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Soil Biochar Application: Assessment of the Effects on Soil Water Properties, Plant Physiological Status, and Yield of Super-Intensive Olive Groves under Controlled Irrigation Conditions

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Abstract: The effects of olive waste biochar and green compost as soil amendments on soil physical properties, as well as on physiological parameters and yield of a super-intensive olive crop cultivated under deficit irrigation conditions, were investigated in south-west Spain during the 2021 growing season. Thus, soils were amended with 40 t ha^{-1} of olive pomace waste biochar, green-compost, or a biochar-compost mixture (50% *w*/*w*), and no amended plots were used as control. On a bi-monthly basis, soil pH, water holding capacity, humidity, and resistance to penetrability were determined. In addition, various indicators of the physiological status and water stress of the plant were also monitored. Finally, the olive yield per tree was measured. Results showed that biochar application was the most effective amendment for increasing soil moisture and reducing soil compaction. The latter was evidenced by the significant reduction of the resistance to the penetrability of the amended soils. Plants of the amended plots showed better leaf water potential. In addition, values of the net photosynthesis rate, the average intrinsic water-use efficiency, and the maximum rate of electron transport in the time before the harvest improved significantly in the trees from the biochar-amended plots, for which olive fruit yields increased by about 15% in comparison with the other treatments. Nevertheless, the estimated net oil yield per tree was similar because the olives from the biochar-amended trees contained more moisture. This field trial shows for the first time that by providing the soil with biochar from olive crop waste as an organic amendment, having high water retention capacity, porosity, and stability, it would be possible to reduce the irrigation water needed and maintain plant yields.

Keywords: soil physical properties; plant physiology; irrigation; water relations; organic amendment; circular economy

1. Introduction

Agriculture is currently facing a complex challenge in the context of climate change: the need to provide quality food to a growing world population. It is expected to reach 9.7 billion inhabitants by 2064 and, as a consequence, more people will suffer from hunger [1]. While greenhouse gas emissions have intensified, the fertility of agricultural soils has declined globally due to the exposure of soils to frequent flooding, desertification, and salinization [2]. Agriculture is one of the economic pillars of the Mediterranean Basin, and of particular interest is the cultivation of olives, of which there are an estimated 7.7 million hectares. This area constitutes about 70% of the existing 1.5 billion olive trees in 56 countries around the world. Spain has more than 2.5 million hectares of olive trees (28% of which are irrigated) [3], and is currently the world's leading producer and exporter [4]. In recent years, in a clear bid to avoid year-to-year fluctuations in productivity and to guarantee profitability, intensive and super-intensive olive growing has been expanding, especially on new farms [5]. In contrast to traditional cultivation, in super-intensive cultivation, the tree is smaller in size (hedgerow shape), up to 2000 olive trees are planted per hectare, compared to 600 trees per hectare for intensive cultivation. In both cases, automatic harvesting is possible, but controlled irrigation (usually drip irrigation) is essential. Nevertheless, the Mediterranean region is characterized by a semi-arid climate. This recent development of intensive olive cultivation that requires irrigation, together with the scarcity of rainfall, means that tensions and conflicts between water users and pressures on the environment are expected to increase.

Another problem affecting agriculture in the Mediterranean region in general, and the olive sector in particular, is the large generation of residual biomass with little commercial value. According to EUROSTAT [6], Spain produced around 5.5 million tons of agricultural and forestry waste in 2012. The olive oil agro-industry also produces abundant residual biomass, such as olive pits, estimated at over 420,000 tons per year (14% fruit weight [7]), pruning residues, dried pomace, etc. The olive pomace, the residual mixture of seeds, olive skins, and pulp [8], has a high content of polyphenols, which makes it a waste of high phytotoxicity and recalcitrance to degradation [8], and, consequently, these biomass residues are normally burned. One of the most promising alternatives being considered to resolve this problem in a sustainable way, is the conversion of these biomasses into biochar [9]. It is the solid and porous by-product resulting from the thermal decomposition of biomass in the absence of oxygen, in a process known as pyrolysis. This process generates a mixture of combustible gases and heavy oils that can be used as an energy source [10]. The agronomic benefits of the application of partially carbonized organic matter as an amendment in agricultural soils were already known by the indigenous people of the Amazon basin in Latin America thousands of years ago, in what has been termed as *terra preta de Indio* (i.e., Portuguese for Indian Black Earth) [11]. Over the last decade, scientific interest in biochar has increased exponentially, with more than 4000 scientific papers published on the subject of biochar and agriculture in 2021 alone. Due to its properties, such as high carbon content, stability, cation exchange capacity, porosity, and moisture retention [12,13], the application of biochar, solely or together with other amendments or fertilizers, in crop soils has been widely experimented, resulting in beneficial effects, especially in coarse-textured soils in arid and semi-arid areas [14]. Biochar can improve the soil physiochemical properties, such as water infiltration, water holding capacity, and saturated hydraulic conductivity to accommodate cultivation [15–20], and increase agricultural productivity [21,22]. Some recent reviews discussed the effects of biochar on soil physical and hydrological properties and indicated that biochar application improved soil compaction [23,24]. Additionally, a meta-analysis [25] has described that biochar application into agrosystems may have proportionate positive effects on crop productivity or soil properties, although they are neither universal nor uniform. For instance, Sorrenty et al. [26] reported that biochar application to sandy-loam soil was effective in reducing the leached amount of NH_4-N in the top-soil layer but in non-limiting conditions (e.g., water availability and soil fertility), the benefits from biochar application in commercial nectarine trees performance were negligible. Recent studies strongly suggest that large rates of biochar application may be necessary to significantly change the penetration resistance of the soil and reduce the risks of soil compaction (e.g., [27]), whereas cost-benefit analyses suggest the application of lower biochar doses [28,29]. However, the economic implications of the implementation of novel organic amendments are not within the scope of this paper, which aims, for the first time, for a holistic study that analyzes the impact of biochar on soil properties (including water content, moisture, penetration resistance, pH, and electrical conductivity), water, physiological plant status, and crop productivity in the field under Mediterranean climate conditions. Furthermore, the recovery of agro-residues for subsequent use in agriculture is important as a circular economy strategy for materials and nutrient reuse [30,31]. In relation to olive agro-industry residues, Lv et al. [32] already reported high yields and good performance in obtaining biochar from olive residues due to their relatively high lignin content compared to other biomasses. Campos & De la Rosa [33] found that biochar produced from olive pits has physical, chemical, and surface properties (water retention capacity greater than 100%, very high porosity (>300 m^2 g^{-1}), low conductivity, and density, among others) that make it very suitable for improving the water properties of soil. The recovery of biochar from olive mill wastes has recently been applied as an eco-friendly and effective method for their sustainable management [30]. Nevertheless, the lack of appropriate pyrolysis reactors means that biochar is scarce, expensive, or not produced under appropriate conditions; therefore, most trials are conducted at the pot scale. At present, the application of high doses of biochar as an arable soil amendment is not economically viable. Nevertheless, if sustainable and cost-effective applications can be found, the development and installation of high-efficiency pyrolytic reactors in which biochar is the solid by-product of the transformation of biomass into syngas would be facilitated by making quality biochar locally available at a highly competitive price.

