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How water table level influences C balance under different fertilization regimes

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ARTICLE INFO

Handling Editor - J.E. Fernández

Keywords: Greenhouse gases Soil organic carbon Soil respiration Groundwater Manure

ABSTRACT

Carbon sequestration in soil has been extensively sought in the agroecosystems through practices which increase organic carbon inputs and/or decrease soil organic carbon (SOC) degradation processes. Less is known about the extent of shallow water table influences in mineral soils, despite being soil moisture a major driver in modifying the C cycle. To examine its effects, a 4-yr lysimetric experiment was set up to measure the C balance components under free drainage and shallow water table at 60 and 120 cm depth. Two levels of N input (250 and 368 kg N ha⁻¹ y⁻¹) were also studied, using dry manure in 2011 and 2012 and fresh manure in 2013 and 2014. Carbon balance was estimated through the difference between inputs (C from organic inputs and root residues and exudates) and outputs (heterotrophic respiration, methane, and C leaching). A negative C balance was measured under all treatments (-3487 kg C ha⁻¹), being respiration not compensated by the consistent C input of organic fertilizer. Furthermore, high N inputs increased SOC mineralization, decreasing the C balance. The role of soil was also observed by the SOC analyses, which confirmed the losses estimated through C balance. The study substantiated also the interacting effect between shallow water table and type of organic carbon, which was revealed crucial for C balance in mineral soils. To conclude, results suggested that water table level around 120-cm depth could limit SOC depletion.

1. Introduction

There is growing interest in sustainable soil management practices that promote soil carbon sequestration (Guo and Gifford, 2002; Kämpf et al., 2016; Minasny et al., 2017). Best practices in this area aim to achieve a positive balance in soil organic carbon (SOC) stock by increasing organic Cinputs and/or reducing SOC degradation processes. Soil carbon dynamics are influenced by both agronomic management and natural drivers, such as temperature, water availability, and soil texture. Among these drivers, soil moisture plays a particularly important role in regulating physical and biochemical processes (Henderson-Sellers, 1996). The water-filled pore space (WFPS) is a key factor that determines microbial respiration rates, with optimal values often found at 60 %, but highly variable depending on soil characteristics (Schjønning et al., 2003). Below this threshold, respiration is limited by

water stress (Aon et al., 2001), while high values limit respiration due to low oxygen levels (Chen et al., 2017; Mukumbuta et al., 2019). Methane (CH₄) production in soils also occurs under strictly anaerobic conditions (Khalil and Baggs, 2005), while methanotrophic microorganisms that require oxygen and CH₄ for their metabolism constrain CH₄ emissions under aerobic conditions (Dutaur and Verchot, 2007). As a result, CH₄ emissions are primarily associated with ponded conditions, such as rice paddies (Sanchis et al., 2012; Uprety et al., 2011) and poorly drained soils. However, even local anoxic conditions can lead to CH₄ emissions (Robertson et al., 2000), particularly when associated with fertilizer N applications that may inhibit CH₄ oxidation (Hu et al., 2013; Mosier et al., 1991). By creating heterogenous WFPS conditions along the soil profile, water table level is a key variable that affects the C cycle and greenhouse gas (GHG) emissions by influencing both C input and microbial processes. On the one hand, capillary rise from groundwater is a

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valuable contributor to crop production (Xu et al., 2013). On the other hand, excessively shallow water tables can impair crop growth (Mueller et al., 2005), which can in turn reduce the input of C derived from roots and residues. Water table fluctuations also play a role in regulating organic matter depletion by affecting redox oscillations (Rezanezhad et al., 2014), particularly in relation to the residence and return times of organic matter (Jeanneau et al., 2020). Increasing the depth of the water table can promote greater CH₄ oxidation by lengthening the diffusion pathway of gases to the surface and increasing their residence time in the soil profile, thereby boosting gas oxidation rates (Crézé and Madramootoo, 2019).

