heads grown with conventional fertilization (Control) or biochar treament (Biochar). Data represented are the mean values (n=16) +/- the standard deviation. Data with different letters mean that there are significantly different.

Table 7. O-rutinoside, luteolin-7-O-glucoside, luteolin-7-O-glucuronide, apigenin-7-O-rutinoside, and apigenin-7-O-glucuronide under both treatments (Figure 6). The major compound identified in

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both treatments was apigenin-7-O-glucuronide. However, Biochar-treated heads had significantly lower levels of both total flavonoids and individual flavonoids compared to the Control treatment.

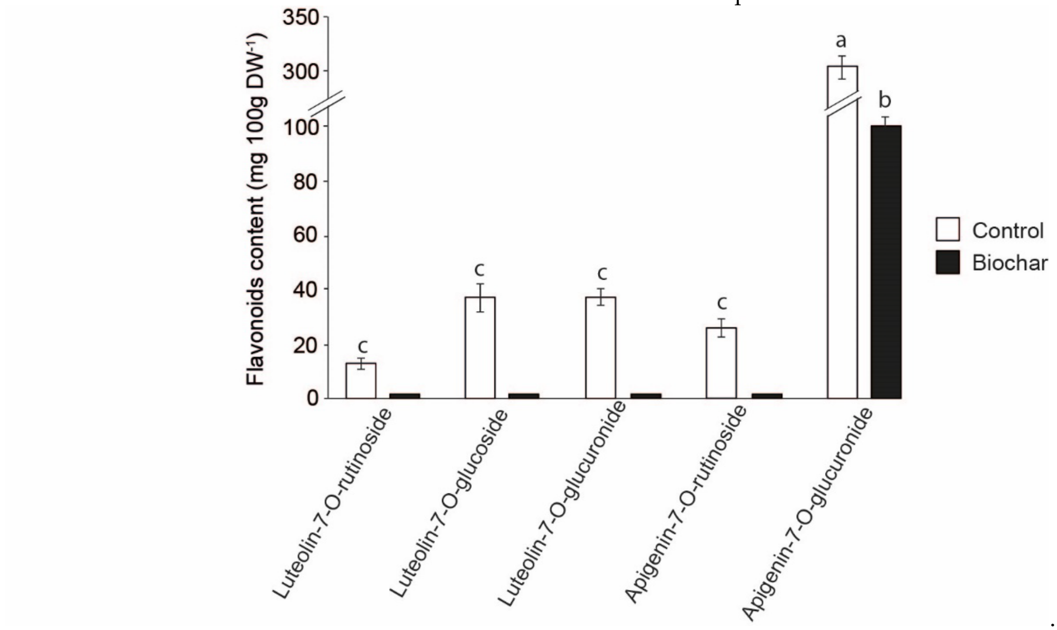

Figure 7. Effect of fertilization on the flavonoids content in heads grown with conventional fertilization (Control) or biochar treament (Biochar). Data represented are the mean values (n=16) +/- the standard deviation. Data with different letters mean that there are significantly different.

4. Discussion

In this study, biochar application promoted higher yields in artichoke production. Although different studies have reported similar findings for biochar treatment, Rondon et al., found that, in beans (*Phaseolus vulgaris* L.), biochar application rates up to 30 g soil Kg^{-1} ha^{-1} soil had a positive effect on yield, while higher concentrations (60 gr soil Kg^{-1} and 90 g soil Kg^{-1}) had no effect or even a negative effect, respectively [24]. Thus, the impact of biochar on yield is likely influenced by both the crop type and the application concentration. Another factor influencing crop yield is the properties of biochar and its decomposition over time. A study using ^{14}C has shown that biochar decomposition in soil under optimal conditions occurs at very slow rates, about 0.26% per year [25]. Studies by Zimmerman (2010) [26], indicated that biochar can initially increase soil organic carbon (SOC) by up to 20% depending on the application rate and the type of feedstock used to produce the biochar. However, the immediate effects on soil properties and crop productivity are often minimal [26]. This finding aligns with our observations over the first two years, where no significant changes in crop productivity or soil properties were detected. In this study, while the primary contribution of biochar is to organic carbon (C) inputs, its impact on N, P, and K levels within the organic fertilization regimen, albeit modest, necessitates continuous evaluation over the duration of its application through soil analysis. For example, the potassium input was 21.6 kg ha^{-1}, based on the application of 1300 kg biochar ha^{-1} with a K_2O content of 2%, applied 2 years prior to the crop yield analysis. In comparison, the potassium input from inorganic fertilizer KNO_3 was 218 kg K ha^{-1}. For plots treated with biochar, the remaining potassium required by the crop was provided via organic fertigation. Nevertheless, it would be advisable to monitor and evaluate the evolution of biochar contributions over time. This observation does not extend to phosphorus, whose soil concentrations were significantly elevated.