The improvement of orchard management practices is compulsory in modern olive-growing intensive systems, planted at high density, with irrigation, and with trees formed to suit mechanical pruning and harvesting. In recent years, increasing attention has been paid to the physiological basis of crop response to cultural practices, with the aim of achieving more efficient use of water [34]. In addition, the positive effects of applying biochar in soil structural properties should be linked to its improvement in above- and belowground growth of plants, and some other phenomena that would directly enhance the process of root area and activity of microorganisms. However, taking into account that the improvements in soil physiochemical properties differ with feedstock type, soil, ageing of biochar with soil, and rate of application [35], its indirect impact on plant physiology is even more uncertain. Recent studies, (e.g., [28,29]) focused on dosage optimization suggested biochar doses $\leq$30 t ha^{-1} due to the declines in growth and physiological performance as a consequence of the induced N limitation at high biochar dosages. On the contrary, Busscher et al. [27] reported that an application of at least 44 t ha^{-1} is needed to significantly reduce the soil resistance to penetrability and improve soil physical properties in a sandy loam soil. It is clear that results differ according to soil type, climate, and crop. Physiological characteristics, including photosynthesis rate (A_N), stomatal conductance (g_s), maximum electron transport rate (ETR_{max}), and leaf water potential (ψ_{leaf}), are key features to determine the general performance and health of plants [36]. Effects of soil amendment by biochar and other organic ameliorants on the physiological properties of olive trees could provide valuable information to benefit farmers. Another question still to be confirmed is whether the combined application of biochar and compost has synergistic effects, as contrasting results have been reported [37,38].

Thus, considering the lack of field trials with biochar in super-intensive olive orchards, the main objectives of this study are:

(i) To verify in the field the effects of the application of biochar produced from olive pomace residues on soil properties, especially water properties, plant physiology, and crop productivity;

(ii) To compare the effects of the application of biochar with the performance of the traditionally-used green-compost.

To our knowledge, this is the first experiment which studies, under real field conditions, how the application of biochar and compost as soil amendments affects soil physical properties, soil composition, and olive tree physiology and productivity. This information is essential to understand how the crop uses water and how biochar and compost could affect soil properties and crop response.

2. Materials and Methods

2.1. Description of Organic Amendments

2.1.1. Olive Pomace Biochar (OB)

Biochar was produced from dry olive pomace (hereinafter referred to as olive pomace biochar-OB) in the pyrolysis reactor of Carboliva S.L., a company in the hamlet of Puente del Obispo (Jaén, Andalusia, southern Spain). The slow pyrolysis was performed at that rotatory cylindrical reactor (8 m long × 2 m diameter) fed by a screw in a continuous process for 15 min at 500 °C. The anoxic conditions were established by a continuous flux of N_2 and CO_2. This reactor converts the olive mill pomace with a capacity of 9 t per hour into a syngas fraction, rich in methane, that is valorized for energy production and used as a green biofuel, and a solid fraction called biochar. The OB sample has a pH of 9.90 ± 0.05, a water holding capacity of 78 ± 15%, ash content of 38.4 ± 0.3%, and total organic C and N contents of 56.3 ± 1.7% and 1.13 ± 0.03%, respectively, a particle size in the range of 5 to 25 mm, and an electrical conductivity of 13,700 ± 389 µS cm^{-1}. The analytical procedures used are briefly described in Section 2.3.

2.1.2. Green Compost (GC)

Green compost (hereinafter GC) was supplied by Fertilizantes Orgánicos Melguizo S.L. (Seville, Spain). This commercial compost is made from a mixture of garden pruning waste and pine wood shavings recovered from discarded pallets. The raw materials are collected in the reception area and mixed prior to transfer to the fermentation area in its facilities located in the town of Los Palacios y Villafranca (Seville, Spain). The composting plant is located in a geographical area where rainfall is infrequent. Thus, the composting was performed using a traditional process in open piles, and aeration was assured by a turning process. GC has pH of 8.3 ± 0.2, the electrical conductivity of 1184 ± 177 µS cm^{-1}, water holding capacity of 66 ± 20%, ash content of 73 ± 3%, and total organic C and N contents of 14.9 ± 0.2 and 0.68 ± 0.02%, respectively.

