land

Article

Ten-Year Impact of Cover Crops on Soil Organic Matter Quantity and Quality in Semi-Arid Vineyards

Marco Antonio Jiménez-González [1], Hugo López-Romano [1], Pilar Carral [1], Ana María Álvarez-González [1], Juan-Emilio Herranz-Luque [1], Blanca Esther Sastre-Rodríguez [2], Andrés García-Díaz [2], Gregorio Muñoz-Organero [2] and María José Marques [1,*]

[1] Geology and Geochemistry Department, Universidad Autónoma de Madrid, C/Francisco Tomás y Valiente, 7, 28049 Madrid, Spain; marco.jimenez@uam.es (M.A.J.-G.); anamaria.alvarez@uam.es (A.M.Á.-G.); hl.juane@gmail.com (J.-E.H.-L.)

[2] Madrid Institute for Research and Rural Development in Food and Agriculture (IMIDRA), Finca El Encín, Carretera A-2, km 38.2, Alcalá de Henares, 28805 Madrid, Spain

* Correspondence: mariajose.marques@uam.es

Abstract: Soil organic matter depletion is a significant concern in agricultural soils, impacting crucial aspects of ecosystem health, especially soil properties such as fertility and soil moisture retention. Adopting sustainable soil management practices, such as cover crops, can mitigate this issue. In this study, we analyzed the soil organic carbon (SOC) content and quality in vineyards using two distinct management methods: permanent spontaneous cover crops and conventional tillage. Dissolved organic carbon (DOC) was quantified and chemically characterized using UV–visible spectroscopy. Our results showed an increase of 4.7 Mg C/ha in the carbon stock (50 cm depth) after 10 years of implementing vegetation covers compared with tilled soil. Additionally, cover crop management increased less humified soluble carbon in surface soil layers, while tillage transformed the solubilized carbon. This finding is important because tilled soil becomes more accessible to microbial degradation and leaching, which, in the long term, leads to a SOM content decrease. In conclusion, an increase in carbon stock was observed when using cover crops due to the incorporation of fresh organic matter, whereas tilled soils showed a depletion of carbon stock, including the mobilization of more stable carbon.

Keywords: dissolved organic carbon; groundcovers; carbon sequestration; sustainable land management practice

Citation: Jiménez-González, M.A.; López-Romano, H.; Carral, P.; Álvarez-González, A.M.; Herranz-Luque, J.-E.; Sastre-Rodríguez, B.E.; García-Díaz, A.; Muñoz-Organero, G.; Marques, M.J. Ten-Year Impact of Cover Crops on Soil Organic Matter Quantity and Quality in Semi-Arid Vineyards. *Land* **2023**, *12*, 2143. https://doi.org/10.3390/land12122143

Academic Editors: Chiara Piccini, Rosario Napoli and Roberta Farina

Received: 31 October 2023
Revised: 22 November 2023
Accepted: 24 November 2023
Published: 8 December 2023

1. Introduction

The benefits derived from an optimal soil organic matter (SOM) content have been extensively discussed in the scientific literature [1]. SOM encompasses a range of enhancements in the physical, chemical, and biological characteristics that are crucial for soil quality, such as better structure [2,3], better infiltration [4], or off-site advantages such as improved surface water quality [5]. From a broader standpoint, good SOM levels improve soil functioning [6] and significantly contribute to sustaining food security [7] by promoting drought-resilient ecosystems.

If organic materials are humified, that is, transformed into more stable organic molecules, another environmental benefit arises: the opportunity to mitigate climate change [8–10]. Soils are the most important carbon storage compartment on earth. Peats can hold more than 100 g C/kg, forests can hold 50 g C/kg, and grasslands can hold 25 g C/kg. Arable and permanent crop lands have the most carbon-depleted soils, with values of ca. 20 g C/kg; only bare lands have a lower concentration, with values of 10 g C/kg [11]. This ranking shows that agricultural soils have lost a significant amount of SOM content, with a decline of about 30% on average [12]. Growing cover crops is recognized as a climate-smart agricultural practice globally, and the scientific literature has shown its effects in different contexts. However, their potential is not fully understood in all cropping systems, climatic zones, or soil types [13]. Different climatic and edaphic conditions and management practices must be addressed over a sufficiently long period since the residence time of SOC may vary in different scenarios.

There are diverse ways of returning organic carbon to the soils: reducing the intensity of tillage, conducting fallows and rotations, and using cover crops. In this work, we address the latter, considering the effect of cover crops (CCs) on vineyard soils while considering SOC content and SOM quality.

Vineyard soils in semi-arid areas are notably depleted of organic carbon [11,13–15]. It is worth noting that an increase in SOC does not necessarily equate to SOC sequestration [16]. The quality of SOM refers to its ability to persist in the soil, forming stable SOM. This capability relies on the creation of stable organic matter, which is formed by long-lived materials [17] that can stabilize carbon in the soil for over 1000 years, surpassing the lifespan of living vegetation [18]. Hence, only humified SOM should contribute to carbon sequestration. Humification is considered the prolonged stabilization of carbon against mineralization or degradation; in this sense, humification contributes to carbon sequestration.

Assessing SOC's stability in soil requires an analysis of the nature of the organic matter being incorporated into the soil. SOM comprises approximately 50% carbon [19], existing in various-sized molecules with different concentrations of aromatic structures and polycondensated materials. These structures and materials form a heterogeneous mixture of celluloses, lignin, tannins, proteins, and lipids, among others, found within the soil, ranging from relatively fresh plant residues to highly processed and stable organic compounds. This continuum reflects the varying degrees of decomposition, transformation, and stabilization that organic matter undergoes within the soil environment.

Classic chemical methods for characterizing SOM distinguish fulvic acids (water-soluble molecules at any pH level), humic acids (which are soluble at alkaline pH levels), and humins (non-soluble). This classification is based on a progressive increase in molecular weight, structural complexity, and aromaticity. The different structural nature of SOM molecules involves different properties, one of which is the above-mentioned greater stability and, therefore, lower susceptibility to decomposition/mineralization and the release of CO_2 into the atmosphere.

Based on the velocity of SOM mineralization, researchers have agreed [20] that SOC can be divided into three different pools: fast (1–2 years), intermediate (10–100 years), and slow (100–1000 years). Slow turnover is due to the highly condensed aromatic chemical structure of organic compounds, which are not easily degraded by microorganisms and have a high potential for carbon sequestration [21].

SOC storage capacity is usually studied in topsoil, and the FAO recommends conducting studies at 30 cm depth [22]. This upper layer is the easiest and most frequently studied layer in the scientific literature, but it is also the most variable due to the influence of land management. A SOC increase in deeper layers of soil may improve its long-term effects due to slower decomposition rates [23,24]. It can be argued that this accumulation is driven by the continuous transport of dissolved organic carbon (DOC) within the soil profile [25]. Therefore, studying DOC offers the opportunity to evaluate potential SOC sequestration in deeper layers of soil. Kalbitz et al. [26] found that applying UV spectroscopy to the compositional study of dissolved organic matter from water extraction procedures was accurate and less time-consuming.

