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Olive Tree Physiology and Productivity Responses under No-Tillage or Digestate Amendment in an Acid Clay Soil

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Abstract: In Mediterranean countries characterized by increasingly extended hot and dry periods, olive trees are often conventionally practiced in low fertility and rainfed soils. This study investigated over a 15-month period how conventional tillage, combined with or without incorporated solid digestate, and no tillage affected selected soil properties, photosynthetic activity and productivity of mature olive trees growing in highly clayey acid soil with an unbalanced nutrient content and Mn excess. Neither in soil nor in drupes were Mn, Fe, Cu and Al contents affected by the managements. However, in soil, exchangeable Mn that was always larger than 200 mg kg^{-1} threshold and unbalanced Ca, Mg, and K contents were evidenced in all treatments. Non-tilled soil showed the highest ($p < 0.05$) stomatal conductance and photosynthetic rate, and the highest ($p < 0.05$) fruits and oil yields. Instead, conventional tillage negatively ($p < 0.05$) affected plant physiology and productivity, likely due to the tilled increase in aeration, enhancing soil water loss and organic C mineralization. Conversely, digestate addition increased TOC, TN and EC. Stomatal conductance, the photosynthetic rate and plant yield significantly recovered (albeit not to no-tillage values) in tillage combined with incorporated digestate, suggesting that digestate-derived organic matter created soil conditions less constraining to plant growth and productivity than the conventional tillage did. Dealing with soil properties and climatic conditions is the key for adopting the best management practice for preserving plant productivity and soil fertility.

Keywords: Mn toxicity; nutrient imbalance; water storage; stomatal conductance; photosynthesis; digestate-DOM

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1. Introduction

Olive (*Olea europaea* L.) represents an extensively and long-held (more than 6000 years) cultivated crop in the Mediterranean dryland area [1]. In 2021, the area under olive tree cultivation in Southern EU countries accounted for approximately 5.0 million ha [2]. The largest cultivated area was recorded in Spain (52.0%), followed by Italy (22.5%), Greece (16.3%), and Portugal (7.6%). France, Slovenia, Cyprus, and Croatia contributed all together with a total cultivated area less than 2%. In most cases, olive tree cultivation is being practiced under rainfed conditions and on soils with a poor fertility status due to, for instance, a low organic matter content, unbalanced nutrient content, adverse physical properties, or characterized by acidic or alkaline excess limits [3]. Although olive tree has shown a preference for neutral and slightly alkaline soils, its cultivation is being expanding to areas with acidic soils, where adverse soil characteristics represent limiting features that severely constrain tree plants productivity [4]. Moreover, another most typical characteristic of Mediterranean olive tree orchards is the presence of mono-cropping with large tree spacing, mostly rainfed farming, and frequent tillage to avoid weed growth in the inter-row [5].

Replacing intensive conventional systems with reduced or no-tillage practices or providing the incorporation of organic amendments have been successfully proposed as improved management systems to contrast soil degradation and the decline in soil fertility [6–10]. The reduction in tillage intensity has shown its potential to protect soil from erosion and to reduce nutrient loss, soil organic matter decline, CO_2 emissions, and preserve soil biodiversity [4,11]. It is also true that the application of organic amendments, such as the recently appeared digestates, can greatly benefit soil fertility, especially in European semi-arid Mediterranean regions [12]. Originated as by-products of the anaerobic digestion (AD) process of mixed organic wastes, digestates are of great potential interest for use in agriculture since they provide essential plant nutrient elements together with easily decomposable organic substrates [13–16]. However, their effects on the soil fertility status can be highly variable, depending on their chemical properties, nutrient and heavy metal content, and biochemical stability [17–21]. For instance, Šimon, et al. [22] found a significant yield increase in winter wheat crops amended with digestate compared to the control, although with no significant differences with mineral fertilizers. Whereas Tan, et al. [23] reported not only a yield increase between 5.19 and 26%, but also a significant increase in macronutrient contents and in both humic- and fulvic-like components in digestate-amended soils.

In a parallel work, the authors focused their attention on assessing changes due to two improved management measures (organic amendment with solid anaerobic digestate and no-tillage) on the chemical and biochemical fertility status in two perennial crop soils (olive grove and citrus grove) with contrasting properties over a one-year observation period [24]. A major finding was that physical and chemical characteristics of the recipient soil represent strong determinants of both the persistence and magnitude of management-induced changes in biochemical and microbial soil properties. Needless to say, soil properties and plant productivity are strongly interconnected.

Then, to gain a comprehensive view of the complex interplay acting at the soil–plant system, we also investigated long-lasting responses of olive trees to solid digestate addition and no-tillage across a two-year time window. A study site characterized by having a clay and acid soil was chosen as it presents more limiting conditions for the growth and productivity of olive trees. The hypothesis assumed for the present study was that the application of certain management techniques, such as no-tillage and incorporation of solid anaerobic digestate, brings about physiological and productive plant responses that are intimately dependent also on the soil type and cannot be widely applied without considering site-specific conditions.

2. Materials and Methods

2.1. Solid Anaerobic Digestate

A local medium-scale (<1 MW) biogas-producing plant provided the solid anaerobic digestate. The plant was constituted by two continuously stirred tank reactors (CSTR) with a total capacity of 7500 m^3 (2500 m^3 tank reactor 1 + 5000 m^3 tank reactor 2) operating at T$\sim$40 °C. The feeding mixture was made of 70% cattle manure, 20% mixed biowastes (solid residues from citrus and olive processing plants, pruning materials, maize silage, and crop residues), and 10% milk serum. The volume loaded was 120 m^3/day, the hydraulic retention time (HRT) was 60 days, and the minimum guaranteed retention time (MGRT) was 16 h at 40 °C. After mechanical separation of the aqueous fraction, which was discarded, the collected solid fraction of the anaerobic digestate was fully characterized [13] to assess compliance with the conformity requirements of the EU Regulation n. 2019/1009. Main properties of the solid anaerobic digestate were as follows: dry matter 18.0%, ash 14.4%, pH 8.77, EC 2140 $\mu S\ cm^{-1}$, total C 39.0%, total N 1.6%, C/N 24.3, NH_4^+-N 5.59 g kg^{-1}, NO_3^--N 0.034 g kg^{-1}, total P 1.24 g kg^{-1}, and total K 2.25 g kg^{-1}. An extended description of this organic by-product is reported by Pathan, et al. [25].