It has been demonstrated that biochar applications combined with organic fertilizer can induce high crop yields by regulating the balance of soil available carbon and nutrients [27]. The results of our study further support the idea that biochar treatment positively influenced artichoke yields, particularly at the specified application rate, within the climatic and soil conditions of our Mediterranean region. However, further research is needed to elucidate the mechanisms underlying

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the relationship between combined biochar and organic fertilization and crop productivity. Increased organic matter in the soil may provide an optimal environment for enhanced ion uptake by plants [28]. In fact, our study found a positive correlation between yield, organic matter content, and ion levels in the artichoke heads, suggesting that biochar treatment may facilitate ion translocation from leaves to heads.

The application of biochar induced changes in mineral content in soil and plant organs. Gavili et al. observed that the large surface area, porous structure, and high CEC of biochar improve the physicochemical properties of soils [29]. In our study, biochar treatment enhanced CEC and facilitated the transport and accumulation of Ca, Mg and S from the soil to the artichoke heads, which has implications for both plant nutrition and human dietary intake.

Similar changes in mineral content have been reported in other crops such as broccoli [6] and tomato [1], where biochar promoted the accumulation of phosphorus (P), potassium (K), iron (Fe), and zinc (Zn) in the edible parts of the plants.

Other mechanisms as stimulated microbial activity, reduced nutrient leaching, and enhanced root growth have been described to explain ion translocations along the plant [30].While experimental evidence supports these benefits, the variability in biochar properties and environmental conditions necessitates careful consideration and further research to optimize its use for sustainable agriculture.

In our study, the macronutrient content in artichoke heads under both conditions fell within the range of concentrations reported by other authors [31,32]. Artichoke heads from plants grown with enriched biochar application had significantly higher levels of Ca and Mg compared to those from control plants. Both Ca and Mg are essential minerals for human health, with daily recommended intakes of around 700-1300 mg for Ca [33] and 300-400 mg for Mg [34] in adult humans.

Calcium plays a crucial role in bone formation, where 99% of the body's Ca is found, while Mg acts as a cellular modulator and is essential for many physiological processes. Imbalances in magnesium are common and are associated with various pathological conditions that contribute to human morbidity and mortality [35]. Therefore, enriching vegetables with Ca and Mg should be considered as a strategy to enhance intake, especially among groups such as postmenopausal women (at risk for osteoporosis) as well as individuals like alcoholics and diabetic patients who are prone to Mg deficiencies [35].

Regarding potassium (K), biochar did not lead to an increase in K content in the artichoke heads, but a lower leaf K content compared to the Control was observed. This observation could be attributed to the plants' adaptation to environmental conditions, particularly the unusually high temperatures during the crop harvest period (Figure 6). It is known that K functions as an osmolyte, aiding in the maintenance of physiological and metabolic processes such as photosynthesis, respiration, and nutrient homeostasis, which helps mitigate damage from high temperatures when K accumulates primarily in the leaves of the plant [36,37]. Therefore, the lower stress effect of elevated temperatures on biochar-treated plants compared to the Control could be inferred, which might also correlate with the lower concentrations of antioxidants (phenolic compounds and flavonoids) observed in these plants.

In terms of micronutrients, biochar induced higher levels of manganese (Mn) but lower levels of cobalt (Co), zinc (Zn), and copper (Cu) in the artichoke heads. Nonetheless, the levels of these micronutrients in both conditions exceeded those reported by other authors [31,32]. Human requirements for Mn vary among countries and are typically set based on average daily intake, ranging from 1.4 mg/day to 5.5 mg/day [38]. Similarly, recommended daily intakes for zinc (Zn) and copper (Cu) vary, typically ranging from 8 mg/day to 14 mg/day and 1 mg/day to 1.25 mg/day, respectively [39]. Cobalt (Co) is considered unlikely to cause adverse effects on human health when blood concentrations remain below 300 µg/L [40]. Considering these established limits and the microelement content found in this study, it can be concluded that our artichokes serve as a valuable source of essential minerals for human health under both treatments.