Cost-effective methods are crucial in this domain given that the chemical characterization of SOM demands substantial economic and temporal resources. Conversely, studying the chemical composition of SOM with spectroscopy offers a rapid and straightforward means of estimating the degree of maturation and transformation of organic matter, thus shedding light on the effects of different management practices on soil ecosystem services [27–29]. Spectroscopy relies on the various shapes of spectra generated by electrons coming from aromatic or unsaturated SOM. The optical density across different wavelengths can be used as a proxy to study the structure, that is, the stability or quality, of SOM.

This characterization helps determine whether using CCs influences the transformation of organic matter, observing changes in UV–vis absorbance variations. Absorbance ratios at different wavelengths have been widely used to study the humic fraction [30]. Among several ratios, E2/E3 and E4/E6 (absorbance from 250 to 365 nm and absorbance from 465 to 665 nm, respectively) have been used in the literature for this purpose. Aromatic compounds are generally considered more stable and recalcitrant than aliphatic compounds and are often associated with better humification and more complex organic matter structures [31,32] related to the degree of SOM transformation [33]. Moreover, being concentration-independent indices, they have significant potential for comparing different solutions in different environments. As general indicators of the degree of transformation or humification and molecular size, they are useful for studying dissolved organic matter. In both cases, lower values in the ratios of E2/E3 and E4/E6 indicate a higher proportion of aromatic or ring-like organic compounds and a higher molecular weight in the dissolved organic material.

The stability and composition of organic carbon in soils can impact soil fertility, carbon sequestration, and resilience to environmental stressors, such as drought and temperature fluctuations. Carbon chemistry is emerging as another crucial factor along with organic carbon content in soils.

In agricultural and land management contexts, increasing SOC stability is often a goal to enhance soil health and promote long-term carbon storage. Practices like reduced tillage, cover cropping, and adding organic amendments (e.g., compost) can contribute to the accumulation of stable organic carbon in soils. Policies to promote Sustainable Soil Development must be supported by concrete data in different places and soils to better establish deadlines and aid farmers willing to change soil management practices to increase stable SOC content. The scope of this study was conducted in vineyards with poor SOM. We aimed to monitor the influence of cover cropping in vineyard rows after 10 years at depths up to 50 cm to address changes in SOC and DOC content and their stability. We also analyzed CCs' effectiveness in enhancing the goods and services of agricultural soils.

2. Materials and Methods

The vineyard is in an experimental research center called El Encín (Figure 1), belonging to the regional Institute of Agronomic Research in Madrid, Spain (IMIDRA; 40°31′34.0″ N, 3°17′21.5″ W). The climate is semi-arid, defined within the Köppen climate classification as "Csa", indicating a typical Mediterranean climate with hot summers [34], an average annual temperature of 13 °C, and accumulated precipitation of 450 mm. The soil of the entire study area has been defined as Calcisol [35].

The vineyard covers 1.5 hectares and contains 23 different grape varieties planted in 2004 under targeted watering, which have been continuously managed with cover crops (CCs) for the last 10 years. During this period, soil management consisted of tilling alternate rows and conserving CCs in the other rows, comprising permanent spontaneous vegetation mowed in spring with clippings left on the soil surface (Figure 1). Mowing is usually performed twice a year, although it can be more frequent depending on the weather. For these soils, when tillage ceased, the most frequent species covering the soil were: *Sisymbrium sophia*, *Fumaria officinalis*, *Veronica hederifolia*, *Chenopodium album*, *Capsella bursa-pastoris*, and *Conyza canadensis* [36]. Under-vine weed control is performed using mechanical tools to trim the vegetation.

For soil sampling, the vineyard was divided into three blocks (Figure 1). Six samples were obtained randomly in each block, three in rows with CCs and three in rows with tilled soils. In total, 54 samples were analyzed (three blocks × three replicates × two management types (CCs and Till) × three depths). A soil auger with a 5 cm diameter was used to obtain soil samples up to depths of 50 cm. The auger was dug into the same borehole to obtain undisturbed soil cylinders and clear limits of soil depth, from 0 to 15 cm, 15 to 35 cm, and the deepest, 35 to 50 cm. The composite samples of each soil depth are identified as 15, 35, and 50 hereafter.

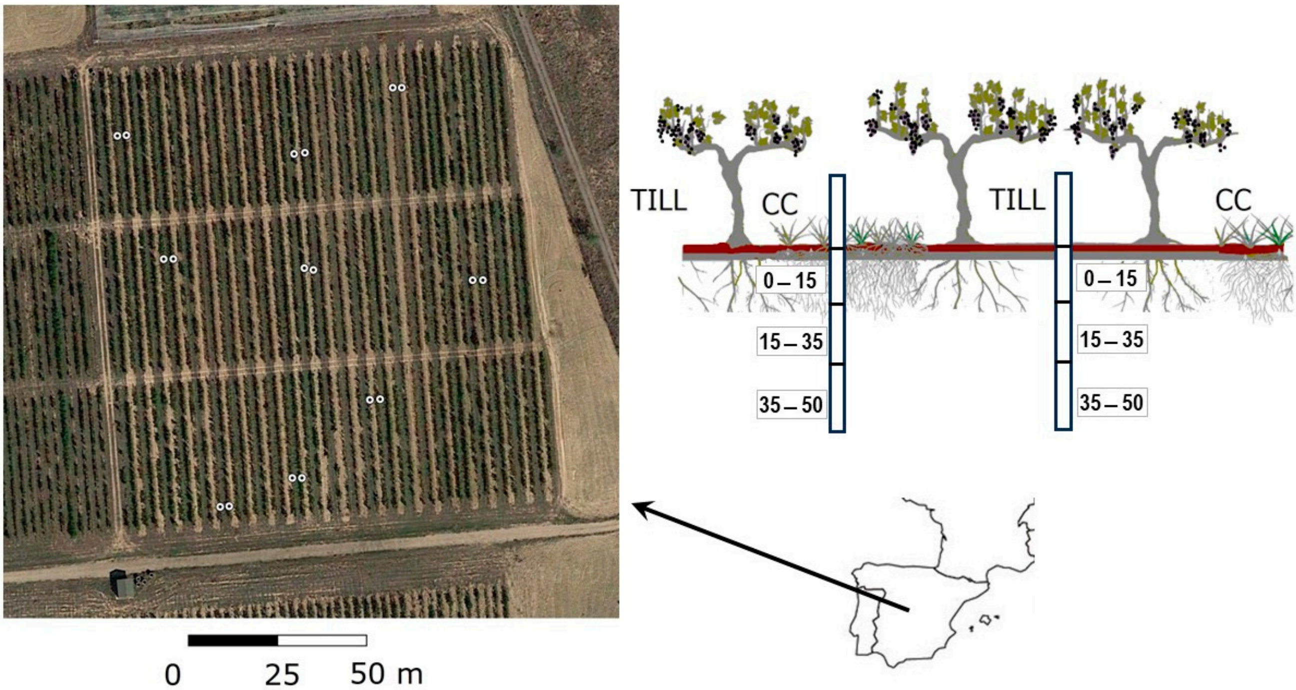

Figure 1. Location of vineyard in Madrid, Spain, with blocks and sampling designs. The vineyard is managed with alternate rows of tillage and cover crops. Three soil samples per block were obtained with augers (right) up to depths of 0–15, 15–35, and 35–50 cm in the middle of the rows, with and without cover crops.