2.2. Site Description and Soil Properties

The field experiment was arranged in a rainfed olive grove (slope < 10%, W exposure, altitude 81 m a.s.l.) located in Southern Italy (Statti Farm, Lamezia Terme, Calabrian Region, Italy; 38°52′31.0′′ N, 16°17′46.0′′ E). Olive trees (*Olea europaea* L. cv. Carolea with a planting distance of 6 × 6 m) were in their productive stage. The study site is characterized by mild and rainy winters and relatively warm and dry summers. The mean annual rainfall and air temperature were, according to the averages recorded over the 1985–2015 period, 1094 mm and +14.3 °C, respectively [26]. During the 2016 and 2017 growing seasons, yearly rainfall and temperature were, respectively, 785 mm and 17.8 °C in 2016, and 661 mm and 17.4 °C in 2017 (Table S1), with slightly shifting climate conditions probably due to ongoing global warming.

The olive grove soil evolves over ancient conoids, forming a terrace plane constituted of Pleistocene sands and brown-reddish conglomerates of metamorphic origin. It shows a typical Ap-Bt-Ct profile (depth > 180 cm) as a result of dominating pedogenetic processes of leaching and illuviation, which have led to increased acidity of the surface horizons and clay migration along the profile with consequent formation of argillic horizons in the subsoil characterized by clay skins (argillans) and FeMn coatings over the faces of peds and walls of voids [27]. Major soil properties are 18.9% sand, 36.1% silt, 45.0% clay, clay texture (USDA), bulk density 1.48 kg dm^{-3}, $pHCaCl_2$ 5.37, pHH_2O 5.54, $EC_{1:2}$ 170 µS cm^{-1}, TOC 21.30 g kg^{-1}, TN 2.03 g kg^{-1}, C/N ratio 10.51, total $CaCO_3$ 0.0 g kg^{-1}, Olsen P 43 mg kg^{-1}, CEC 51.9 $cmol_+$ kg^{-1}, Ca^{2+} 11.12 $cmol_+$ kg^{-1}, Mg^{2+} 2.53 $cmol_+$ kg^{-1}, K^+ 2.68 $cmol_+$ kg^{-1}, Na^+ 0.17 $cmol_+$ kg^{-1}, base saturation 31.8%, Ca/Mg ratio 4.35, and Mg/K ratio 0.94. The available water-holding capacity (AWC, available moisture between the field capacity and the wilting point) equals 170 mm. Soil thermal and moisture regimes (first 150 cm depth) are thermic and udic, respectively [27]. The soil is classified as Typic Hapludalf fine, mixed thermic [28] or Cutanic Profondic Luvisol [24] and has been kept continuously cultivated with olive trees and periodically tilled in the 0–20 cm layer since the mid-1950s.

2.3. Experimental Design and Soil Management

The experimental design consisted of 75 × 18 m field plots arranged in a randomized complete block design, with three replications, to compare the following three treatments: (1) no-tillage (NT), where weeds were mechanically removed and left upon the soil surface as a residue mulch; (2) conventional tillage (TILL) consisting of an inter-row harrowing (~20 cm) followed by a slight rolling; and (3) conventional tillage coupled with soil incorporation of the solid fraction of anaerobic digestate (DIG) at a rate of 30 Mg ha^{-1}, a dose close to that usually employed in agriculture and to that suggested by Caracciolo, et al. [29] and Fernández-Bayo, et al. [30] in their field trials testing C and N mineralization (Figure 1). The digestate was supplied during the 2016 growing season at the beginning of the trial (May 2016), while plant responses to the treatments were monitored over the following season in order to evaluate the mid-term effects on plant physiology in accordance with what was reported by Knoop, et al. [31] and Jaša, et al. [32]. Chemical fertilizers were applied only during the 2016 growing season (in early spring) before the start of the trial. All field plots were fertilized at once with 400 kg ha^{-1} of a 20N-10P_2O_5-10K_2O chemical fertilizer, supplying 80 kg N ha^{-1}, 18 kg P ha^{-1} and 34 kg K ha^{-1}. Each treatment was assigned to 36 trees, divided into three plots of 12 trees each. Each plot included three rows of trees. To avoid any border effect, only the central row of each plot was considered, and all measurements, and plant and soil samples were taken from the inner trees of the central row.

(**a**) (**b**) (**c**)

Figure 1. View of the three treatments, (**a**) NT, (**b**) DIG and (**c**) TILL (as in M&M), of the olive orchard at the beginning of the field trial (May 2016).

2.4. Soil Sampling and Analysis

Soil samples were collected 6 days before (T0, early May) and then 2 days (T1, May), 7 weeks (T2, late June), 18 weeks (T3, mid-September) and one year (T4, early May) after the treatment applications. Three individual non-rhizosphere soil cores (approx. 200 g each) were surface collected (Ap horizon, 0–20 cm soil layer) and then accurately mixed to form a unique composite sample. Three composite samples (each from 9 individual inter-row soil cores) were taken per treatment. Nine composite soil samples were collected (3 treatments × 3 replicates) at each sampling time in order to have 45 composite soil samples at the end of the experiment. Field-moist samples were air-dried, sieved to a 2 mm particle size, and then stored at room temperature before chemical analysis. Soil chemical properties were determined according to the standard methods recommended by the Soil Science Society of America [33]. Briefly, soil acidity was potentiometrically measured in a 1:2.5 (w/v) soil-to-0.01 M $CaCl_2$ solution mixture ($pHCaCl_2$); electrical conductivity was measured at 25 °C in a 1:2 (w/v) soil-to-water ratio slurry ($EC_{1:2}$ 25 °C). Total organic C and N were determined using the elemental analyzer LECO CN628 (LECO Corporation, St. Joseph, MI, USA).

2.5. Gas Exchange and Chlorophyll Fluorescence Measurements

Gas exchange measurements (photosynthetic rate, stomatal conductance, and transpiration rate) were measured on 54 mature leaves from the outer layer of each tree (6 leaves × 3 plants × 3 blocks) using a portable photosynthesis system (LI-COR 6400XT; LI-COR Biosciences, Lincoln, NE, USA). Data collection was limited to 3 plants per treatment to avoid the risk of collecting data over a too extended time range with changing environmental conditions.

Chlorophyll fluorescence parameter data were determined using a chlorophyll fluorimeter (LI-COR 6400-40; LI-COR Biosciences) from clips of the same mature leaves chosen for gas exchange measurements after 30 min preconditioning in the dark. The following chlorophyll fluorescence parameters were determined: F0 minimum fluorescence and Fm maximum fluorescence. These values were subtracted and divided [(Fm − F0)/Fm] to calculate the maximum quantum efficiency of photosystem II (PSII) photochemistry (Fv/Fm), which provides quantitative information on the current state of plant performance under stress conditions [34]. The non-photochemical quenching coefficient (NPQ) was also calculated.