Shallow water table influences 22-32% of the world global land area, including major production regions (Fan et al., 2013). In particular, a considerable portion of the Veneto low plain (NE, Italy) is occupied by a water table (ARPAV, 2014) that fluctuates between -0.8and -1.0 m in the winter and -2.0 and -3.0 m in the summer. In this region, the Veneto Regional Government has subsidized in past Rural Development Programs several agri-environmental schemes aimed at promoting sustainable use of organic and mineral fertilizers. These measures were designed to increase SOC stocks and mitigate GHG emissions and N water pollution (Dal Ferro et al., 2016; Longo et al., 2021). Their effects, however, are strongly influenced by the interaction between C and N input and water table level. In a recent meta-analysis, Huang et al. (2020) noted that N addition can stimulate the storage of soil organic C through two mechanisms: by increasing C input and, at high N rates, by reducing the decomposition of old SOC. Conversely, when C-rich biomass inputs are associated with low N availability, microorganisms can produce a priming effect by decomposing soil organic matter to acquire N, which can lead to SOC depletion (Blagodatskaya and Kuzyakov, 2008).

Unfortunately, to the best of our knowledge, there is currently a lack of robust literature on the effects of fertilization practices on the SOC cycle and GHG emissions under shallow water table conditions in mineral soils. Most studies have focused on organic soils, and there is a need to fill this research gap. To address this, we conducted a four-year lysimeter experiment starting in 2011 to investigate how the interaction between shallow water table and fertilization practices affects the SOC balance and C-derived GHG emissions, including $\rm CO_2$ and $\rm CH_4$. Our hypothesis was that the high anaerobic conditions associated with high N input under shallow water table could limit SOC sequestration, leading to an increase in $\rm CO_2$ release from mineral soils.

2. Materials and methods

2.1. Experimental Site

The experiment was conducted over a 4-year period from 2011 to 2014 at the L. Toniolo Experimental Farm of the University of Padova in Northern Italy (45°19′N, 11°31 E). The site consists of 12 free draining lysimeters, each with a volume of 1.5 m³ (surface area of 1 m² and depth of 1.5 m). The lysimeters are buried and delimited by cement side walls, and the soil level within each is aligned with that of the surrounding landscape. The soil is a Fluvic-Calcaric Cambisol (35% sand, 48% silt, 17% clay), a widespread typology extending over almost 50% of the low Veneto plain at an estimated 420,922 ha. The typical pH is 8.1 and SOC content is 0.9%. Each lysimeter is filled while preserving the original soil horizons from an adjoining experimental farm. Each lysimeter (with dimension $100 \times 100 \times 150 \text{ H}$) is filled with 20 cm of draining layer (i.e., gravel 3-5 cm diameter) at the base, and 130 cm of excavated soil. The bottom is funnel-shaped and connected via an underground drainage pipe (1% slope) to an external cylinder of height (150 cm) filled with water. The water level inside the cylinder is maintained at the targeted value using an electronic water level sensor; this design allows fine tuning of the water table level, as well as collection of percolation water. A piezometer spans the center of each lysimeter and allows measurement of the water table level and sampling of groundwater.

Each lysimeter is buried at the center of a plot area of 200×200 cm. Lysimeter and surrounding plot area are managed in the same way except for the water table depth treatments.

The experimental site is also equipped with a sliding automatic rain shelter; this is used to prevent unintended inputs of rainwater, protect against hail and avoid shading.

2.2. Treatments and management techniques

A full factorial design was chosen combining two experimental factors: i) N input (250 N and 365 N); and ii) Water table depth (Free Drainage (i.e., no water table)(FD), deeper water table (WT) at $-120\ cm$ (WT120) and shallow Water table at $-60\ cm$ (WT60) from soil surface). Three levels of water table conditions and two levels of fertilization resulted in a total of six treatments each with two completely randomized replicates. The number of replicates was limited by the high cost of the equipped lysimeters, as already reported in Cocco et al. (2018).