2.2. Experimental Setup and Orchard Characteristics

The experimental plots were located at La Hampa experimental farm, which is located in Coria del Río municipality (37°17′ N, 6°3′ W, 30 m above sea level; Figure 1). This area has a typical Mediterranean climate, with mild winters and dry summers, and rain episodes concentrated during autumn and spring. The average minimum and maximum air temperatures in the area are 12 °C and 26 °C, respectively. The cumulative mean rainfall was 496 L m^{-2} for the period 2015–2020 [39] and 443 L m^{-2} during 2021, and the yearly potential evapotranspiration is about 1500 L m^{-2} [40]. Temperature and precipitation were recorded daily during the experiment by an automated agroclimatic station (Figure 1).

The topsoil at the experimental site is sandy loam (Xerochrept) with a pH of 7.6 ± 0.3, and total C, organic C, and N contents of 2.2 ± 0.4, 1.2 ± 0.3, and 0.18 ± 0.03%, respectively. The trees are 6-year-old *Olea europapea* L. (Arbequina) planted in lines in a 4 m × 1.5 m configuration (1667 trees ha^{-1}). All of the trees were irrigated with 30% regulated deficit irrigation (RDI) [41,42]; i.e., so as to satisfy 30% of irrigation needs every time [43]. The temporal distribution of irrigation is shown in Figure 1. The highest irrigation requirements were observed throughout the summer period and amounted to 460 L m^{-2} of the 752 L m^{-2} yr^{-1} applied, which corresponds to 7520 m^3 ha^{-1} yr^{-1}.

Taking into account the layout of the olive trees in this super intensive olive grove, and the drip irrigation system, four different treatments were stablished on 15 January 2021, consisting of control plots with no amendment (C; control), olive pomace biochar (OB), green compost (GC), and a 1:1 (*w/w*) of OB: GC mixture (OB + GC). The organic amendments were not applied to the entire surface of the farm but solely to plots of 1 m × 1 m around each tree using a frame (Figure 1d). Thus, 4 kg m^{-2} (equivalent to 40 t ha^{-1}) were applied and subsequently mixed with the first 5 cm of the soil at each plot using a rake, to leave a uniform distribution. Taking into account the distribution of the trees and the distance between them, the net real amendment applied is 6.67 t ha^{-1} for

BC and GC, and 3.33 + 3.33 t ha^{-1} for the mixture BC + GC, respectively. Each treatment was applied per complete row of 24 plots and not to a random distribution of trees in the parcel to avoid cross-contamination due to erosion or wind transport, flowing in the order of: C/C/GC/OB + GC/GC/OB + GC/OB/OB/OB/C. Consequently, the number of plots was n = 72, 72, 48, and 48, for treatments C, B, GC, and OB + GC, respectively, making a total of 240 plots of 1 m^2 each.

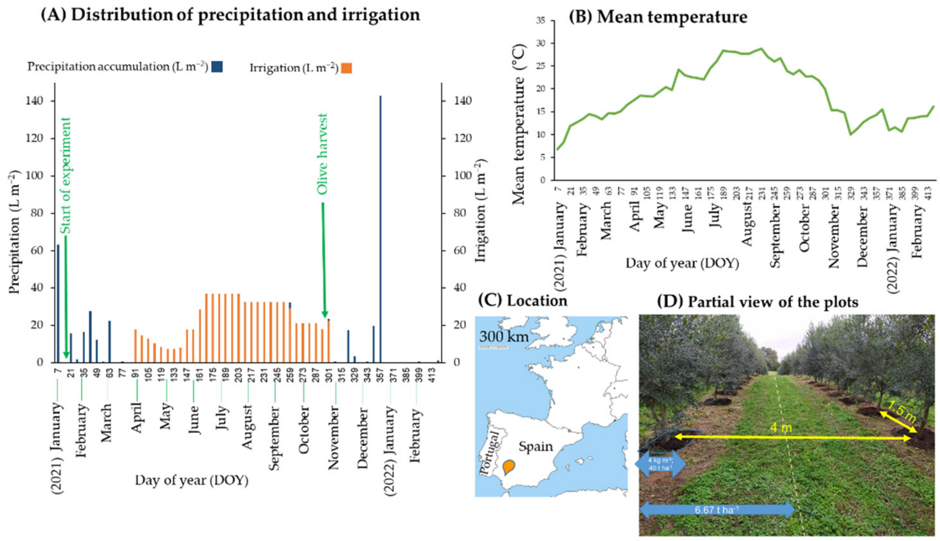

Figure 1. Distribution of precipitation and irrigation in liter per square meter (L m^{-2}) (**A**); mean temperature (°C) during the field experiment (**B**); location of the experimental station La Hampa at SW Spain (**C**); and partial view of the plots and indication of the dosage (**D**).

Soil samples were taken from 0 to 5 cm (topsoil), 5 to 10 cm, and 10 to 20 cm depths with a manual auger at three points and four different plots for each treatment on the days of the year (DOY) 104, 118, 140, 159, 173, 187, 251, 260, 287, 294, 334, and 418. To allow statistical comparison, the same number of measurements per treatment has always been considered for each of the studied parameters.

2.3. Analyses of Physical and Chemical Properties of Amendments and Soils

Total carbon (TC) and total nitrogen (TN) contents of OB, GC, and soil were determined by dry combustion, using a Flash 2000 HT elemental micro-analyzer (Thermo Instruments, Bremen, Germany) equipped with a thermal conductivity detector at 1020 °C. The pH and EC were measured by the procedure described by Campos et al. [44] in a 1:5 (w/w) soil: distilled water mixture, whereas for pure organic amendments, a 1:10 (w/w) mixture was used. Total moisture (%) was determined for amended and un-amended soils by weight difference after drying the samples at 105 °C for 24 h in order to calculate the total water content, and water holding capacity (WHC) was measured by the method of Campos et al. [45].

2.4. Field Determination of Soil Humidity and Resistance to Penetrability

Soil moisture (%) was measured in the field at 0–5 and 5–10 cm depths with a PMS710 soil moisture meter (Tsingtao Toky Instruments Co., Ltd., Qingdao, China). For each treatment and depth, soil moisture was measured at three points and four different trees on DOY 104, 118, 140, 159, 187, 251, 260, 287, 294, 334, and 418.