All these measurements were carried out with mature, fully expanded leaves from the middle of the main shoot. Measurements were performed at natural full light intensity (from 12:00 a.m. to 01:00 p.m.) during clear sunny days in July (both in 2016 and 2017, Table S1), when olive plants were expected to undergo the most stressful condition in terms of high temperature and water deficit.

2.6. Olive Tree Fruit Sampling and Processing

Ten adult, healthy, randomly chosen olive trees were assigned to each treatment. At harvest (in November 2017), 100 fruits were randomly sampled to measure the average fruit weight, and the total number of fruits per tree was calculated by dividing the crop yield by the average fruit weight [35]. The resistance to detachment (i.e., the force required to detach the fruit peduncle from the branch) was evaluated using a dynamometer TR 53208 (Turoni®, Forlì, Italy). Soon after sampling, forty drupes per treatment were immediately used to determine the following biometric parameters: equatorial (D) and polar diameter (H) using a precision caliber; after skin removal, pulp firmness (PF) was measured using a penetrometer with a 3 mm diameter tip set on opposite sides of the equatorial zone of the fruit (fruit firmness tester, Turoni®); the skin color was measured using a portable Minolta spectrophotometer CM-700d (Minolta, Inc., Tokyo, Japan), and expressed in terms of CIELab and HSB color spaces; fresh (FW) and dry (DW) weights were determined gravimetrically before and after oven drying at 60 °C for 72 h. The oil content of the fruit mesocarp (pulp) was measured using a Soxhlet extraction (Soxhlet extractor E-812/816, BUCHI, Flawil, Switzerland). The oil yield of individual trees was calculated after measuring the mesocarp oil content on a dry weight basis, the fruit fresh yield, the pulp-to-fruit ratio, and the ratio between DW and FW [36].

The remaining sixty fresh fruits per treatment (Figure 2) were gently washed with deionized water and stored at −80 °C in plastic bags prior to chemical analysis.

(**a**) (**b**) (**c**)

Figure 2. Size and color of drupes collected from the three different treatments, (**a**) NT, (**b**) DIG and (**c**) TILL (as in the M&M), at the end of the observation period (November 2017).

2.7. Extraction and Quantification of Mn, Fe, Cu and Al from Soil and Olive Tree Fruits

Exchangeable Mn, Fe, Cu, and Al were soil-extracted with 0.1 M $CaCl_2$ (1:10, *w*/*v*, soil solution) under shaking for 16 h at room temperature. After centrifugation (3000 *g*, 5 min) and filter paper filtration (Whatman® no. 42), clean filtrates were stored at −20 °C before analysis.

Pulps from twenty drupes per treatment were crunched in a mortar to form a homogeneous paste. An aliquot equivalent to 0.5 g dry weight was acid digested in a HNO_3–H_2O_2 (9:1, *v*/*v*) solution using a microwave (Milestone Ethos Easy, Sorisole, Italy) oven operating at 200 °C for 30 min. Clean extracts were brought to 100 mL of volume with ultrapure deionized water and then stored at −20 °C before analysis.

Analytical measurements of Mn, Fe, Cu, and Al both in soil and olive tree fruit extracts were carried out using inductively coupled plasma-optical emission spectrometry (ICP OES, Perkin Elmer Optima 8000, Walthan, MA, USA).

2.8. Statistical Analysis

Soil and plant data shown in Tables 1–8 were analyzed with a multiple pairwise comparison of means (Tukey's HSD test at $p < 0.05$). Soil chemical data shown in Tables 1 and 2 and physiological data in Tables 4 and 5 were first tested for deviation from normality (Kolmogorov–Smirnov test) and homogeneity of within-group variances (Levene's test) before the two-way ANOVA (Time × Management). The statistical analysis was performed by using SAS 9.3 software (SAS Institute, Cary, NC, USA).

3. Results

3.1. Soil Chemical Properties

The two-way ANOVA revealed that time, soil management, and, in most cases, their interaction significantly influenced the major soil chemical variables (Table 1). Precisely, soil pH showed a time-dependent declining trend in all treatments (−5% on average), even though the amendment with anaerobic digestate brought about a significant (+7%) and transient (only at T1) increase soon after its addition. Seasonal fluctuations in EC were observed in all treatments, being greater in magnitude (up to 3-fold higher than the T0 value) and in persistence (still appreciable at the final stage) in DIG-treated soil. Note also that soil tillage raised the EC by 52% (at T1, T2, and T3) and remained still significantly higher than in NT, although lowering by roughly 40% in T4 compared to T1 (Table 1). Finally, the addition of solid digestate immediately increased both soil TOC and TN (by 30%) in a way that values higher than the initial ones were still noticeable at the final stage. A slight increase in the no-tillage treatment was also noticed over the 1-year observation period when compared to conventionally tilled soil (Table 1).

Table 1. Variations in soil chemical properties (means ± SD, n = 3) under different treatments (NT, DIG, and TILL) at five sampling times (as in the M&M) during the 2016/2017 growing season.

Variable	Management	Sampling Time				
		T_0	T_1	T_2	T_3	T_4
pH_{CaCl2}	NT	5.35 ± 0.07	5.28 ± 0.06 ^{b}	5.13 ± 0.06	5.18 ± 0.18	5.15 ± 0.24
	DIG	5.41 ± 0.25	5.79 ± 0.14 ^{a}	5.17 ± 0.12	5.42 ± 0.19	5.49 ± 0.29
	TILL	5.37 ± 0.14	5.17 ± 0.24 ^{b}	5.06 ± 0.16	5.12 ± 0.13	5.05 ± 0.09
	T: $F_{4,30}$ = 5.60 **; M: $F_{2,30}$ = 16.86 ***; T × M: $F_{8,30}$ = 2.02 *					
$EC_{1:2}$				μS cm^{-1}		
	NT	162.7 ± 23.6	164.1 ± 23.1 ^{c}	198.7 ± 15.8 ^{c}	253.8 ± 31.1 ^{b}	195.3 ± 22.0 ^{b}
	DIG	159.0 ± 25.8	485.3 ± 17.8 ^{a}	389.5 ± 19.8 ^{a}	347.0 ± 22.6 ^{a}	278.8 ± 11.3 ^{a}
	TILL	173.4 ± 40.2	263.8 ± 41.6 ^{b}	255.6 ± 29.8 ^{b}	246.3 ± 21.0 ^{b}	174.3 ± 13.7 ^{b}
	T: $F_{4,30}$ = 64.88 ***; M: $F_{2,30}$ = 118.08 ***; T × M: $F_{8,30}$ = 20.03 ***					
TOC				mg kg^{-1}		
	NT	20.2 ± 2.1	20.3 ± 1.4 ^{b}	19.7 ± 1.4 ^{b}	22.2 ± 0.7 ^{b}	24.7 ± 1.8 ^{b}
	DIG	22.0 ± 2.8	28.8 ± 1.3 ^{a}	24.9 ± 0.4 ^{a}	25.7 ± 2.3 ^{a}	28.0 ± 1.4 ^{a}
	TILL	19.6 ± 0.9	23.5 ± 2.5 ^{b}	21.1 ± 2.4 ^{b}	19.2 ± 1.0 ^{c}	22.2 ± 1.3 ^{b}
	T: $F_{4,30}$ = 5.14 **; M: $F_{2,30}$ = 23.56 *; T × M: $F_{8,30}$ = 2.33 *					
TN				mg kg^{-1}		
	NT	1.8 ± 0.1	1.9 ± 0.1 ^{b}	1.8 ± 0.2 ^{b}	2.2 ± 0.1 ^{ab}	2.0 ± 0.1 ^{ab}
	DIG	1.9 ± 0.2	2.6 ± 0.1 ^{a}	2.3 ± 0.1 ^{a}	2.4 ± 0.3 ^{a}	2.3 ± 0.2 ^{a}
	TILL	1.8 ± 0.1	1.9 ± 0.1 ^{b}	1.8 ± 0.1 ^{b}	1.9 ± 0.1 ^{b}	1.8 ± 0.2 ^{b}
	T: $F_{4,30}$ = 4.99 **; M: $F_{2,30}$ = 7.36 **; T × M: $F_{8,30}$ = 1.92 ^{ns}					