Particle size analysis was conducted using the Bouyoucos method. We first shook 50 g of soil samples (<2 mm) with 400 mL of distilled water and 20 mL of a dispersant solution ($(NaPO_3)_6$). Finally, the suspension was transferred to a test tube and 1 L of distilled water was added until complete. The silt and clay content was estimated by determining the density of the suspension at 40 s (silt + clay) and 2 h (clay) after agitation [37]. The sand content was obtained by sieving (0.05 mesh sieve) and weighing after drying at 105 °C.

The *SOC* analysis was performed with wet oxidation using potassium dichromate in an acid medium [38]. The conversion of *SOC* into organic matter was accomplished using the van Bemmelen factor (1.724). The stock of *SOC* (t/ha) was calculated using the equation:

$$SOC\ (\text{tC/ha}) = SOC\ (\%) \cdot BD\ (\text{g/cm}^3) \cdot [1 - (VG/100)] \cdot LT\ (\text{cm}), \quad (1)$$

where *BD* is the soil bulk density; *VG* (%) is the soil fraction > 2 mm, and *LT* represents soil thickness [14].

DOC was obtained from undisturbed soil samples. The procedure used 2.5 g of previously sieved (2 mm mesh size) dry-room temperature soils in 25 mL of distilled water (1:10 *w*/*v* ratio), which was then agitated (200 rpm) for 15 min in 50 cm^3 polyethylene bottles and filtered using a 0.45 µm Nylon Syringe [39]. Soil extracts were then centrifuged at 4500 rpm for 15 min to eliminate clays. All the extractions were performed in duplicate in the layer samples considered in this study. The aqueous extracts were analyzed using a Multi N/C Analytik Jena Analyzer (Jena, Germany). Liquid samples can be evaluated for both inorganic and total carbon using this method. Subsequently, the soluble organic carbon was determined as the difference between both inorganic carbon and total carbon.

The DOC fraction was also used to measure the wavelength absorbance ranging from 190 to 800 nm using a UV–VIS Genesys 150 spectrophotometer (Thermo Fisher Scientific S.L. Waltham, MA, USA) to obtain a proxy for its molecular composition.

We analyzed differences due to management practices and differences found between soil layers using the Kruskal–Wallis test [40], a non-parametric alternative to one-way variance analysis between groups in Statistica StatSoft Inc. (North Melbourne, Victoria, AU) 8.0 software [41].

3. Results and Discussion

The particle size distribution is crucial to understanding a soil's properties. Our results indicate that the soil has a sandy loam texture (Table 1), no matter the depth considered. There were no significant differences between the samples.

Table 1. Soil texture in different management types and depths.

Management	Depth (cm)	n	Sand (%) 2–0.05 mm	Silt (%) 0.05–0.002 mm	Clay (%) <0.002 mm
Tillage	15	6	71.1 ± 1.6	16.5 ± 1.9	12.4 ± 1.3
	35	6	71.1 ± 1.6	15 ± 1.7	13.9 ± 1.3
	50	6	70.1 ± 1.8	15.8 ± 2.3	14 ± 1.5
Cover crops	15	6	70.4 ± 4.3	16.3 ± 3.7	13.3 ± 0.8
	35	6	70.2 ± 1.6	16.2 ± 1.6	13.6 ± 0.8
	50	6	69.9 ± 4.2	15.7 ± 3.6	14.4 ± 1.1

Mean and standard deviation.

3.1. Soil Organic Carbon

According to the literature, the soils of this experimental farm had 1.1% SOM (0–18 cm depth) in the seventies [42]. Currently, the median shows a value of 0.85 compared with 1.1% forty years ago. A more recent review of organic carbon content in different soils and land uses, published by Calvo de Anta et al. [14], refers to 0.9% as the average value of SOC for depths of 0–30 cm in Calcisols used for woody crop production in this area.

The CC treatment increased the SOM concentration, particularly in the upper layers (Table 2). Tillage, performed in a business-as-usual manner, yielded 0.85% SOM in the topsoil compared with 1.23% under CCs.

Table 2. Differences in soil organic matter (SOM) and bulk density (BD). Significant differences according to the Kruskal–Wallis test between layers in the same management (BL), and between management types for the same layer (BM). Values with different letters showed $p < 0.001$.

Management	Depth (cm)	n	SOM (%) Median	Q25–Q75	BL	BM	BD (Mg m^{-3})
Tillage	15	6	0.85	0.71–0.97	a	a	1.52 a
	35	6	0.45	0.32–0.57	b	a	1.62 a
	50	6	0.46	0.26–0.69	b	a	1.57 a
Cover crops	15	6	1.23	1.11–1.62	a	b	1.64 a
	35	6	0.56	0.49–0.78	b	a	1.56 a
	50	6	0.29	0.2–0.36	c	a	1.62 a

Median and quartiles (Q25–Q75).

In deeper layers, the SOM at 35 cm under CCs was 0.56% compared with 0.45% under tillage without significant differences between management types ($p = 0.27$). There were no significant differences in SOM content at depths of 50 cm.

The increase in SOM produced by CCs led to a significant difference in the topsoil's C stock compared with tillage management ($p < 0.001$). The layer up to 35 cm underneath showed a tendency to increase but not significantly in this period. These results align with others provided for vineyards in Mediterranean environments, e.g., 0.78 Mg C/ha yr [43].

The results for the deepest layer, ranging from 35 to 50 cm, were similar in both treatments (Figure 2).