Maize (Zea mays L.) was grown in succession throughout the 4-year study. The N input doses were defined following rules for Nitrate Vulnerable Zones (NVZs) according to the Nitrates Directive 91/676/ EEC (European Commission, 2018). The two levels at which N was applied were: 170 kg organic N ha⁻¹ + 80 kg mineral N ha⁻¹ ("250 N") and 250 kg organic N ha⁻¹ + 118 kg mineral N ha⁻¹ ("368 N"). Mineral N was applied as urea in three split doses (30% incorporated at sowing in spring, 35% top-dressed in mid-May, and 35% top-dressed in mid-June). Organic N was applied in a single dose before sowing. Two types of organic fertilizers were tested to evaluate the effects of agrienvironmental schemes aiming at replacing mineral N with organic N. It was a commercial mix of beef cattle manure and poultry litter (84% d. m., 5.2% OC, 2.8% N) in 2011-2012 and as fresh cattle slurry (8% d.m., 3.6% OC, 0.26% N) in 2013-2014. After the distribution of mineral and organic N fertilizer before sowing, the soil was tilled manually with a spring spade to a 25-cm depth to simulate harrowing. Maize was sown in two rows (with an interrow distance of 70 cm) at a density of 8 plants m ². Plants were harvested at commercial maturity stage removing all aboveground crop residues. Sowing in 2011 was delayed due to set up of the instrumentation. Consequently, a short cycle corn variety was planted (FAO class 300). In 2012–2014, a medium-late cycle variety was planted (FAO class 600). Every crop cycle received two pesticide treatments: Clorpirifos at seeding and Deltamethrin at pollen shed. In 2014, treatment with Lambda-cyhalothrin was also included. Total annual water input was 900 mm and was provided by a series of irrigation events during the spring-summer growing season (according to average crop need in FD treatments) and by rainfall simulations during the postharvest period (randomly distributed over the typically rainy months). Water table levels were manually adjusted daily during the summer and every three days in other seasons. The maximum allowed fluctuation was \pm 10 cm from the set reference level (Cocco et al., 2018).

2.3. Soil moisture and temperature monitoring

A time domain reflectometry (TDR) sensor (Moisture Point MP-917, ESI Environmental Sensors Inc., Canada) connected to vertical wave guides monitored water content across three soil profiles: 0–15, 15–30, and 60–90 cm. Starting in 2013, every lysimeter was equipped with an automated monitoring system that allowed the continuous measurement of soil water content. The system was composed of CS635 TDR probes (Campbell Scientific, USA) installed at 15-, 30-, and 60-cm depths connected to a CR-10X datalogger (Campbell Scientific) through a series of multiplexers. Total porosity was derived from soil bulk density and particle density, with the latter measured by a helium pycnometer (Micro Ultrapyc 1200e, Quantachrome, Austria). Waterfilled pore space (WFPS, %) was calculated as the ratio between water content and total porosity. A thermocouple system monitored soil temperature at a depth of 5–30 cm (Model 107 temperature probe, Campbell Scientific).

2.4. C balance component measurements

2.4.1. Root and manure C input

At harvest, maize grain and crop residues were weighed, shredded and dried at 65 °C. C input from above-ground crop residues and roots, including rhizodeposition, was estimated on the basis of total above-ground biomass at harvest, following the procedure of Kätterer et al. (2011). A C concentration of 0.45 g C g $^{-1}$ d.m. in all plant parts was assumed. To limit lysimeter disturbance, root biomass was measured only at the end of experiment using the monolith method (25 cm \times 25 cm x 30 cm) (Böhm, 1979). The vertical root distribution was then estimated using such measurements of root derived biomass in a Michaelis Menten-type function (Kätterer et al., 2011). Root C input was then adjusted to account for exudates, considered as 9% of the total assimilated C (Derrien et al., 2004).

For organic fertilizers, the C input was estimated through elemental analysis method (VARIO MACRO, Elementar Analysensysteme GmbH, Germany), leading to a total input in cumulative periods equal to 7710 and $11346~{\rm kg}~{\rm C}~{\rm ha}^{-1}$ under 250 N and 368 N, respectively.

2.4.2. CO₂ Soil respiration and CH₄ emission

To monitor emissions of carbon dioxide (CO₂) and methane (CH₄) from the soil, an automated closed dynamic chamber system was installed (Delle Vedove et al., 2013, 2007); twelve chambers were installed (i.e., one per lysimeter) in the center row of each lysimeter between plants and oriented to south in order to prevent any shadowing effect on the soil surface's temperature inside the chamber. The closed chamber is a top-closed and base-open steel cylinder (20 cm inner diameter and 10 cm high) placed over a collar inserted in the soil at a depth of around 7 cm. A motorized steel lid, with a neoprene sheet on the inner surface, and a rubber O-ring on the cylinder's perimeter, warrant a tight seal of the chamber volume. A vent installed on the lid, maintains equilibrium with external pressure variations and reduces temperature and humidity differences between chamber headspace air and the surrounding atmosphere during chamber deployment (Hutchinson and Livingston, 2001; Xu et al., 2006). For more detailed methodological features refer to Delle Vedove et al. (2013).