When present, different letters in a column indicate significant differences within each sampling time (Tukey's HSD at $p < 0.05$). Significant effects due to time (T), soil management (M), and their interaction are presented as F-values and the level of significance (* $p < 0.05$; ** $p < 0.01$; *** $p < 0.001$; ns $p > 0.05$) estimated using two-way ANOVA (time × management).

3.2. Soil Exchangeable Mn, Fe, Cu, and Al

The statistical analysis revealed that soil management did not result in a main factor influencing the variability of exchangeable micronutrients and aluminum (Table 2), whereas time was an effective factor affecting the variability of Mn and Fe. Regarding this latter point, time-dependent changes in soil exchangeable Mn were especially found in both tilled plots. An opposite behavior was shown in exchangeable Fe content, which declined most in tilled plots (Table 2). No factor influenced the exchangeable soil Cu, which remained practically unvaried in all treatments at any sampling time. A significant time × treatment interaction was found only for Mn and Al (Table 2). Considering the average contents of soil exchangeable micronutrients, the following trend was found: Fe < Cu << Mn. Thus, a strong iron deficiency (well below 2 mg kg^{-1}), a slightly deficient level of copper, and an extremely high content of soil exchangeable Mn was revealed, particularly in DIG-treated plots. Finally, the exchangeable Al soil content showed marked time-dependent fluctuations, with no significant differences among treatments (Table 2).

Table 2. Variations in exchangeable Mn, Fe, Cu, and Al (means ± SD, *n* = 3) in soil under different treatments (NT, DIG, and TILL) at five sampling times (as in the M&M) during the 2016/2017 growing season.

Metal	Management	Sampling Time				
		T_0	T_1	T_2	T_3	T_4
Mn				mg kg^{-1}		
	NT	247.0 ± 35.4	248.3 ± 22.9	237.5 ± 64.2	256.3 ± 28.7	248.9 ± 25.6
	DIG	247.4 ± 24.4	298.1 ± 28.3	241.7 ± 9.0	276.0 ± 54.2	247.8 ± 7.8
	TILL	229.2 ± 35.0	261.5 ± 91.2	199.3 ± 29.7	232.7 ± 39.8	250.0 ± 28.7
	T: $F_{4,30}$ = 8.95 *; M: $F_{2,30}$ = 3.18 ^{ns}; T × M: $F_{8,30}$ = 10.25 *					
Fe				mg kg^{-1}		
	NT	0.04 ± 0.07	0.03 ± 0.05	0.12 ± 0.05	0.07 ± 0.07	0.02 ± 0.03
	DIG	0.03 ± 0.06	b.d.l.	0.03 ± 0.05	0.01 ± 0.02	0.01 ± 0.02
	TILL	0.22 ± 0.12	0.02 ± 0.04	0.05 ± 0.05	0.06 ± 0.10	0.04 ± 0.06
	T: $F_{4,30}$ = 6.53 *; M: $F_{2,30}$ = 3.91 ^{ns}; T × M: $F_{8,30}$ = 4.61 ^{ns}					
Cu				mg kg^{-1}		
	NT	0.57 ± 0.35	0.47 ± 0.28	0.73 ± 0.32	0.48 ± 0.27	0.62 ± 0.24
	DIG	0.47 ± 0.17	0.54 ± 0.12	0.71 ± 0.35	0.40 ± 0.13	0.55 ± 0.11
	TILL	0.74 ± 0.18	0.48 ± 0.31	0.56 ± 0.15	0.34 ± 0.08	0.63 ± 0.27
	T: $F_{4,30}$ = 0.10 ^{ns}; M: $F_{2,30}$ = 0.17 ^{ns}; T × M: $F_{8,30}$ = 0.36 ^{ns}					
Al				mg kg^{-1}		
	NT	0.61 ± 0.68	0.42 ± 0.70	0.84 ± 0.89	0.20 ± 0.17	0.34 ± 0.55
	DIG	0.41 ± 0.72	b.d.l.	0.65 ± 0.12	b.d.l.	0.07 ± 0.13
	TILL	0.68 ± 0.82	0.13 ± 0.22	1.51 ± 2.22	0.11 ± 0.18	0.32 ± 0.56
	T: $F_{4,30}$ = 0.08 ^{ns}; M: $F_{2,30}$ = 0.84 ^{ns}; T × M: $F_{8,30}$ = 0.99 *					

b.d.l. = Below detection limit. When present, different letters in a column indicate significant differences within each sampling time (Tukey's HSD at $p < 0.05$). Significant effects due to time (T), soil management (M), and their interaction are presented as *F*-values and the level of significance (* $p < 0.05$; ns $p > 0.05$) estimated using two-way ANOVA (time × management).

3.3. Total Mn, Fe, Cu, and Al Contents in Olive Tree Fruits

Metal contents in pulps of olive tree fruits did not evidence any significant differences among treatments, except in the case of copper, which was found to be higher in DIG and NT in comparison with TILL (Table 3).

Table 3. Total Mn, Fe, Cu, and Al contents (mean ± SD, *n* = 3) in olive tree fruits under different treatments (NT, DIG, and TILL) during the 2016/2017 growing season.