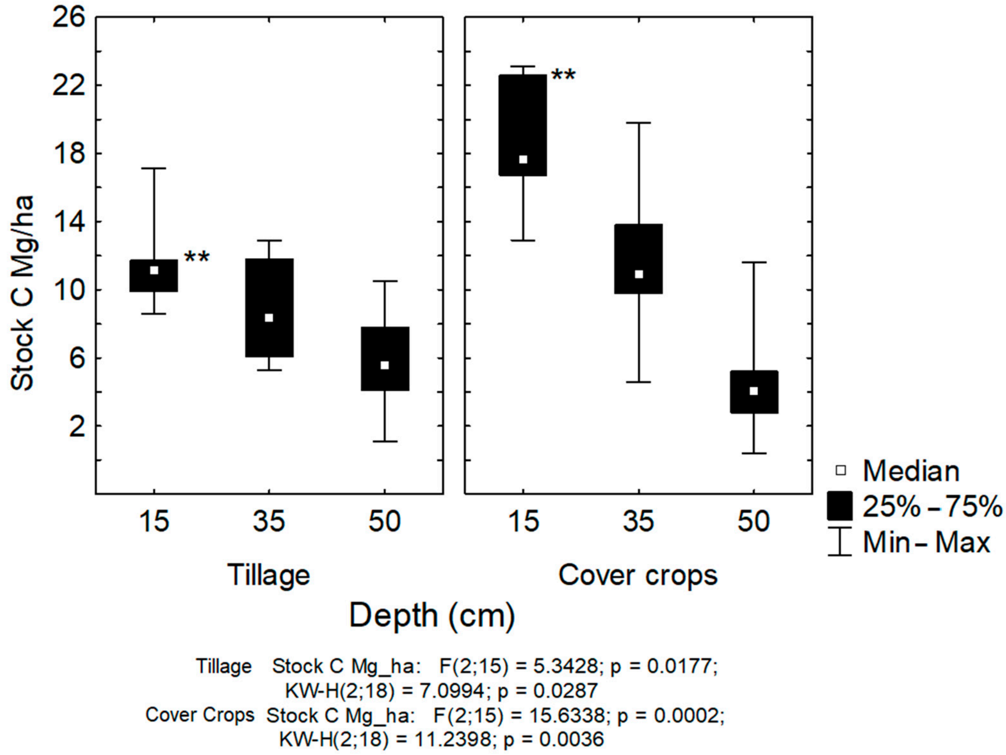

Figure 2. Variation between soil management and soil depth in the organic carbon stock of the three soil layers considered (C stock, Mg/ha). Significant differences between management practices at different depths: ** $p < 0.001$.

One of the most cited effects of CCs on agricultural fields was an increase in soil bulk density. We detected a slightly smaller bulk density value in the topsoil of the tillage treatment, at 1.5 Mg/m^3, compared with 1.6 Mg/m^3 in the rest of the layers and CCs (Table 1). Although not significantly different, this can still influence the calculation of SOC stock. Considering all the studied layers (0–50 cm) and the equivalent mass to avoid the effects of different compaction states [44], the C stock in tilled soils was 24.4 compared with 29.1 Mg C/ha in CC soils. Therefore, this management practice increased by 4.7 Mg C/ha in 10 years compared with the tillage practice. Although the increase in C was not measured yearly, we obtained a proxy for the annual rate of 0.47 Mg C/ha·yr at a 50 cm depth in soils managed with CCs. This value is substantial considering the classical research published by Schlesinger [45], which documented an annual increase of 0.024 Mg C/ha for all ecosystems with significant variations depending on environmental conditions. The magnitude of C stock in these vineyards (24.4–29.1 Mg C/ha) coincides with that from other published research studies on SOC for agricultural soils in Spain, ranging from 10 to 30 Mg C/ha up to 30 cm depth [14], or ca. 30 Mg C/ha for vineyards in France [46].

3.2. Dissolved Organic Carbon

Plant and crop residues that have been recently added to the soil are not typically water-soluble. These materials contain a combination of complex organic compounds, cellulose, lignin, and other organic matter that cannot easily dissolve in water. Over time, as plant residues undergo decomposition by soil microorganisms and other factors, SOM becomes more humified, yielding a generally non-water-soluble organic material. SOM's solubility is primarily influenced by the presence of certain functional groups and the degree of decomposition. DOC includes simple carbohydrates and amino acids, organic acids containing carboxyl and phenolic groups, and organic molecules with high polarity such as hydroxyl and carboxyl groups.

DOC is the most oxidized fraction and, hence, the most chemically and microbially altered fraction of organic compounds in soils [47]. Figure 3 shows that CCs produced a significant increase in DOC. The median value was 9 mg/L in the tillage treatment compared with 11 mg/L in the CC treatment in the topsoil (0–15 cm deep). The layer underneath (35 cm) also experienced a notable increase in DOC due to CCs, from 6.5 to 7.6 mg/L ($p = 0.1$, tillage and CCs median values, respectively). There were no differences at the 50 cm depth, where both management types showed 5.5 mg/L. The conventional significance level is typically set at 0.05 in many scientific studies; however, in the complex soil context of this exploratory analysis, with a small sample size, the limit of $p = 0.1$ may be accepted as a more lenient criterion. These differences are not due to chance and may demonstrate a trend over time.

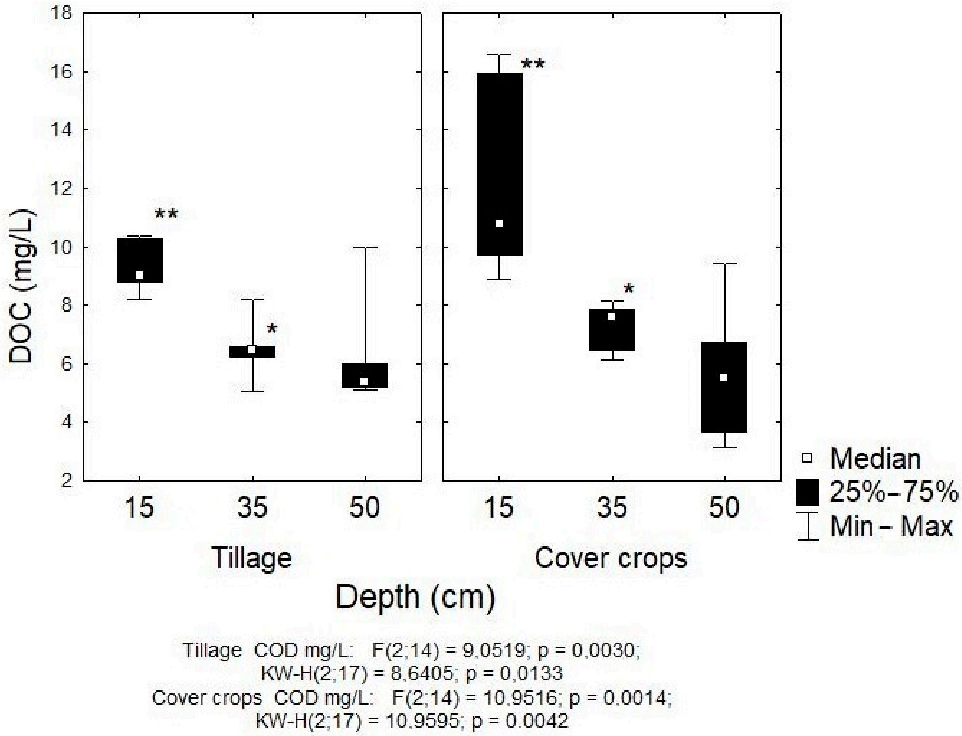

Figure 3. Variation in dissolved organic carbon (DOC, mg/L) between soil management and soil depth. Significant differences between management practices at different depths: ** $p < 0.001$; * $p = 0.10$ between management practices).