Soil resistance to penetrability (kg cm^{-2}) was measured at soil surface using the HM-502 hand penetrometer (Gilson Company, Inc., Lewis Center, OH, USA). Soil resistance to penetrability was determined at nine points and four different trees for each treatment on the same DOY as soil moisture.

2.5. Olive Trees Physiological Status

Plant physiology measurements were carried out on DOY 251, 287, 334, and 418 to provide data for the growth, fruit ripening, and post-harvest stages. Leaf water potential (ψ_{leaf}) was measured with a Scholander type pressure chamber (PMS Instrument Company, Albany, OR, USA). Measurements were made at noon (GMT) on sun-exposed, healthy, fully developed leaves of representative current-year branches. Eight leaves were measured in four representative trees per treatment.

Leaf gas exchange and chlorophyll fluorescence were measured simultaneously using an open gas exchange system (LI-6400, Li-Cor, Lincoln, NE, USA) equipped with the LI-6400-40 chamber. Twelve leaves per treatment were measured between 9:00 and 11:00 GMT, which are the times of the day when stomatal conductance is maximum. Measurements were performed in four representative trees, in leaves that were sun oriented and fully mature. Conditions in the chamber were set at 400 μmol air s^{-1}, ambient CO_2 = 430 μmol CO_2 mol^{-1}, and PPFD = 1600 μmol $m^{-2}s^{-1}$. The actual photochemical efficiency of photosystem II (ϕ_{PSII}) was determined by measuring steady-state fluorescence (F_s') and maximum fluorescence during a light-saturating pulse of 10,000 μmol$m^{-2}s^{-1}$ (F_m') following the procedures of Genty et al. [46]:

$$\phi_{PSII} = (F_m' - F_s')/F_m' \quad (1)$$

The maximum electron transport rate (ETR_{max}) was then calculated as:

$$ETR_{max} = \phi_{PSII}\ PPFD\ \beta \quad (2)$$

where PPFD is the photosynthetic photon flux density and β is a term which includes the product of leaf absorptance, and the partitioning of absorbed quanta between PSI and PSII, considered as 0.5. Leaf absorptance was assumed to be 0.93 as measured in Perez-Martin et al. [47]. Briefly, leaf absorptance was determined using an integrating sphere with a portable spectroradiometer (LI-1800; Li-Cor) and calculating absorptance as 1–reflectance–transmittance.

2.6. Olive and Oil Yields per Tree

On 21 October 2021 (DOY: 294), the olives were harvested from the entire plot under study. The fruit was collected by hand, by a crew of four people who first spread a net entirely covering the soil surface area of four trees, and weighed immediately. Thus, for each line of 24 trees, 6 measurements of olive weight were obtained. For each batch, 2 kg of olives was taken for the analysis of moisture and fat content.

Olive water content (%; w/w) was calculated following the normalized methodology described in ISO 662:2016 [48]. Briefly, 20 g of fresh olive as dried at 103 ± 2 °C. Fat content was subsequently determined by using the ISO procedure [41]. An amount of 10 g of dry and milled sample was placed in a cellulose thimble and then into a Soxhlet chamber (50 mL) fitted to a distillation flask containing 100 mL of *n*-hexane (HPLC grade) and boiling glass regulators. After extraction for 4 h the thimble was dried to remove the solvent and the sample was weighed again.

The percentage of oil yield for the olive was calculated using the formula:

$$\text{Olive oil yield (\%)} = \text{Mass before extraction} - \text{Mass after extraction}/\text{Mass before extraction} \times 100\ (\%) \quad (3)$$

Free acidity of olives (oil acidity) is a parameter that is typically used to check the quality of the olive oil and to differentiate between extra virgin, virgin, and ordinary virgin. Acidity of fresh olives (%) was determined by Visible/Near Infrared (VIS/NIR) spectroscopy, following the procedure described by Garrido-Varo et al. [49].

2.7. Statistical Analysis

The IBM SPSS Statistics 26.0 software (SPSS, Chicago, IL, USA) was used for performing statistical analysis. One-way ANOVA and HSD Tukey (p = 0.05) as a post hoc analysis were performed after testing the normality and homoscedasticity of the data (Shapiro–Wilk and Levene tests). For non-normal variables, Kruskal–Wallis and Mann–Whitney U tests were performed. Linear correlations were tested using the obtained results to better discern the effects of organic amendments on soil properties and their consequences on productivity.

3. Results and Discussion

3.1. Physical and Chemical Properties of Soils

3.1.1. Laboratory Analyses

Compared to the control, not amended soils, the addition of biochar (either as OB or GC + OB) significantly increased pH and EC of the topsoil, from 7.6 to ≥9.4 and from 191 to >700 µS cm^{-1}, respectively. This increase is due to the high alkalinity and EC of OB, and was maintained throughout the duration of the experiment for the topsoil (Table 1). However, the differences were practically negligible at a soil depth greater than 10 cm.

Table 1. Physical and chemical properties of soils.