Management	Metal			
	Mn	Fe	Cu	Al
		mg kg^{-1}		
NT	5.20 ± 0.79	12.12 ± 1.49	16.60 ± 2.09 ^{a}	4.50 ± 0.66
DIG	4.51 ± 0.31	10.94 ± 1.21	17.25 ± 0.66 ^{a}	4.97 ± 4.27
TILL	4.25 ± 0.15	13.33 ± 2.64	11.37 ± 2.31 ^{b}	6.33 ± 3.39

When present, different letters in a column indicate significant differences within each sampling time (Tukey's HSD at $p < 0.05$).

3.4. Gas Exchange and Chlorophyll Fluorescence Measurements

The two-way ANOVA revealed that soil management was the main factor influencing the variability of physiological responses related to the photosynthetic and transpiration rates (Table 4). Conversely, time influenced the photosynthetic rate and the stomatal conductance, but not the transpiration rate. It is worth noting that, in addition to season-dependent fluctuations in absolute values, gas exchange measurements (that is CO_2 fixed during the photosynthetic process and H_2O lost by transpiration through stomatal openings) showed a similar trend among treatments (that is: NT > DIG > TILL), which was maintained unaltered across the 2-year field experiment (Table 4). In particular, olive trees from NT showed leaf gas exchange readings that were always higher than those from DIG and TILL. Differently, olive tree plants from DIG-treated plots showed values for the photosynthetic rate, stomatal conductance and transpiration rate that were significantly higher than those from TILL only during 2016 (when the solid anaerobic digestate was applied), but not during the following growing season (Table 4).

Table 4. Gas exchange measurements (photosynthetic and transpiration rate) and stomatal conductance (mean ± SD, n = 3) of leaves of olive trees under different treatments (NT, DIG, and TILL as in the M&M) during the July sampling of the 2016 and 2017 growing seasons.

Year	Management	Photosynthetic Rate	Stomatal Conductance	Transpiration Rate
		μmol CO_2 m^{-2} s^{-1}	mol H_2O m^{-2} s^{-1}	mmol H_2O m^{-2} s^{-1}
2016	NT	12.32 ± 0.67 ^{a}	0.16 ± 0.04 ^{a}	1.97 ± 0.33 ^{a}
	DIG	10.59 ± 0.54 ^{b}	0.12 ± 0.04 ^{a}	1.50 ± 0.07 ^{b}
	TILL	4.93 ± 0.27 ^{c}	0.05 ± 0.02 ^{b}	0.94 ± 0.20 ^{b}
2017	NT	9.09 ± 0.84 ^{A}	0.08 ± 0.01 ^{A}	1.68 ± 0.09 ^{A}
	DIG	6.05 ± 0.32 ^{B}	0.05 ± 0.01 ^{B}	0.98 ± 0.15 ^{B}
	TILL	4.50 ± 0.39 ^{B}	0.05 ± 0.01 ^{B}	1.03 ± 0.18 ^{B}
Photosynthetic rate T: $F_{1,27}$ = 36.38 ***; M: $F_{2,27}$ = 58.90 ***; T × M: $F_{2,27}$ = 7.15 **				
Stomatal conductance T: $F_{2,27}$ = 7.52 *; M: $F_{2,27}$ = 4.25 *; T × M: $F_{2,27}$ = 1.89 ^{ns}				
Transpiration rate T: $F_{2,27}$ = 2.67 ^{ns}; M: $F_{2,27}$ = 11.32 ***; T × M: $F_{2,27}$ = 1.40 ^{ns}				

When present, different letters in a column and within each sampling time indicate significant differences among treatments (Tukey's HSD at p < 0.05). Significant effects due to time (T), soil management (M), and their interaction are presented as F-values and the level of significance (* p < 0.05; ** p < 0.01; *** p < 0.001; ns p > 0.05) estimated using two-way ANOVA (time × management).

As for changes in the absolute values between the two years, the photosynthetic rate, stomatal conductance, and transpiration rate declined, respectively, by ~26%, ~50%, and ~15% in NT-treated olive plants; these physiological parameters decreased by ~43%, ~59%, and ~35% in the DIG treatment. Whereas only a slight decrease (ca 8%) in the photosynthetic rate was observed in olive plants from TILL (Table 4).

The same as seen for gas exchange measurements, physiological parameters representing photosystem II activity in plants under the differing treatments followed the trend TILL < DIG < NT across the 2-year experiment (Table 5). Note that absolute values generally increased from the sampling of July 2016 to that of July 2017. Precisely, the observed increases in Fv/Fm and NPQ were +20.7% and +29.4%, respectively, in the NT treatment, and were equal to +4.2% and +2.6% in DIG treatment. Whereas both parameters remained practically unaltered in TILL treatment (Table 5). The strong effect of time and management was separately considered and the effect of their interaction on Fv/Fm and NPQ was confirmed by the two-way ANOVA.

Fruit and oil yields responded significantly to the differing management practices, resulting in higher values under NT and being decreased in DIG and TILL treatments (Table 6). However, the mesocarp oil content result was lower in DIG-treated plants, and significantly decreased in TILL when compared to NT. Olive oil yield showed significant differences among treatments, being the highest under no-tillage management and the lowest in DIG-treated plants (Table 6).

Weights and biometric properties of olive tree fruits followed the same trend of DIG < TILL < NT seen before, resulting in fruits from DIG-treated olive plants that were the smallest in dimension and the lightest in weight (Table 7). It is also worth noting that there was an opposite trend between the fruits per tree and the fruit weight, whereas under conventional tillage the number of fruit per tree markedly declined. In addition, fruit characteristics such as pulp firmness and dynamometer measures suggest that drupes from the DIG treatment more rapidly ripened due to the lowest force needed to detach them from the tree branch (Table 7). However, the colorimetric indices did not show any differences among olive fruits from differing soil treatments (Table 8).

Table 5. Photosystem II activity, as expressed by the chlorophyll fluorescence parameters Fv/Fm and non-photochemical quenching coefficient (NPQ) (mean ± SD, n = 3), of leaves of olive trees under different treatments (NT, DIG, and TILL as in the M&M) during the July sampling of the 2016 and 2017 growing seasons.