The predominant fatty acids found in our artichoke heads were linoleic acid (C18:2n-6) and palmitic acid (C16:0), which aligns with findings by Dabbou et al. [41]. Similarly, a variety of

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artichokes grown in Iran showed seed oil containing oleic, linoleic, and palmitic acids [42]. Linoleic acid is particularly important due to its beneficial effects in protecting the cardiovascular system, stimulating immune defenses, and supporting nervous system function in humans [43].

Biochar application has been demonstrated to alter the lipid profile of plants. Organic fertilization also influences soil biological and chemical properties, including macro- and micro-nutrient content, which in turn affects mineral uptake by plants and the genes and enzymes involved in fatty acid biosynthesis. It has been observed changes in the levels of unsaturated fatty acids in wheat plants exposed to biochar, but similar changes were not observed in maize plants [44]. In nigella seeds, combined biochar application and minimum tillage increased palmitoleic, oleic, and linoleic acids while decreasing myristic, palmitic, and stearic acids compared to those without biochar treatment [12]. However, in our study, biochar increased the saturated fatty acid palmitic acid and had no effect on the unsaturated linoleic acid content. The ratio of polyunsaturated fatty acids to saturated fatty acids (PUFA/SFA) is associated with impacts on coronary heart disease, with a higher PUFA/SFA ratio indicating more positive health effects [45]. In our research, we determined the PUFA/SFA ratio to evaluate the nutritional value of artichokes and the influence of both inorganic and biochar-amended organic fertilization on this ratio. Despite biochar application resulting in higher levels of palmitic acid in artichoke heads, which has been associated with cardiovascular and obesity-related diseases [46], the PUFA/SFA ratio in the heads of biochar-treated plants was higher (0.95) than those from the Control (0.83). Additionally, palmitic acid is essential during fetal development and for brain development, with recommended average intake set around 20-30g/day [47]. Considering this, the content of palmitic acid in biochar-treated artichoke heads is considered suitable for human health.

The impact of biochar application on phytochemicals such as flavonoids and phenolic acids varies across different plant species and environmental conditions. For instance, in Viola cornuta cultivars, the content of phytochemicals is influenced by both biochar and applied fertilizers, with biochar sometimes exerting a negative effect that fertilization can mitigate [14]. Conversely, biochar derived from tobacco waste has been shown to upregulate genes involved in flavonoid synthesis in Chinese cherries, and similar positive effects on flavonoid levels have been observed in cowpeas when biochar was used as a soil amendment [5,15]. In our study with artichokes, biochar fertilization did not enhance the accumulation of flavonoids and even led to reduced contents of these compounds, similar to the findings for hydroxycinnamic acids [48,49]. Discrepancies across studies may arise from differences in plant species, soil characteristics, biochar properties, and conventional fertilization methods. Additionally, the timing of harvest significantly affects polyphenol content in artichokes, with higher accumulation observed under conditions of increased solar radiation and lower air temperature [50]. This partly explains the decrease in phytochemical content observed in our study, as high temperatures were recorded during the head harvest period (see Figure 7). Despite the observed decrease in phytochemical content associated with biochar fertilization in our experiments, the levels of these compounds in artichokes grown with biochar remained sufficient to qualify as a good source of phenolic compounds (including glycosylated flavonoids and hydroxycinnamic acids). Indeed, fruits and beverages are recognized as primary sources of dietary polyphenols, with recommended daily intakes averaging around 1g per day [51]. Thus, despite the challenges posed by our harsh climatic conditions, the support provided by biochar treatment ensured that artichokes still contributed significantly to the intake of these natural antioxidants in the diet.

5. Conclusions

In conclusion, the application of biochar combined with organic fertilization emerges as a promising strategy to enhance artichoke yields in semiarid Mediterranean regions while preserving nutritional quality. This study demonstrated that biochar application positively influenced soil properties such as cation exchange capacity, organic matter, total organic carbon, and the availability of essential cations like Ca, Mg, and S. These improvements in soil conditions fostered enhanced ion uptake and accumulation in artichoke heads, contributing to increased productivity. Notably, anomalies in temperature during harvest appeared to influence potassium accumulation in leaves and the levels of phenolic compounds and flavonoids in artichoke heads of control plants, possibly as a defense mechanism. The potential protective role of biochar under adverse environmental conditions, such as anomalous temperatures, warrants further investigation. Moving forward, future research should delve into the long-term effects of biochar application on mineral dynamics in agricultural soils. It is crucial to unravel the molecular mechanisms that underlie biochar-induced changes in mineral uptake and transport in plants. Furthermore, optimizing biochar application strategies by considering its nature and the specific types of crops and agroecosystems will be essential to maximize its beneficial impacts.

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