The results obtained in this study are consistent with previous findings reported by researchers such as Kaiser and Kalbitz [25]. They argued that the DOC increase throughout depth is a proxy for SOM transport while also representing high SOM cycling ability, which gives positive insight into the framework of soil functions.

3.3. Dissolved Organic Carbon Composition

The SOM in topsoil is formed by recently made photosynthesis molecules, as demonstrated by its radiocarbon signature, and also by DOC [48]. It experiences various degrees of decomposition, transformation, and stabilization that can follow a continuum pattern within the soil profile. Long-term intensive land use has led to an increase in the polycondensation of dissolved organic molecules, which is particularly intense in aqueous extracts of topsoil [49]. In deeper layers, the interaction with mineral surfaces may differ from that of topsoil. These interactions can alter the chemical structure of organic matter and affect its spectral characteristics, possibly resulting in lower ratios of E2/E3 or E4/E6 values depending on environmental conditions. At the same time, microbial communities in deeper soil layers may have different metabolic pathways and preferences for organic matter decomposition compared with surface layers. Microbial processing influences the composition and aromaticity of organic matter. Significant correlations between these ratios and the molecular weight and aromaticity of DOC have been described in previous research [50–52].

Figure 4 represents the optical density at different wavelengths (250, 365, 465, and 665 nm) and the ratios obtained from these wavelengths, E2/E3 and E4/E6. We observed a decrease in the E2/E3 and E4/E6 ratios with depth. The presence of fresh organic matter inputs near the surface in the CC treatment, with materials being less humified, especially influences the E4/E6 absorbance response in this management practice. This fact explains the high values of these ratios in the first 15 cm of the soil profile. At different depths, the organic matter in the solution becomes more humified and stable, leading to lower E2/E3 values in the deeper layers. Similarly, the E4/E6 ratio provides chemical information on the structure of humic to fulvic fractions and shows lower values with depth, suggesting more transformed and polymerized organic matter, which indicates stabilized organic matter. Both indicators show an increase in the chemical structure quality of DOC with depth.

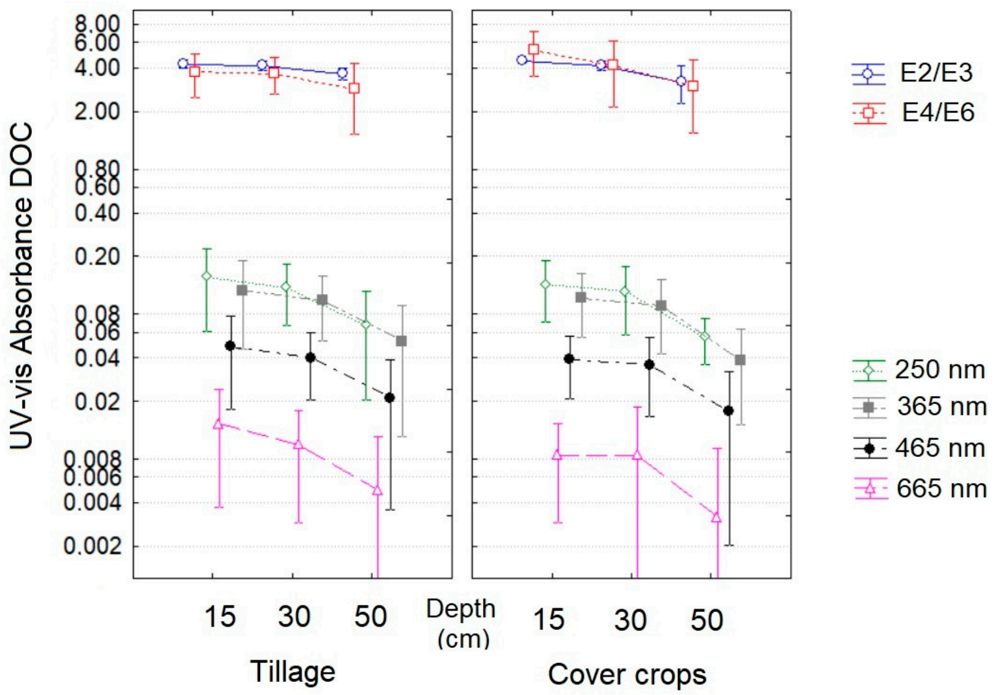

Figure 4. Variation in UV–vis dissolved organic carbon (DOC, mg/L) absorbance between soil management and soil depth (mean and confidence intervals 95%).

Table 3 shows the chemical characterization results of DOC using UV–visible spectroscopy considering the ratios E2/E3 and E4/E6 in the different soil management types. The results show the continuum of absorbance and differences between management types and layers.

As previously mentioned, the E2/E3 ratio is significantly lower in the deepest layer of both soils, which is the expected pattern. A low E2/E3 ratio signifies a high degree of transformation and potentially more oxidized organic matter. Consequently, the organic matter solubilizing in the deep layers of both soils appears to be the most transformed.

We found a significant difference in depth while comparing different management types. Soils managed with CCs showed lower E2/E3 ratios than soils managed with tillage ($p = 0.1$) at the 50 cm depth.

Table 3. Differences in the E2/E3 and E4/E6 ratios. Significant differences according to the Kruskal–Wallis test between layers in the same management type (BL) and between management types for the same layer (BM). Values with different letters showed $p < 0.001$. * $p < 0.1$.

Manage.	Depth (cm)	n	E2/E3 Median	Q25–Q75	BL	BM	E4/E6 Median	Q25–Q75	BL	BM
Tillage	15	6	4.27	4.2–4.3	a	a	3.46	3.1–3.6	a	a *
	35	6	4.30	4.2–4.3	a	a	3.43	2.9–4.8	a	b
	50	6	3.62	3.5–3.9	b	b *	2.98	1.4–3.6	b	b
Cover crops	15	6	4.48	4.4–4.5	a		4.70	4.1–6	a	
	35	6	4.13	3.9–4.5	a		3.96	2.6–5.5	a	
	50	6	3.51	2.5–3.8	b		3.16	1.4–4.2	b	

Median and quartiles (Q25–Q75).

In soils managed with CCs, we can observe how the E4/E6 value decreases over depth. The DOC in the topsoil presents the highest value (4.70), while the lowest is at the 50 cm depth (3.16). These data may indicate that the DOC presents labile structures in the solution obtained from the topsoil while being transformed into deeper layers. It is probably due to the incorporation of new fresh organic matter after cover crops were implemented 10 years ago. Soil management can increase DOC content by mobilizing fresh organic matter through the soil profile and increasing the transformation with depth.