Year	Treatment	F_v/F_m	NPQ
2016	NT	0.53 ± 0.01^{a}	2.14 ± 0.06^{a}
	DIG	0.47 ± 0.03^{ab}	1.91 ± 0.11^{b}
	TILL	0.40 ± 0.04^{b}	1.70 ± 0.12^{b}
2017	NT	0.64 ± 0.01^{A}	2.77 ± 0.04^{A}
	DIG	0.49 ± 0.01^{B}	1.96 ± 0.02^{B}
	TILL	0.41 ± 0.01^{C}	1.71 ± 0.02^{B}
F_v/F_m T: $F_{1,27}$ = 68.22 **; M: $F_{2,27}$ = 12.29 ***; T × M: $F_{2,27}$ = 6.52 **			
NPQ T: $F_{1,27}$ = 26.54 ***; M: $F_{2,27}$ = 103.89 ***; T × M: $F_{2,27}$ = 21.01 ***			

When present, different letters in a column and within each sampling time indicate significant differences among treatments (Tukey's HSD at $p < 0.05$). Significant effects due to time (T), soil management (M), and their interaction are presented as F-values and the level of significance (** $p < 0.01$; *** $p < 0.001$) estimated using two-way ANOVA (time × management).

Table 6. Plant yield and fruit characteristics at harvest from olive trees under different treatments (NT, DIG, and TILL) during the 2016/2017 growing season.

Treatment	Fruit Yield kg Olives Tree^{-1}	Pulp-To-Fruit Ratio	Mesocarp Oil % DW	Oil Yield kg Plant^{-1}
NT	25.3 ± 3.15^{a}	39.12 ± 3.16	60.24 ± 1.50^{a}	6.0 ± 0.9^{a}
DIG	21.1 ± 1.15^{b}	35.29 ± 1.67	45.63 ± 0.38^{c}	3.4 ± 0.2^{c}
TILL	20.6 ± 2.01^{b}	35.85 ± 1.08	53.11 ± 0.97^{b}	3.9 ± 0.4^{b}

When present, different letters in a column indicate significant differences among treatments (Tukey's HSD at $p < 0.05$).

Table 7. Main fruit biometric characteristics at harvest from olive trees under different treatments (NT, DIG, and TILL) during the 2016/2017 growing season.

Treatment	Weight g	Fruit per Tree	Equatorial ∅ mm	Polar ∅ mm	Pulp Firmness kg cm^{-2}	Dynamometer g
NT	5.62 ± 0.19^{a}	4502 ± 581^{b}	19.42 ± 0.22^{a}	24.91 ± 0.36^{a}	305.2 ± 13.5	522.56 ± 25.7^{ab}
DIG	3.88 ± 0.14^{b}	5438 ± 335^{a}	17.11 ± 0.23^{b}	22.48 ± 0.31^{b}	312.1 ± 15.3	491.56 ± 25.0^{b}
TILL	5.36 ± 0.18^{a}	3843 ± 397^{c}	19.17 ± 0.27^{a}	24.43 ± 0.32^{a}	280.1 ± 11.4	599.67 ± 29.8^{a}

∅ = diameter. When present, different letters in a column indicate significant differences among treatments (Tukey's HSD at $p < 0.05$).

Table 8. Colorimetric indices of drupes at harvest from olive trees under different treatments (NT, DIG, and TILL) during the 2016/2017 growing season.

Treatment	Lightness	Chroma	Hue
NT	48.92 ± 2.33	19.77 ± 2.48	115.00 ± 3.10
DIG	46.70 ± 1.80	18.40 ± 2.01	120.32 ± 3.62
TILL	44.87 ± 2.03	15.92 ± 2.11	124.24 ± 3.13

When present, different letters in a column indicate significant differences among treatments (Tukey's HSD at $p < 0.05$).

4. Discussion

4.1. Soil Chemical Properties

The persistent and large increases in both soil organic C and total N pools observed in digestate-treated plots across the entire experimental period is consistent with what was reported, for instance, by Knoop, Dietrich, Heinrich, Dornack and Raab [31], who found a 21-month lasting effect after the soil addition of digestate. Needless to say, most authors' reports corroborate this finding [37–39] However, it must be also noted that opposite results were shown by other authors [20,40]. For instance, Adani, et al. [41] observed a 50% increase in labile carbon loss as a result of a 75% increase in the compost dose. Nevertheless, the potential storage of freshly added C pools through digestate amendment is strongly dependent on either the anaerobic biogas properties or the recipient soil characteristics [42–45]. We surmise that the pedogenetic features of the studied olive grove soil rich in fine-textured particles, poor in calcium carbonate, and with limited aeration produced soil fabric conditions suitable for physical protection and reduced mineralization of newly added organic pools, whatever their content of easily degradable organic substrates. Finally, total C and N readings from non-tilled soil confirm that the high clay content leads to the formation of stable microaggregates in which carbon is stabilized and sequestered in the long term, in accordance with what was reported by Six, et al. [46] and Denef, et al. [47].

It is also worth noting that soil EC, which is a highly variable soil chemical characteristic generally subjected to seasonal fluctuations due to continuously occurring soluble nutrient addition (mineralization and fertilization) and removal (leaching, plant uptake, and microbial immobilization) processes, remained significantly higher in DIG-treated plots. This finding suggests that soluble salt accumulation was the preferential process occurring in the olive grove soil as a consequence of a reduced internal drainage (K_{sat}), which can be estimated as low as 0.27 mm h^{-1} by considering the clay content. This physical feature protects groundwater from eutrophication and excessive N leaching, but rises the risk of secondary salinization under repeated and large applications of solid digestate. Finally, other than an immediate and ephemeral increase observed soon after the start of the trial, no significant variations in soil pH were appreciated over the entire experimental period in all treatments, thus confirming the strong buffering activity of the acid clay-rich olive grove soil.

4.2. Soil Exchangeable Micronutrients and Aluminum Content

Soil acidity is one of the main constraints to crop production because it exerts multiple critical influences on chemical and biological soil properties. In detail, it has a pronounced impact on the growth and activity of soil organisms, including plants, animals and microbes; it affects the mobility of many contaminants; and creates conditions of a nutrient imbalance and aluminum toxicity [48]. Regarding this last point, it is needed to point out that aluminum toxicity is rarely a problem when the soil pH measured in $CaCl_2$ is above 4.8 (pHH_2O 5.2), because little aluminum exists in the solution or in exchangeable pools above this pH level. This is in full accordance with our findings from the acid olive grove soil, where aluminum was equally found at an estimated concentration around 1 μM in all three treatments: a value widely lower than the threshold value of 50 μM indicated by [49] as the upper limit interfering with Al-sensitive plants, which represent almost one-third of currently cultivated olive germplasms. To sum up, in the present study, aluminum toxicity cannot be considered a factor influencing the olive tree physiology and productivity, either directly or indirectly, because it was found to be well below the critical threshold and not to have negatively altered the Ca/Al balance, which was always much larger than the critical value of one.