Analysis of CO_2 gas emissions was performed by means of an infrared gas analyzer (IRGA). A datalogger (CR1000, Campbell Sci. Inc., USA) controlled all monitoring operations (chamber closure, activation and heating of the IRGA analyzer, air circulation pumps, and aperture/closure of chamber valves). Air circulation within the system was regulated by two pumps and 26 solenoid valves. Each of the 12 chambers took six measurements a day (every 4 h, i.e., 0:00, 4:00, 8:00, 12:00, 16:00, 20:00) for a deployment time of 180 s and took 120 measurements of CO_2 concentration during that time. This resulted in 8640 gas concentration readings daily. Measurements of CO_2 concentration were taken every 1.5 s, beginning 10 s before chamber closure (150 s). For each measurement, the system recorded the fraction of water vapor (W, mmol mol-1), air temperature (T, $^{\circ}C$) and pressure (P, kPa). The dry air CO_2 molar fraction during chamber deployment time C_t was fitted by the following non-linear equation:

$$C_{(t)} = C_x - (C_x - C_0)e^{-a(t-t_0)}$$
 (1)

Where $C_{(t)}$ is CO_2 concentration at time t (µmol $CO_2 \cdot mol^{-1}$ dry air), corrected for the molar fraction of vapor, pressure and temperature. C_0 is the initial concentration of CO_2 at chamber closure (the intercept of a linear interpolation of the last 10 points of the mixing). C_x and a are regression parameters that define the horizontal asymptote and the form of the function. Finally, t_0 is the time at which C_x equals C_0 .

The rate of CO_2 gas exchange over time (dC/dt) was obtained by deriving the previous equation for time:

$$\frac{dC}{dt} = a(C_x - C_0)e^{-a(t - t_0)} \tag{2}$$

The flux of CO₂ (i.e., soil respiration) was expressed as follows:

$$SR = \left(\frac{V}{S}\right) \left(\frac{dC}{dt}\right) \frac{P_0}{R(T_0 + 273.15)} \tag{3}$$

Where SR is soil respiration (μ mol CO₂ m⁻² s⁻¹), P₀ is pressure at t₀ (kPa), T₀ is temperature at t₀ (Celsius), R is the universal gas constant (8.31 J mol⁻¹ K⁻¹), V is the volume of the system (cm³, chamber + tubing) and S is chamber area (cm²).

Analysis of CH₄ gas emissions was performed by means of a gas auto-sampler connected to the 12-chamber automated system by high-density PE tubing (10 m L, 4/6 mm in inner/outer diameter). After chamber closure, air circulated between the chamber and vials for 90 s. For each deployment time after chamber closure, three 20-mL samples (taken at 0, 15 and 30 min after closure) were withdrawn from the chamber headspace air and injected into pre-evacuated 20-mL crimped glass vials, each with a gas-tight butyl rubber septum. Gases contained in the vials were analyzed using a gas chromatograph (7890 A model G3440A, Agilent, USA), equipped with a flame ionizer detector (FID) to determine CH₄ concentration. The three concentration points were linearly interpolated, and the slope is the dCH₄·dt⁻¹ molar change rate. The CH₄ flux rate (µmol CH₄ m⁻² s⁻¹) was then computed using Eq. 3.

For CH_4 analysis, a daily gas sampling was performed two days prior to fertilizer application and during the following eight days. Thereafter, the frequency decreased to twice per month until harvest, except for the periods of topdressing N fertilization, when frequency was again intensified. During the winter-spring period, sampling was performed once per month, except when the soil was frozen (i.e., January). On average, each growing season provided 23 sampling events.

2.4.3. Autotrophic versus Heterotrophic soil respiration

The SR is the sum of both autotrophic respiration (roots derived soil ${\rm CO}_2$ efflux) and heterotrophic respiration (SOC derived ${\rm CO}_2$ efflux). The soil C balance was calculated considering only heterotrophic respiration. To separate and quantify the two types of respiration, an approach based on the Arrhenius law was used. This model expresses the relationships between kinetic reaction and temperature and have been shown to accurately represent soil respiration (Guo et al., 2019; Xu and Baldocchi, 2004). In our study, soil moisture was also considered. Thus, heterotrophic respiration (*Rh*) was estimated according to:

$$Rh_{(T,\theta)} = (f + b\theta)e^{k(T - T_x)} \tag{4}$$

Where Rh is heterotrophic respiration as a function of soil temperature (T) and moisture (θ), f and b are calibration parameters, θ is the volumetric soil moisture in the 0–30 cm layer, k the reaction rate constant, and T_x the soil threshold temperature above which microbial reactions are active. Volumetric soil water content in each lysimeter was measured using TDR.