	Sample Depth	Day of Year	Month	Treatment C	OB + GC	GC	OB
pH (1:5)	0–5 cm	118	April	7.6 ± 0.5 ^{c,}	9.4 ± 0.1 ^{b}	8.1 ± 0.2 ^{c}	9.7 ± 0.4 ^{a}
		159	June	7.7 ± 0.1 ^{c}	9.4 ± 0.0 ^{b}	7.4 ± 0.0 ^{d}	9.9 ± 0.0 ^{a}
		251	September	8.2 ± 0.0 ^{c}	9.5 ± 0.0 ^{a}	8.3 ± 0.0 ^{c}	9.3 ± 0.0 ^{b}
		287	October	8.9 ± 0.1 ^{b}	10.1 ± 0.1 ^{a}	8.6 ± 0.0 ^{c}	9.1 ± 0.0 ^{b}
		418	February	9.7 ± 0.7	8.5 ± 0.2	9.3 ± 0.7	9.4 ± 0.6
	5–10 cm	118	April	8.1 ± 0.2 ^{b}	9.1 ± 0.2 ^{a}	8.2 ± 0.2 ^{b}	9.3 ± 0.4 ^{a}
		159	June	7.7 ± 0.1 ^{d}	8.1 ± 0.1 ^{c}	8.3 ± 0.1 ^{b}	9.7 ± 0.0 ^{a}
		287	October	9.1 ± 0.1 ^{c}	10.1 ± 0.1 ^{a}	9.4 ± 0.0 ^{b}	9.0 ± 0.0 ^{c}
		418	February	9.4 ± 0.1 ^{a}	9.0 ± 0.4 ^{a}	9.9 ± 0.3 ^{a}	9.3 ± 0.4 ^{a}
	10–20 cm	118	April	7.9 ± 0.3 ^{b}	9.0 ± 0.1 ^{a}	8.6 ± 0.2 ^{a}	9.0 ± 0.6 ^{a}
		251	September	9.1 ± 0.1 ^{a}	9.0 ± 0.2 ^{a}	9.0 ± 0.0 ^{a}	8.8 ± 0.0 ^{a}
		287	October	9.0 ± 0.0 ^{a}	9.2 ± 0.0 ^{a}	9.1 ± 0.0 ^{a}	8.9 ± 0.0 ^{a}
		418	February	9.4 ± 0.0 ^{a}	9.2 ± 0.2 ^{a}	9.6 ± 0.3 ^{a}	9.0 ± 0.1 ^{a}
EC [µS cm^{-1}] (1:5)	0–5 cm	118	April	191 ± 56 ^{c}	743 ± 330 ^{ab}	450 ± 167 ^{b}	754 ± 211 ^{a}
		159	June	227 ± 81 ^{d}	475 ± 39 ^{c}	1242 ± 22 ^{a}	789 ± 33 ^{b}
		251	September	1343 ± 12 ^{c}	1798 ± 102 ^{b}	2330 ± 42 ^{a}	1185 ± 49 ^{c}
		287	October	904 ± 13 ^{c}	1964 ± 12 ^{a}	1193 ± 6 ^{b}	473 ± 30 ^{d}
		418	February	240 ± 82 ^{a}	130 ± 29 ^{a}	160 ± 38 ^{a}	188 ± 52 ^{a}
	5–10 cm	118	April	167 ± 32 ^{c}	307 ± 109 ^{ab}	276 ± 112 ^{b}	353 ± 110 ^{a}
		159	June	431 ± 21 ^{a}	180 ± 10 ^{b}	309 ± 85 ^{ab}	435 ± 26 ^{a}
		287	October	305 ± 25 ^{b}	478 ± 2 ^{a}	274 ± 7 ^{b}	217 ± 4 ^{c}
		418	February	119 ± 25 ^{a}	109 ± 6 ^{a}	169 ± 52 ^{a}	107 ± 8 ^{a}
	10–20 cm	118	April	211 ± 69 ^{a}	381 ± 262 ^{a}	219 ± 38 ^{a}	257 ± 76 ^{a}
		251	September	379 ± 28 ^{a}	480 ± 89 ^{a}	410 ± 32 ^{a}	506 ± 3 ^{a}
		287	October	226 ± 3 ^{b}	330 ± 13 ^{a}	243 ± 8 ^{b}	189 ± 5 ^{c}
		418	February	144 ± 11 ^{a}	137 ± 11 ^{a}	142 ± 55 ^{a}	104 ± 9 ^{a}

Table 1. *Cont.*

	Sample Depth	Day of Year	Month	Treatment C	OB + GC	GC	OB
Water content (105 °C; %)	0–5 cm	118	April	17.4 ± 1.6 ^{a}	15.9 ± 2.9 ^{a}	16.5 ± 1.6 ^{a}	19.1 ± 3.7 ^{a}
		251	September	11.7 ± 0.4 ^{b}	15.2 ± 0.3 ^{a}	12.6 ± 0.5 ^{b}	15.5 ± 0.1 ^{a}
		287	October	13.7 ± 0.3 ^{b}	15.3 ± 2.4 ^{b}	14.7 ± 0.5 ^{b}	26.8 ± 0.6 ^{a}
		418	February	9.7 ± 0.1 ^{ab}	7.4 ± 1.3 ^{b}	5.4 ± 0.2 ^{b}	18.6 ± 6.5 ^{a}
	5–10 cm	118	April	16.8 ± 1.7 ^{a}	13.5 ± 3.0 ^{a}	15.2 ± 1.9 ^{a}	17.3 ± 3.0 ^{a}
		287	October	14.4 ± 0.3 ^{b}	14.1 ± 0.7 ^{b}	13.7 ± 0.3 ^{b}	17.6 ± 0.2 ^{a}
		418	February	11.1 ± 3.2 ^{b}	7.0 ± 0.3 ^{b}	8.6 ± 0.7 ^{b}	18.6 ± 0.6 ^{a}
	10–20 cm	118	April	15.9 ± 2.2 ^{a}	15.9 ± 2.2 ^{a}	16.1 ± 0.3 ^{a}	17.1 ± 1.9 ^{a}
		251	September	15.1 ± 0.1 ^{a}	15.1 ± 0.1 ^{a}	15.8 ± 0.2 ^{a}	15.9 ± 0.1 ^{a}
		287	October	14.9 ± 0.9 ^{a}	14.9 ± 0.9 ^{a}	14.8 ± 0.4 ^{a}	16.9 ± 0.5 ^{a}
		418	February	12.6 ± 2.0 ^{b}	7.0 ± 1.7 ^{c}	12.6 ± 0.4 ^{b}	18.3 ± 0.5 ^{a}

Different letters indicate significant differences between treatments in the same sampling ($p < 0.05$). C: Control; OB + GC: Olive Biochar+ Green compost; GC: Green Compost; OB: Olive Biochar.