Note that in strongly acidic soils, as the one tested in the present research, the availability of macronutrients (Ca, Mg, K, P, N, and S), as well as two micronutrients, Mo and B, is strongly restricted. In contrast, the availability of micronutrient cations (Fe, Mn, and Cu, but also Zn and Ni) is increased, even to the extent of toxicity [48]. Unbalanced and low Ca, Mg, and K contents are certainly a major pedogenetic-linked factor limiting olive tree yield and quality. Similarly, the parent material and pedogenetic processes can be invoked as main factors responsible for deficiencies of exchangeable Cu and Fe levels, irrespective of differing soil management treatments. Needless to say, iron toxicity can become a serious problem to plants at very low pH levels (usually less than 4.0) and under anaerobic conditions, which did not occur in the olive grove soil here.

Manganese is an essential plant nutrient whose toxicity, when taken up in excessive amounts, can represent a growth-limiting factor to olive tree plants, especially in acid soils derived from manganese-rich parent material [50]. In fact, Mn becomes increasingly available as the pH drops, but unlike aluminum, Mn toxicity starts to appear at pH_{H2O} levels as high as 5.6. Given the chemical properties and nature of the parent material of the olive grove soil, not unexpectedly, the available Mn concentration in soil reached a value as high as 200 mg kg^{-1} (above the 50 mg kg^{-1} limit considered toxic to many crop species [50,51]), and it was further enlarged, rather than restricted, by the addition of the organic amendment, which increased its bioavailability (albeit not significantly), as observed in the present study. In fact, the Ca/Mn (moles to moles) ratio, calculated using the soil data, shows a clear declining trend across treatments, from 15.3 and 12.9 (in TILL and NT soil, respectively) to 10.2 (in DIG-treated soil), thus corroborating the role of the added organic matter in promoting Mn mobility in soil.

We argue that, given the feeding material used for the anaerobic digestion process, the low micronutrient content, and the used application rate, the solid anaerobic digestate supplied in the present research did not contribute to significantly rise the metal load of the recipient soil. In fact, in accordance with several authors [16,19,52], biogas digestates have generally very low amounts of heavy metals, which primarily depend on the quality of the input feedstock [18]. Thus, direct enrichment of soil micronutrients cannot be considered here a relevant process linked to the soil addition of an agricultural solid digestate. From the other side, it cannot be excluded that reduced mineral N forms entering the soil with digestate addition could have promoted the nitrification process with the consequent release of additional acid cations capable of displacing Ca and Mg from the exchange complex, thus enabling their leaching to occur and a further decrease soil pH [48]. If this is true, excess ammoniacal N sources entering the soil with a high clay content (45%) and low base saturation (<32%) would have increased the nutrient unbalance at the soil–water interface and further contributed to rise acidic conditions and soluble Mn excess. Finally, Mn forms complexes with organic ligands, but how these ligands are stable, and increases or decreases in Mn mobility in soil, and its availability to plants depend on several factors (soil pH, CSC, water and temperature regimes, and microbial activity), which make the question still matter of debate [12,53].

To conclude, whatever the role of the solid digestate addition on affecting Mn mobility in soil, it must be noted that the Ca/Mn values obtained for the field treatments were always below the critical threshold of 30, a finding representing a soil condition characterized by a constitutive excess of soluble Mn and Mn-related nutrient imbalance, which both severely limit plant growth and productivity.

4.3. Olive Tree Fruit Micronutrients and Aluminum Content

The metal content evaluation in drupes shows a similar Cu concentration in DIG and NT, suggesting a higher Cu uptake than TILL plants; this could be connected to higher CO_2 fixation and Fv′/Fm′ ratio, taking into account that Cu is a fundamental co-factor in plastocyanin function and, consequently, in photosynthetic activity [54]. Furthermore, it is interesting to note how NPQ is composed by several factors, indicating not only a heat dissipation process but also the efficiency of other metabolic pathways related to photosystems II and I, like the quenching state transition (*q*T) that indicates phosphorylated proteins transferring light energy from photosystem II to photosystem I [55]. So, we can argue that the higher NPQ found in DIG and NT could be interpreted as a positive response, although it was not statistically different between DIG and TILL.

Noteworthy, the total Mn content in olive tree fruits was found within the expected range (4.9–8.7 mg kg^{-1}), whereas a slightly lower amount of Fe was observed (reference range: 18–25 mg kg^{-1}) [56], suggesting that (i) olive plant tolerance to excess Mn involves an alteration of the distribution of the metal within the plant (i.e., favoring root accumulation while restricting shoot translocation, especially towards tree fruits) [50]; and (ii) iron and manganese contents are negatively correlated, and excess Mn brings about inhibition of Fe uptake and metabolism [51]. In fact, in addition to direct negative effects, excess Mn in the soil solution can interfere with the absorption, translocation, and metabolism of other essential nutrients, such as Ca, Mg, P, and Fe, thus magnifying their deficiencies. This becomes particularly relevant in acid soils where a nutrient imbalance is common.

From the other side, aluminum remained unchanged in olive tree fruits, thus mirroring the evenly distributed and tiny amount of soil exchangeable Al detected in all treatments.

4.4. Physiological Responses

Based on photosynthesis and stomatal conductance, it appears clear that soil tillage negatively affected the photosynthetic rate of olive leaves, thus providing values which are generally lower than those generally reported in the literature, as in [50]. Then, it is not surprising that the chlorophyll fluorescence parameters—which allow us to estimate the photochemistry of the photosynthetic system II (PSII) from the fluorescence decline, termed "quenching", and provide an early assessment of plant responses to most types of stresses [57]—declined. In particular, the reduction of the Fv/Fm ratio (i.e., the maximum quantum yield of PSII), which was well below the threshold value of 0.7, indicates that olive tree plants were under stress conditions [58]. To explain these findings, we argue that the mechanical disturbance due to the repeated tillage events could have altered the soil physical properties in a way that, given the fine-textured features of the olive grove soil and the rainfed cultivation regime, water loss through evaporation increased due to improved soil aeration. It is also worth considering that, within the context of global warming, soil water deficiency is being exacerbated by increasing temperatures and declining precipitation, as evidenced during the 2016–2017 experimental period (Table S1), when compared to the time series data registered over the 2000–2015 period (average T 17.4 °C, annual rainfall 889.7 mm, [26]. In brief, readings from physiological responses indicate that in conventionally tilled soil, factors other than Mn toxicity and a nutrient imbalance negatively affected the functionality of PSII in photosynthesis and hence olive tree productivity. We hypothesize that the tillage-induced water deficit, which was further exacerbated by the rainfed regime, would have resulted in a decline in the olive fruit number and oil content, but not in their biometric features. In a few words, water-stressed plants produced less olives of a similar size. This finding is in agreement with [59,60].