The Eq. (4) was calibrated using an independent data set obtained with an EGM-4 environmental gas analyzer (PP Systems, USA) during the 2014 growing season. The EGM-4 is a portable instrument with a chamber for static and dynamic measurements of CO2 fluxes, an air pumping system connected to an infrared analyzer, and external sensors for the measurement of agri-environmental parameters (in this case, soil temperature). EGM-4 measurements were compared with those of the automatic 12-chamber system, with favorable results ($R^2 = 0.75$). Following this, two lysimeters (not instrumented with automatic chambers) were used to precisely determine CO2 respiration fluxes. Both lysimeters were cultivated with maize (as previously described) and assigned the treatments 368 N FD and 368 N WT60, respectively. Autotrophic respiration of maize was calculated as the difference between total respiration (in the presence of roots) and respiration measured in undisturbed PVC soil cores (15 cm ID) previously inserted to a depth of 90 cm. Model parameters were calibrated using Excel Solver with the objective of minimizing the average square deviations between observed and calculated respiration.

2.4.4. C leached

Water samples were collected from percolation water, stored at -20 °C, and later analyzed for dissolved organic C (DOC) through the Walkley-Black method (Walkley and Black, 1934). Organic carbon lost through leaching and measured in percolation water (C_{LCH}) was calculated using the sum of leached organic C for each i-th percolation event:

$$C_{LCH} = \sum_{i=1}^{n} P_i DOC_i \tag{5}$$

where P_i is the percolation (L) for each individual i-th event and DOC_i is the respective concentration of C (mg C L⁻¹).

2.5. Net C balance

In this study, the system boundaries include the main components of the C balance of the mesocosm studied. The soil C balance was calculated as the difference between inputs and outputs:

$$\Delta C = C_{manure} + C_{roots} - C_{Rh} - C_{CH_4} - C_{LCH}$$
(6)

Where ΔC is the change of the organic C soil stock in the considered range (kg ha⁻¹) and C_{manure} is the C input from the manure (kg ha⁻¹), C_{roots} is the C input from roots and exudates (kg ha⁻¹), C_{Rh} and C_{CH_4} are the C loss through heterotrophic respiration and methane (kg ha⁻¹), and C_{LCH} is the C loss through leaching (kg ha⁻¹).

In addition, ΔC was compared to the SOC stock change measured by soil analysis. Lysimeter soil profiles were sampled at the beginning (May 2011) and at the end of the experiment (December 2014). Soil samples were taken at the following depths: 0–5, 5–30, 30–55, 55–75, 75–95, 95–120 cm. The samples were air-dried, sieved to 500 μ m and analyzed for SOC using the elemental analysis method (VARIO MACRO, Elementar Analysensysteme GmbH, Germany). Bulk density was measured with the core method.

2.6. Statistical analyses

Both daily and yearly cumulated daily values of SR were tested for each treatment (organic C input and mineral N fertilizer dose). All data were investigated for normality using the Shapiro-Wilk or Anderson-Darling (sample number >5000) normality test. Furthermore, daily SR was split into three periods: bare ground soil (from harvest to following organic C fertilization), 20 days interval between organic C addition and crop emergence, and the crop growing season period (from 20 days after C addition to harvest). Daily SR were tested with a linear mixed model with a repeated statement to account for repeated measurements considering WFPS and temperature as continuous factors and N dose as categorical factors. Post-hoc pair-wise comparisons of least-squares means were performed using the Tukey test to adjust for multiple comparisons. The non-parametric Kruskal-Wallis test was used for daily

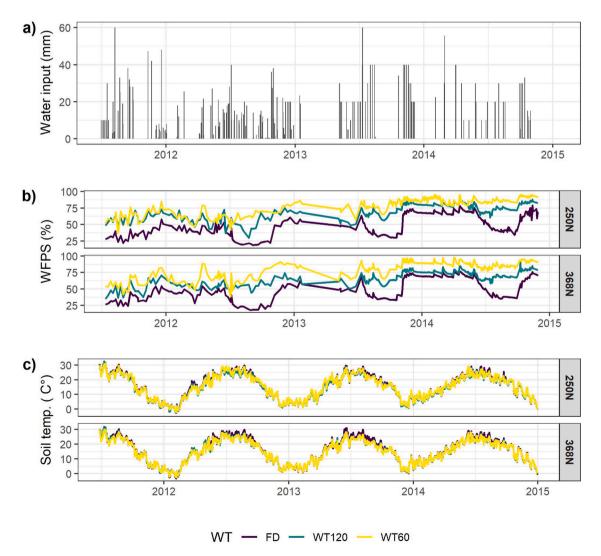


Fig. 1. a) Surface water input, b) Water-filled pore space (WFPS), and c) soil temperature in lysimeters under free drainage (FD), and with water tables set at 120-and 60-cm depths (WT120 and WT60).