It is worth mentioning that these DOM concentrations were obtained using direct extraction from distilled water. Furthermore, they were not purified of other cations or salts that solubilize. As a result, we found that the E2/E3 ratio was more appropriate for identifying changes in DOC quality between sites because it is more stable and less affected by environmental fluctuations (e.g., heavy rainfalls) due to minor differences between the used wavelengths. On the contrary, the E4/E6 ratio can fluctuate with environmental conditions, for example, at sites with high iron concentrations, which can influence absorbance by 600 nm [52].

When we compare different management types, the increasing trend in the E4/E6 ratio in CC soil suggests the addition of fresh organic carbon. The E4/E6 ratio in the tilled topsoil (Table 3) was 3.46, which is notably smaller than 4.70 in the CC topsoil ($p = 0.08$), indicating a higher proportion of aromatic and more complex and recalcitrant organic compounds in permanently tilled soil's DOM. Changes in the E4/E6 ratio were not detected at the 50 cm depth when managed with CCs. Similarly, there were no differences between management types at the 35 and 50 cm depths.

The E4/E6 index in tilled soils generally registers lower values than it does in soils managed with CCs, except for values at 50 cm, where they are relatively similar. This finding indicates a greater transformation of organic carbon solubilized in tilled soils [53–55]. The migration of this carbon to the liquid phase favors the accessibility of this organic matter to microorganisms promoting its mineralization. We hypothesized that tilled soil has a decreased SOC content for this reason. Another supporting aspect of this statement is that there is no difference between the values at 15 and 35 cm. Both layers show similar transformation DOC degrees, likely due to the tilling effect of mixing surface and subsurface soils. Tillage destroys the soil structure, and deeper soil layers are moved to the surface, exposing them to atmospheric conditions. This process promotes SOM oxidation, leading to mineralization and loss of carbon content. These conditions potentially induce the emergence of more transformed chemical structures of organic matter in the topsoil ($p = 0.08$), coinciding with a decline in carbon content.

Various environmental factors and biological processes can affect these variations in the chemical structure of organic matter. Factors such as temperature and moisture, closely linked to microbial activity, and vegetation type can influence the humification degree and relative abundance of humic and fulvic substances at different depths. New land management practices in vineyards, such as CCs, were found to influence SOC and DOC values with depth.

4. Conclusions

SOC changes occur slowly, especially in deeper soil layers, and may not be uniform across all depths. Detecting statistically significant differences often demands decades of continuous monitoring. In this vineyard, the adoption of CCs has increased by 4.7 Mg C/ha in 10 years (considering the 50 cm depth) compared with traditional tillage. This increase is especially notable in areas with low SOC, such as the site described in this study, with <1% SOM.

(i) Managing vineyards with CCs notably increased SOM in the upper soil layer up to the 15 cm depth ($p < 0.001$), from 0.85 in tilled soils to 1.23% in CCs. Both treatments showed similar results from 35 to 50 cm depths. Although tillage had a slightly lower bulk density (1.5 Mg/m^3) than CCs (1.6 Mg/m^3) in the topsoil layer, it did not differ significantly. The CCs increased C stock by 4.7 t C/ha in ten years. This increase exceeds the average annual increases documented in classical research. The C stock, ranging from 24 to 29 Mg C/ha at a 50 cm depth, aligns with previously conducted studies conducted in Spain and France for agricultural soils and vineyards.

(ii) Concentrations of dissolved organic matter (DOM), obtained using direct extraction with water, were also different depending on management type. CC management notably increased DOC levels in the upper soil layer (0–15 cm), with values of 11 mg/L, which is 20% higher than in tilled soils. It also increased slightly at a depth of 35 cm. The significance level of these differences was $p = 0.1$, which is higher than the typical significance level, but it demonstrates a trend toward an increase over time at a depth of 35 cm. On the contrary, at 50 cm, both management methods showed similar values of 5.5 mg/L. This similarity indicates that management at this depth has no effect after 10 years.

(iii) In terms of organic matter stabilization, areas under CC management displayed elevated E4/E6 ratios, signifying additional fresh organic carbon compared with permanently tilled soils. Regardless of the management approach, as soil depth increases, both the E2/E3 and E4/E6 ratios decrease, indicating a progression toward more stable organic matter at greater depth.

Tilled topsoil layers exhibited lower E4/E6 ratios, suggesting a higher proportion of complex and recalcitrant organic compounds than those under CC management. Tillage disturbs the soil structure, exposing deeper layers and accelerating SOM oxidation. According to some arguments, transformed carbon is transferred to the liquid phase in tilled soils, promoting mineralization, which may decrease the SOC content in these tilled soils. However, the composition of the remaining organic matter is more complex.

Author Contributions: Conceptualization, M.A.J.-G. and M.J.M.; methodology, H.L.-R., A.M.Á.-G., P.C., B.E.S.-R., and A.G.-D.; software, H.L.-R. and J.-E.H.-L.; validation, M.J.M. and M.A.J.-G.; formal analysis, P.C., A.M.Á.-G. and H.L.-R.; investigation, B.E.S.-R., A.G.-D. and G.M.-O.; resources, B.E.S.-R., A.G.-D. and G.M.-O.; data curation, H.L.-R.; writing—original draft preparation, M.A.J.-G. and M.J.M.; writing—review and editing, all authors. All authors have read and agreed to the published version of the manuscript.

Funding: This research was partially funded by the European Joint Programme on Agricultural Soil Management (EJP Soil).

Data Availability Statement: Datasets are available from the authors upon request. The data are not publicly available due to intellectual property/confidentiality issues.

Conflicts of Interest: The authors declare no conflict of interest.