Soil water content did not differ between treatments in the spring season (DOY 118), even at depth. However, in samples taken from September onwards (DOYs 251, 287, and 418), it was observed that OB, and, to a lesser extent, GC amendment, increased the water content significantly, also for the soil samples taken below 5 cm depth. Previous studies have reported an increase in soil water retention, a decrease, or no change [35,45,50,51] after biochar application. Razzaghi et al. [24] concluded that the impact of biochar on soil water content might be soil type-dependent.

3.1.2. In Situ Analyses

The average values of soil moisture and penetration resistance measured in the field for each treatment are shown in Table 2. The soil moisture measured in the field ranged from 7.1% (C; spring) to 20.3% (OB + GC; summer). The application of any amendment increased soil moisture of the top-soil in the spring and summer seasons. During summer and autumn, irrigation with up to 140 L m^{-2} per month during these periods increased the average soil moisture for all treatments. The differences in soil moisture between the treatments are reduced for the soil fraction of 5 to 10 cm depth.

Control plots showed average penetration resistance values of 4 MPa, whereas 1.9, 1.9, and 2.5 MPa were measured for OB, OB + GC, and GC treated plots, respectively. Thus, the penetration resistance decreased considerably with the application of both GC and OB compared to the soils of the control plots throughout the entire experiment (Table 2). Exceeding 3 MPa for coarse-textured soils can significantly reduce root growth [52]. Similarly, Blanco-Canqui [53] reported that biochar application rates >20 Mg ha^{-1} reduced penetration resistance.

Soil moisture (0–5 cm depth) and penetration resistance measured in the field were strongly inversely correlated ($R^2 = 0.758$, $p < 0.05$; Figure S1 and Table S1). This result indicates how soil physical properties are interrelated. The addition of an organic amendment, such as biochar or compost, allowed improved penetrability, reduced compaction and allowed moisture to increase, with a similar water supply. This result may suggest that soils with biochar could be trafficked at higher water content, without causing compaction, than soils without biochar [53]. A possible advantage of biochar application over compost is the greater stability of the former. This, in turn, could extend the beneficial effects of the amendment for a longer period of time, but this has yet to be demonstrated in long-term trials.

Table 2. Average soil moisture and resistance to penetrability (in situ measurements) of not amended and amended soils.

	Sample Depth	Season	Treatment C	OB + GC	GC	OB
Soil moisture (%)	0–5 cm	Spring	7.1 ± 6.5 ^{c}	13.6 ± 6.0 ^{b}	13.4 ± 6.7 ^{b}	15.5 ± 5.4 ^{a}
		Summer	11.6 ± 7.8 ^{b}	20.3 ± 0.3 ^{a}	19.9 ± 1.8 ^{a}	20.1 ± 1.7 ^{a}
		Autumn	16.7 ± 5.5 ^{a}	19.9 ± 1.2 ^{a}	18.6 ± 3.3 ^{a}	19.0 ± 2.6 ^{a}
		Winter	11.2 ± 7.9 ^{c}	12.2 ± 6.9 ^{bc}	17.5 ± 5.3 ^{ab}	18.7 ± 2.6 ^{a}
	5–10 cm	Spring	11.5 ± 6.2 ^{b}	17.0 ± 3.3 ^{a}	17.3 ± 2.9 ^{a}	18.0 ± 2.3 ^{a}
		Summer	17.1 ± 4.8 ^{b}	20.6 ± 0.1 ^{a}	20.6 ± 0.2 ^{a}	20.2 ± 3.0 ^{a}
		Autumn	19.5 ± 2.2 ^{a}	20.5. ± 0.2 ^{a}	20.1 ± 1.3 ^{a}	20.5 ± 0.2 ^{a}
		Winter	17.3 ± 3.8 ^{a}	16.1 ± 4.0 ^{a}	20.4 ± 0.3 ^{a}	20.4 ± 0.2 ^{a}
Resistance to penetrability (kg cm^{-2})		Spring	3.8 ± 1.5 ^{a}	2.5 ± 1.5 ^{b}	2.6 ± 1.5 ^{b}	2.0 ± 1.1 ^{c}
		Summer	4.0 ± 1.9 ^{a}	1.7 ± 1.1 ^{b}	1.8 ± 1.0 ^{b}	1.3 ± 0.8 ^{b}
		Autumn	3.5 ± 1.7 ^{a}	1.2 ± 0.3 ^{c}	1.8 ± 0.9 ^{b}	1.5 ± 0.9 ^{b}
		Winter	4.7 ± 1.6 ^{a}	2.0 ± 1.0 ^{c}	3.7 ± 1.7 ^{b}	2.6 ± 1.2 ^{c}

Different letters indicate significant differences between treatments in the same sampling ($p < 0.05$).

3.2. Effects on Olive Tree Physiological Parameters

Measurements of midday stomatal conductance (g_s), net photosynthesis rate (A_N), and maximum electron transport rate (ETR_{max}) on DOY 251, 287, 334, and 418 are presented in Figure 2 and Table S2. On DOY 251, g_s was about 0.20 mol m^{-2}s^{-1} for all the treatments. Nevertheless, on DOY 287, g_s significantly decreased for the OB plots to reach values of about 0.11 mol m^{-2} s^{-1}. Post-harvest measurements (DOY 334 and 418) showed g_s values between 0.10 and 0.12 mol m^{-2} s^{-1}, with no significant differences between treatments. Stomatal conductance (g_s) is considered one of the best indicators of plant water stress, since it is placed at the intersection of water and CO_2 fluxes at the leaf level. In all cases, all these values are above 0.1 mol m^{-2} s^{-1}, indicating that the plants were not water stressed, according to previously published models for the same crop type [54].