On the other hand, solid digestate addition, which brought about a marked and persistent increase in total soil C and N pools, seemed to alleviate the adverse physiological effects found in olive plants from tilled plots in terms of increased CO_2 fixation, stomatal conductance, and transpiration, even though they never reached response values as high as observed in NT-treated olive plants. Three possible interacting factors could be invoked to explain this result.

First is the reduced soil water loss due to an improved soil structure and the greater water-holding capacity of digestate-derived organic matter. Needless to say, solid anaerobic digestate exerts beneficial effects on soil physical properties, in full accordance with what was reported by several authors [19,53]. Moreover, it has been shown that digestate-released organic matter can contrast salt addition effects, thus enhancing soil water availability [61]. We also observed that under rainfed conditions and in a fine-textured soil, the no-tillage practice was even more effective in maintaining the soil water-holding capacity and alleviating any water stress, as shown by the photosynthesis-related parameters that were always higher in NT than in both tilled plots (either amended or not). Both these findings are consistent with Gimenez, et al.'s [62], who reported photosynthetic rates ranging between 7 and 18 μmol CO_2 m^{-2} s^{-1} when olive tree plants were grown under non-limiting water availability. On the other hand, the photosynthetic rate < 5 μmol CO_2 m^{-2} s^{-1} observed in conventionally tilled plots is certainly a clue of a stress response due to a water deficit. To sum up, adverse plant responses due to unfavorable physical conditions created in the clay soil by the tillage event were partially mitigated by the soil incorporation of solid digestate.

Second is a bio-stimulating effect on plant growth due to hormones and hormone-like molecules released from the solid digestate into the soil. Although poorly investigated, this topic was recently addressed by [63–65], who went on to show the presence of auxin-like properties located in a fraction of the dissolved organic matter extracted from the digestate from full-scale digester plants. Within this context, the potential for the application of digestate-derived bioactive compounds used as biofertilizer has been proposed by Du, et al. [66]. Needless to say, anaerobic digestion treatment has increased enormously during the last years and has become a very important sector for waste treatment and renewable energy production in Europe [67]. As a by-product of the anaerobic digestion process, digestate is of great potential interest not only as a soil amendment [15] but also as a source of biostimulant molecules [68], which are concentrated in the most biologically reactive DOM fraction and are directly involved in plant nutrition processes [69,70]. Further inside, due to the photosynthetic performances observed in DIG treatment that were higher or equal to TILL, a lower oil yield and mesocarp oil percentage cannot be explained with a reduced C fixation rate, but with a hormone-like activity due to digestate-released humic substances. That is also consistent with the acceleration of the drupe ripening process in plants from DIG-treated soil, which could in turn justify the lower force needed to detach them from branch, in accordance with [71,72]. Moreover, less decreased photosynthetic performances observed in DIG than in TILL justify the difference in plant productivity (in terms of fruits per tree and fruit yield) found between these two treatments. Then, it becomes clear that digestate addition exerted a mitigating action on plant physiology and productivity when compared to unamended tillage. To sum up, it cannot be excluded that a pool of bioactive organic molecules entering the soil after amendment with digestate directly and indirectly affected the olive tree's physiology, and hence its productivity, following a dose-response relationship (hormetic effect) [73].

Third are changes in soil microbial activity and compositional shifts in prokaryotic community composition, which in turn altered the soil nutrient release and the nutrient use efficiency, as well as rhizosphere interactions at the soil–plant interface. Needless to say, it is known that soil incorporation of anaerobic digestate affects the soil microbial activity and brings about changes in the phylogenetic composition of prokaryotic communities [16–18,20,74–77]. In two parallel papers focused on olive grove soil, it was found that major changes involved the phylogenetic shift in the r/K-strategist balance, being copiotrophic bacterial taxa favored (such as Proteobacteria) and oligotrophic taxa (i.e., Acidobacteria) constrained [25], accompanied by higher microbial C-use efficiency and a long-lasting release of soluble C and N forms [78]. This would lead to the conclusion that over the long term, digestate addition helped create soil ecological conditions that were less unfavorable (or less constraining) to olive tree growth than the conventional tillage did.

5. Conclusions

No-tillage represents a widely accepted and well-established practice for sustainable management of agroecosystems, including woody crops in the Mediterranean climate [79]. This becomes true also in the studied olive tree orchard where, despite adverse pedoclimatic features due to a high clay content, low pH, excess Mn mobility, and rainfed farming, no tillage has not provided a negative response in terms of photosynthetic rates and crop yield when compared to conventional tillage. In other words, even though adverse climatic and soil physico-chemical (low pH, excess Mn, nutrient imbalance, and clay content) conditions can negatively affect olive plant responses and productivity in the Mediterranean area [5,50], the present research evidenced that no tillage provided less unfavorable conditions for managing the rainfed olive orchard. This was likely due to a positive influence on soil water storage ability, which prevented olive plants from suffering a water deficit under increasing aridity and warm temperatures. Such a finding corroborates also the need to maintain ecological conditions conducive to oligotrophic prokaryotic taxa, as reported in Pathan, Roccotelli, Petrovičovà, Romeo, Badagliacca, Monti, and Gelsomino [25], and protect soil C pools [24] in order to prevent the overexploitation of soil resources, including plant-available soil moisture. On the contrary, soil tillage, which is usually performed to remove weeds and thus avoid water and nutrient competition, showed detrimental effects on olive tree yield. This finding corroborates [80]. Interestingly, soil incorporation of solid digestate mitigated tillage-induced adverse effects on plant physiology and productivity, thus contrasting the negative impact of the tillage event. However, productivity levels in digestate-treated soil never reached those of the non-tilled soil. Several direct and indirect effects can be invoked to explain this finding, which are related to the multifunctional and the long-lasting role played at the plant–soil interface and on soil microbiota by the partially decomposed digestate-derived organic matter entering the soil. To conclude, even though it cannot be considered and managed like a common mineral fertilizer nor as a commercial composted material, solid anaerobic digestate confirms its potential as a feasible and effective measure to properly dispose and valorize organic by-products from agro-forestry and agro-energy sectors without altering trace metal availability and their toxicity. Moreover, it represents also a win-win strategy to manage the soil fertility status under changing climate conditions in vulnerable, dry-prone farmland areas and in less productive soils.

Supplementary Materials: The following supporting information can be downloaded at https://www.mdpi.com/article/10.3390/soilsystems8010013/s1, Table S1: Thermo-pluviometric dataset related to the study site reporting air temperature (in °C) and rainfall (in mm) monitored across the 2016 and 2017 growing seasons.

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