References

1. Wiesmeier, M.; Urbanski, L.; Hobley, E.; Lang, B.; von Lützow, M.; Marin-Spiotta, E.; van Wesemael, B.; Rabot, E.; Ließ, M.; Garcia-Franco, N.; et al. Soil Organic Carbon Storage as a Key Function of Soils—A Review of Drivers and Indicators at Various Scales. *Geoderma* **2019**, *333*, 149–162. [CrossRef]
2. Singh, M.J.; Khera, K.L. Physical Indicators of Soil Quality in Relation to Soil Erodibility Under Different Land Uses. *Arid Land. Res. Manag.* **2009**, *23*, 152–167. [CrossRef]
3. Blavet, D.; De Noni, G.; Le Bissonnais, Y.; Leonard, M.; Maillo, L.; Laurent, J.Y.; Asseline, J.; Leprun, J.C.; Arshad, M.A.; Roose, E. Effect of Land Use and Management on the Early Stages of Soil Water Erosion in French Mediterranean Vineyards. *Soil Tillage Res.* **2009**, *106*, 124–136. [CrossRef]
4. Aljibury, F.K.; Christensen, L.P. Water Penetration of Vineyard Soils as Modified by Cultural Practices. *Am. J. Enol. Vitic.* **1972**, *23*, 35. [CrossRef]
5. Auzet, A.-V.; Le Bissonnais, Y.; Souchère, V. France. In *Soil Erosion in Europe*; John Wiley & Sons: Hoboken, NJ, USA, 2006; pp. 369–383. [CrossRef]
6. Thiele-Bruhn, S.; Bloem, J.; de Vries, F.T.; Kalbitz, K.; Wagg, C. Linking Soil Biodiversity and Agricultural Soil Management. *Curr. Opin. Environ. Sustain.* **2012**, *4*, 523–528. [CrossRef]
7. van der Wal, A.; de Boer, W. Dinner in the Dark: Illuminating Drivers of Soil Organic Matter Decomposition. *Soil. Biol. Biochem.* **2017**, *105*, 45–48. [CrossRef]
8. Lal, R. Soil Carbon Sequestration to Mitigate Climate Change. *Geoderma* **2004**, *123*, 1–22. [CrossRef]
9. Chenu, C.; Angers, D.A.; Barré, P.; Derrien, D.; Arrouays, D.; Balesdent, J. Increasing Organic Stocks in Agricultural Soils: Knowledge Gaps and Potential Innovations. *Soil Tillage Res.* **2019**, *188*, 41–52. [CrossRef]
10. Rumpel, C.; Amiraslani, F.; Chenu, C.; Garcia Cardenas, M.; Kaonga, M.; Koutika, L.-S.; Ladha, J.; Madari, B.; Shirato, Y.; Smith, P.; et al. The 4p1000 Initiative: Opportunities, Limitations and Challenges for Implementing Soil Organic Carbon Sequestration as a Sustainable Development Strategy. *Ambio* **2020**, *49*, 350–360. [CrossRef]
11. de Brogniez, D.; Ballabio, C.; van Wesemael, B.; Jones, R.J.A.; Stevens, A.; Montanarella, L. Topsoil Organic Carbon Map of Europe. In *Soil Carbon*; Springer: Berlin/Heidelberg, Germany, 2014; pp. 393–405. [CrossRef]
12. Murty, D.; Kirschbaum, M.U.F.; Mcmurtrie, R.E.; Mcgilvray, H. Does Conversion of Forest to Agricultural Land Change Soil Carbon and Nitrogen? A Review of the Literature. *Glob. Chang. Biol.* **2002**, *8*, 105–123. [CrossRef]
13. Chiti, T.; Gardin, L.; Perugini, L.; Quaratino, R.; Vaccari, F.P.; Miglietta, F.; Valentini, R. Soil Organic Carbon Stock Assessment for the Different Cropland Land Uses in Italy. *Biol. Fertil. Soils* **2012**, *48*, 9–17. [CrossRef]
14. Calvo de Anta, R.; Luís, E.; Febrero-Bande, M.; Galiñanes, J.; Macías, F.; Ortíz, R.; Casás, F. Soil Organic Carbon in Peninsular Spain: Influence of Environmental Factors and Spatial Distribution. *Geoderma* **2020**, *370*, 114365. [CrossRef]
15. Meersmans, J.; Martin, M.P.; Lacarce, E.; De Baets, S.; Jolivet, C.; Boulonne, L.; Lehmann, S.; Saby, N.P.A.; Bispo, A.; Arrouays, D. A High Resolution Map of French Soil Organic Carbon. *Agron. Sustain. Dev.* **2012**, *32*, 841–851. [CrossRef]
16. Baveye, P.C.; Berthelin, J.; Tessier, D.; Lemaire, G. Storage of Soil Carbon Is Not Sequestration: Straightforward Graphical Visualization of Their Basic Differences. *Eur. J. Soil Sci.* **2023**, *74*, e13380. [CrossRef]
17. Kögel-Knabner, I. The Macromolecular Organic Composition of Plant and Microbial Residues as Inputs to Soil Organic Matter. *Soil Biol. Biochem.* **2002**, *34*, 139–162. [CrossRef]
18. Lützow, M.V.; Kögel-Knabner, I.; Ekschmitt, K.; Matzner, E.; Guggenberger, G.; Marschner, B.; Flessa, H. Stabilization of Organic Matter in Temperate Soils: Mechanisms and Their Relevance under Different Soil Conditions—A Review. *Eur. J. Soil Sci.* **2006**, *57*, 426–445. [CrossRef]
19. Pribyl, D.W. A Critical Review of the Conventional SOC to SOM Conversion Factor. *Geoderma* **2010**, *156*, 75–83. [CrossRef]
20. FAO; ITPS. *Status of the World's Soil Resources (SWSR)—Main Report*; Food and Agriculture Organization of the United Nations and Intergovernmental Technical Panel on Soils: Rome, Italy, 2015; Volume 607.
21. Blair, G.J.; Lefroy, R.D.B.; Lise, L. Soil Carbon Fractions Based on Their Degree of Oxidation, and the Development of a Carbon Management Index for Agricultural Systems. *Aust. J. Agric. Res.* **1995**, *46*, 1459–1466. [CrossRef]
22. FAO; ITPS. Global Soil Organic Carbon Map (GSOCmap) Technical Report, Rome. 2018. Available online: https://www.fao.org/3/I8891EN/i8891en.pdf (accessed on 6 September 2023).
23. Post, W.M.; Kwon, K.C. Soil Carbon Sequestration and Land-Use Change: Processes and Potential. *Glob. Chang. Biol.* **2000**, *6*, 317–327. [CrossRef]
24. Rumpel, C.; Chabbi, A.; Marschner, B. Carbon Storage and Sequestration in Subsoil Horizons: Knowledge, Gaps and Potentials. In *Recarbonization of the Biosphere: Ecosystems and the Global Carbon Cycle*; Lal, R., Lorenz, K., Hüttl, R.F., Schneider, B.U., von Braun, J., Eds.; Springer: Dordrecht, The Netherlands, 2012; pp. 445–464. [CrossRef]
25. Kaiser, K.; Kalbitz, K. Cycling Downwards—Dissolved Organic Matter in Soils. *Soil Biol. Biochem.* **2012**, *52*, 29–32. [CrossRef]
26. Kalbitz, K.; Geyer, S.; Geyer, W. A Comparative Characterization of Dissolved Organic Matter by Means of Original Aqueous Samples and Isolated Humic Substances. *Chemosphere* **2000**, *40*, 1305–1312. [CrossRef] [PubMed]
27. Jimenez-Gonzalez, M.A.; Alvarez, A.M.; Carral, P.; Almendros, G. Influence of Soil Forming Factors on the Molecular Structure of Soil Organic Matter and Carbon Levels. *Catena* **2020**, *189*, 104501. [CrossRef]
28. de Guardia, A.; Brunet, S.; Rogeau, D.; Matejka, G. Fractionation and Characterisation of Dissolved Organic Matter from Composting Green Wastes. *Bioresour. Technol.* **2002**, *83*, 181–187. [CrossRef] [PubMed]