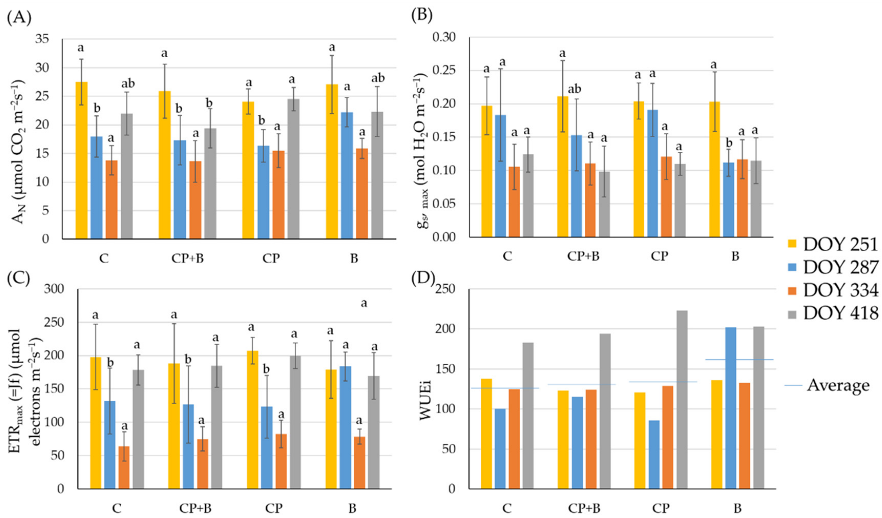

Figure 2. Physiological parameters measured in olive trees: (**A**) net photosynthesis rate (A_N), (**B**) stomatal conductance (g_s), (**C**) maximum rate of electron transport (ETR_{max}), and (**D**) calculated value of intrinsic water-use efficiency (WUEi) at day of the year 251, 287, 334, and 418. C: Control; OB + GC: Olive Biochar+ Green compost; GC: Green Compost; OB: Olive Biochar. Different letters indicate significant differences between treatments in the same sampling ($p < 0.05$).

The net photosynthetic rates (A_N) reached the maximum values (27.5 to 24.1 µmol CO_2 m^{-2} s^{-1}) on day of year 251. A general drop in the A_N values is reported on DOY 287 and 334 without significant differences between the four treatments, except for the increase for the OB amended trees (22.2 µmol CO_2 m^{-2} s^{-1} on day of year 287). Strong seasonal variations in photosynthetic parameters have previously been reported for deciduous species under Mediterranean climate conditions in relation to water availability [55]. The general decline in g_s and A_N values during autumn and the post-harvest period for all the treatments suggests stomata closure, probably due to the cut in the irrigation water supply coupled with a dry autumn season.

A strong linear correlation between g_s and A_N is shown for OB + GC amended plants ($R^2 = 0.63$; $p < 0.05$), and very poor for the control plants ($R^2 = 0.415$; $p < 0.05$). At the leaf level, intrinsic water-use efficiency (WUEi) is defined as the instantaneous ratio between net CO_2 assimilation rate (A_N) and stomatal conductance to water vapor (g_s). WUEi of the olive trees from the biochar-amended plots was on average 20% greater than those of the remaining plots (0.168 vs. 0.137 to 0.140; Figure 2 and Table S2). Values of WUEi in olive have been previously reported by Angelopoulos et al. [53] and Diaz Espejo et al. [56]. Although linear A_N vs. g_s relationships have been reported for olive [57], A_N is affected later than g_s under water stress [58]. This is a key trait for the adaptation of olive tree to drought, and explains usually greater WUEi recorded in plants. Gale and Thomas [29] recently reported that biochar addition resulted in substantial positive effects on plant ecophysiology increasing WUEi with biochar dose up to 30 t ha^{-1}. The disturbance of the g_s-A_N relationship, especially in biochar-amended plots, suggests an alteration of soil water availability due to the application of organic amendments, which may induce modifications in plant physiology. Nevertheless, results of the present study should be taken with care due to the possible excess supply of mineral nutrients by biochar, in particular K, in proportion to N, inducing N deficiency at high biochar doses [29].

A linear relationship between the values of maximum electron transport rate (ETR_{max}) at saturating irradiance and A_N was observed ($R^2 = 0.81$; $p < 0.05$, Figure S2). There were no differences of ETR_{max} between treatments, except for the significant increase that was found in plants from plots amended with biochar during the harvest period ($ETR_{max} = 184$; day of year 287; OB treatment). This result is opposite to the lack of response to biochar addition for the physiology of apple trees reported by Eyles et al. [59]. Cultivars that have lower values of ETR_{max} are less efficient in transporting electrons and potentially fix less CO_2 [60].

Figure 3 shows the evolution of the water potential (ψ_{leaf}) of olive tree leaves measured on DOY 287, 334, and 418. A greater water stress is shown on DOY 287 and 418, and for the plants from the control plots, whose potential reached the most negative values ($\Psi_\pi \leq -2.0$ MPa; day of year 287). However, no significant differences were found in the post-harvest analyses. Irrigation was effective in maintaining the leaf osmotic potential within values considered for non-stressed trees.

3.3. Effects of the Amendment on Plant Yields

Plant yield ranged from 10.1 to 11.9 kg of olives per tree for control and biochar-treated plots, respectively (Table 3). The significant increase in the net fruit weight for biochar-amended olive trees did not result in a linear rise of the estimated oil production, as olives from biochar-amended olive trees contained more moisture and less relative abundance of fat. Thus, this increase in net weight is due to a greater accumulation of water at the fruit from the trees on the biochar-amended plots, which is suitable for table olives. In contrast, a lower water content would be desirable for varieties destined for oil production. The latter could be achieved either by reducing irrigation rates or by delaying the harvest date.