29. Prudent, P.; Domeizel, M.; Massiani, C.; Thomas, O. Gel Chromatography Separation and U.V. Spectroscopic Characterization of Humic-like Substances in Urban Composts. *Sci. Total Environ.* **1995**, *172*, 229–235. [CrossRef]
30. Aranda, V.; Ayora-Cañada, M.J.; Domínguez-Vidal, A.; Martín-García, J.M.; Calero, J.; Delgado, R.; Verdejo, T.; González-Vila, F.J. Effect of Soil Type and Management (Organic vs. Conventional) on Soil Organic Matter Quality in Olive Groves in a Semi-Arid Environment in Sierra Mágina Natural Park (S Spain). *Geoderma* **2011**, *164*, 54–63. [CrossRef]
31. Senesi, N.; D'Orazio, V.; Ricca, G. Humic Acids in the First Generation of EUROSOILS. *Geoderma* **2003**, *116*, 325–344. [CrossRef]
32. Stevenson, F.J. *Humus Chemistry. Genesis, Composition, Reactions*; Wiley: New York, NY, USA, 1994; 496p.
33. Tinoco, P.; Almendros, G.; González-Vila, F.J.; Sanz, J.; González-Pérez, J.A. Revisiting Molecular Characteristics Responsive for the Aromaticity of Soil Humic Acids. *J. Soils Sediments* **2015**, *15*, 781–791. [CrossRef]
34. Aguirre de Carcer, Í.; Carral-González, P. *Apuntes de Meteorología y Climatología Para El Medio Ambiente*; UAM Ediciones: Madrid, Spain, 2013; 346p.
35. WRB, I. *Reference Base for Soil Resources 2014, Update 2015. International Soil Classification System for Naming Soils and Creating Legends for Soil Maps. No. 106*; World Soil Resources Report; FAO: Rome, Italy, 2015.
36. Navarrete Martínez, L.; Sanchez del Arco, M.J.; Hernanz, J.L.; Sánchez-Girón, V.M. Evolución de La Vegetación Arvense En Cultivos de Secano Bajo Diferentes Sistemas de Laboreo Durante Los Últimos Tres Años. In *Herbologia e biodiversidade Numa Agricultura Sustentável.*; Actas XII Congresso da SEMh/XIX Congresso da ALAM/II Congresso da IBCM: Lisboa, Portugal, 2009; pp. 123–126.
37. Boyoucos, G.J. Hydrometer Method Improved for Making Particle Size Analyses of Soils. *Agron. J.* **1962**, *54*, 464–465. [CrossRef]
38. Walkley, A.; Black, I.A. An Examination of the Degtjareff Method for Determining Soil Organic Matter, and a Proposed Modification of the Chromic Acid Titration Method. *Soil Sci.* **1934**, *37*, 29–38. [CrossRef]
39. Jones, D.L.; Willett, V.B. Experimental Evaluation of Methods to Quantify Dissolved Organic Nitrogen (DON) and Dissolved Organic Carbon (DOC) in Soil. *Soil Biol. Biochem.* **2006**, *38*, 991–999. [CrossRef]
40. Kruskal, W.H.; Wallis, W.A. Use of Ranks in One-Criterion Variance Analysis. *J. Am. Stat. Assoc.* **1952**, *47*, 583–621. [CrossRef]
41. StatSoft. *Statistica*; Data Analisys Software System; V10-2011; StatSoft: Tulsa, OK, USA, 2011.
42. *El Encín, Suelo y Clima. Departamento de Ecología. CRIDA 6*; INIA, Ed.; Instituto Nacional de Investigaciones Agrarias: Madrid, Spain, 1977.
43. Vicente-Vicente, J.L.; García-Ruiz, R.; Francaviglia, R.; Aguilera, E.; Smith, P. Soil Carbon Sequestration Rates under Mediterranean Woody Crops Using Recommended Management Practices: A Meta-Analysis. *Agric. Ecosyst. Environ.* **2016**, *235*, 204–214. [CrossRef]
44. Wendt, J.W.; Hauser, S. An Equivalent Soil Mass Procedure for Monitoring Soil Organic Carbon in Multiple Soil Layers. *Eur. J. Soil Sci.* **2013**, *64*, 58–65. [CrossRef]
45. Schlesinger, W.H. Evidence from Chronosequence Studies for a Low Carbon-Storage Potential of Soils. *Nature* **1990**, *348*, 232–234. [CrossRef]
46. Arrouays, D.; Deslais, W.; Badeau, V. The Carbon Content of Topsoil and Its Geographical Distribution in France. *Soil Use Manag.* **2001**, *17*, 7–11. [CrossRef]
47. Huang, Y.; Eglinton, G.; Van Der Hage, E.R.E.; Boon, J.J.; Bol, R.; Ineson, P. Dissolved Organic Matter and Its Parent Organic Matter in Grass Upland Soil Horizons Studied by Analytical Pyrolysis Techniques. *Eur. J. Soil Sci.* **1998**, *49*, 1–15. [CrossRef]
48. Sanderman, J.; Baldock, J.A.; Amundson, R. Dissolved Organic Carbon Chemistry and Dynamics in Contrasting Forest and Grassland Soils. *Biogeochemistry* **2008**, *89*, 181–198. [CrossRef]
49. Kalbitz, K.; Geyer, W.; Geyer, S. Spectroscopic Properties of Dissolved Humic Substances—A Reflection of Land Use History in a Fen Area. *Biogeochemistry* **1999**, *47*, 219–238. [CrossRef]
50. Chin, Y.-P.; Aiken, G.; O'Loughlin, E. Molecular Weight, Polydispersity, and Spectroscopic Properties of Aquatic Humic Substances. *Environ. Sci. Technol.* **1994**, *28*, 1853–1858. [CrossRef]
51. Wang, L.; Wu, F.; Zhang, R.; Li, W.; Liao, H. Characterization of Dissolved Organic Matter Fractions from Lake Hongfeng, Southwestern China Plateau. *J. Environ. Sci.* **2009**, *21*, 581–588. [CrossRef]
52. Peacock, M.; Evans, C.D.; Fenner, N.; Freeman, C.; Gough, R.; Jones, T.G.; Lebron, I. UV-Visible Absorbance Spectroscopy as a Proxy for Peatland Dissolved Organic Carbon (DOC) Quantity and Quality: Considerations on Wavelength and Absorbance Degradation. *Environ. Sci. Process Impacts* **2014**, *16*, 1445–1461. [CrossRef] [PubMed]
53. Chen, Y.; Senesi, N.; Schnitzer, M. Information Provided on Humic Substances by E4/E6 Ratios. *Soil Sci. Soc. Am. J.* **1977**, *41*, 352–358. [CrossRef]
54. Eyheraguibel, B. Caractérisation Des Substances Humiques Biomimétiques- Effets Sur Les Végétaux. Ph.D. Thesis, Sciences des Agroressources, L'institut National Polytechnique de Toulouse, Toulouse, France, 2004.
55. Duval, J. Méthodes D´évaluation de La Maturité Des Composts. Projet Pour Une Agriculture Écologique. 1993. Available online: http://www.eap.mcgill.ca./Agrobio/ab-310-02. (accessed on 6 September 2023).