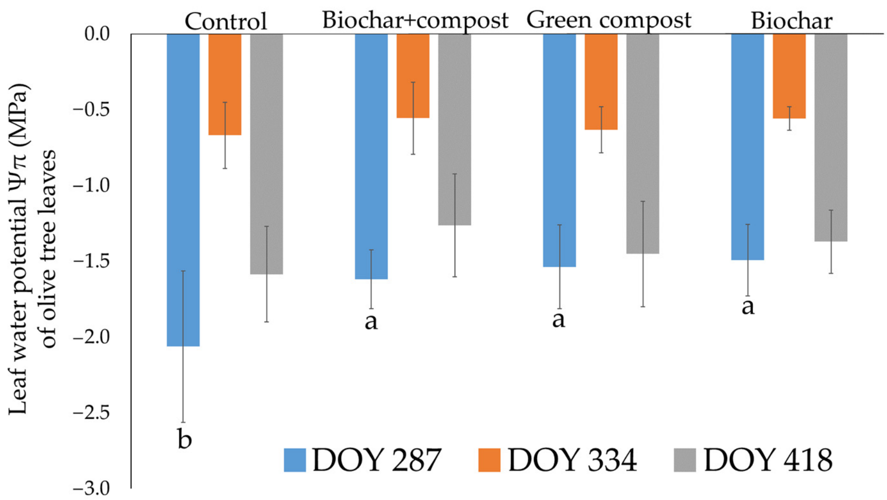

Figure 3. Water potential (ψ_{leaf}) of olive tree leaves measured at DOYs 287, 334, and 418. Different lowercase letters indicate significant differences between treatments in the same sampling ($p < 0.05$), whereas no letter indicate no significant differences.

Table 3. Primary productivity parameters per olive tree for each treatment.

Treatment	Olive Fruit Yield (kg per Tree)	Olive Water Content (% *w*/*w*)	Total Fat (% *w*/*w*)	Oil Free Acidity	Olive Oil Yield (kg per Tree)	Total Fat (% Dry Weight)
C	10.1 ± 0.4 [a]	61.7 ± 0.3 [a]	15.1 ± 0.2 [a]	0.31 ± 0.02	1.52	39.4
GC	10.5 ± 0.3 [a]	61.0 ± 0.4 [a]	15.8 ± 0.4 [a]	0.35 ± 0.03	1.66	40.5
OB + GC	10.8 ± 0.2 [a]	61.7 ± 0.3 [a]	15.2 ± 0.2 [a]	0.31 ± 0.03	1.64	39.7
OB	11.9 ± 0.3 [b]	63.9 ± 0.5 [b]	14.1 ± 0.3 [b]	0.29 ± 0.04	1.68	39.1

Different letters indicate significant differences between treatments for the same parameter ($p < 0.05$), whereas no letter indicates no significant differences.

Regarding olive oil quality, the levels of free acidity ranged from 0.29 (OB) to 0.35 (GC) without significant differences (Table 3). All of them are within the limits established by the International Olive Council (IOC) for extra virgin olive oils (EVOOs). Caruso et al. [61] suggested that olive oil acidity was not influenced by the amount of irrigation and tree water status. In contrast, other authors obtained higher levels of acidity in olive oil from well-watered olive trees [62].

4. Conclusions

Soil physical properties are crucial for crop yields of olive groves because they strongly determine water uptake and transport by plants, which can affect plant physiology and yield. The results of this study have shown that the application of olive pomace biochar (OB) and compost (GC) significantly increased soil moisture and reduced soil resistance to penetrability. Moreover, biochar led to a relative olive production increase of about 15% compared with all other treatments. However, the lower fat and greater moisture content of olives from the OB amended plots attenuated the differences in oil production per tree. Concerning the effects on the plant physiology of olive trees, stomatal conductance showed values of no water stress at any stage. Nevertheless, plants from the control plots showed the most negative water potential values (ψ_{leaf}) on day of year 287, just before the harvesting. On the contrary, olive trees from the biochar-amended plots obtained the greatest net photosynthesis rate (A_N), water-use efficiency (WUEi), and maximum electron transport rate (ETR_{max}), indicative of better water and photosynthetic conditions. The results of this field study show that the application of organic amendments with high porosity and water retention capacity modified the soil physical properties, reducing soil compaction and improving the water status of olive trees in super-intensive olive trees plantations. However, no synergistic effect on the physiological response of olive trees was observed when the mixture of OB and GC was added to the soil. The increase in fruit yield, determined as kg of olives per plant, in the olive trees of the plots amended with biochar was mainly due to a greater water content of the fruits. This finding, together with the better physiological parameters of the plants from the biochar amended plots in the period just before the harvesting, could suggest that the application of olive pomace biochar as soil amendment under super-intensive olive groves could maintain oil yields with a reduction of the irrigation needs. However, long-term studies, including nutrient content and dosage effects, are needed to confirm this finding.

Supplementary Materials: The following are available online at https://www.mdpi.com/article/10.3390/agronomy12102321/s1. Supplementary Figure S1: Representation of linear correlation between soil resistance to penetration and soil moisture (0–5 cm depth); Supplementary Figure S2: Linear correlations between the maximum electron transport rate (ETR_{max}) and net photosynthetic rate (A_N). (a) All treatments; (b) Correlations per treatment: Control, Olive Biochar + Green compost, Green compost, and Olive Biochar. Supplementary Table S1: Soil moisture and resistance to penetrability (in situ parameters) of not amended and amended soils.; Table S2: Physiological parameters measured in olive trees: Net photosynthesis rate (A_N), stomatal conductance (g_s), maximum rate of electron transport (ETR_{max}) and calculated value of intrinsic water-use efficiency (WUEi) at day of the year 251, 287, 334 and 418.

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Data Availability Statement: The soil samples and datasets analyzed during the current study are available from the corresponding author on reasonable request.

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