values of C leaching and methane emissions, being their distribution not normal. Cumulative values for all balance components (roots, SR, Rh, $\mathrm{CH_4}$, C_{LCH}) were tested through ANOVA and post-hoc pair-wise comparisons of least-squares means were performed. Statistical analysis were performed using R (R Core Team, 2017).

3. Results

3.1. Water-filled pore space and temperature

In 2011-2014, water applied through irrigation and rainfall equaled 791, 959, 937 and 837 mm, respectively (Fig. 1a). The WFPS at the topsoil (0-15 cm) appeared to be intensively affected by evapotranspiration, water up-flux and infiltration. Similar dynamics were also observed in the intermediate soil layer (15-30 cm), which was influenced by surface water supply. Considering all depths, WFPS showed to be affected by WT (p < 0.001, WT60 > WT120 > FD) but not by fertilization. Different trends were identified in the WFPS dynamics. During growing season, WFPS in the upper 15 cm averaged 25%, 34%, and 43% for FD, WT120, and WT60, respectively, and 32%, 47%, and 67% at the 15-30 layer. In the deeper (60-90 cm) layer, WFPS fluctuated depending on deep water input, and produced averages of about 51% (FD), 92% (WT120), and 99% (WT60) (Fig. 1b). During base fertilizer application, WFPS of the top 0-30 cm layer measured about 32% in FD, 43% in WT120, and 52% in WT60; at top N dressing, 36 (FD), 46 (WT120), and 61% (WT160). Finally, during the winter period (November to February), values showed marked increases and reached 75% (FD), 83% (WT120), and 100% (WT60) in the deepest (60–90 cm) layer (Fig. 1b).

Soil temperatures in the 0–30 cm layer averaged 23 °C during the growing season (April-September) and fell below 10 °C during winter, with a few days in January and February 2012 below 0 °C (Fig. 1c). In summer, W60 and WT120 soil temperatures decreased by about 0.9 °C with respect to FD. Considering winter months alone, soil temperatures were similar across all treatments and averaged 9.7 °C.

3.2. Carbon in root biomass

The ratio between root (including exudates) and aboveground biomass (R/S) averaged 0.22, being minimum under 368 N-WT (0.16) and maximum under 250 N-FD (0.31) (Table 1). Root C input was influenced by the water table and fertilization treatments in both periods (dry manure in 2011–2012 and fresh manure in 2013–2014), with a significant interaction in 2011–2012 (Table S1). Despite a short cycle maize hybrid being planted in 2011, root C input was greater using dry manure than fresh manure, ranging from 3971 in FD to 6718 kg C ha⁻¹ in WT120 with a more pronounced difference among 368 N lysimeters (Fig. 2). On the other hand, using fresh manure, the combination of free drainage and lower N fertilization (250 N) stimulated root growth, 6231 kg C ha⁻¹ vs 5516 and 4703 kg C ha⁻¹ in WT120 and WT60, respectively (Fig. 2).

3.3. Total and heterotrophic soil respiration

Total soil respiration (SR) showed a pronounced seasonal cyclicity for all treatments, with lower emissions during the colder months (bare soil) and higher values in summer (cropping season).

During cropping season, which lasted on average 181 days, emission peaks originated from the implementation of management practices

 $\label{thm:continuous} \textbf{Table 1} \\ \textbf{Root-to-shoot (R/S) ratio estimated according to the Michaelis Menten-type function.} \\$

Fertilization level	FD	WT120	WT60	
250 N	0.31	0.25	0.25	
368 N	0.20	0.16	0.16	

(Fig. 3). An early spring peak of about 20 days followed the addition of manure/slurry and its incorporation. It was exceptionally high in 2012, when emissions reached values up to 224 kg C-CO₂ ha ¹ d⁻¹ (Fig. 3). A second lower peak was observed 50-60 days after sowing, dominated by the combined effect of temperature and plant development. Peaks ranged from 75 to 125 kg C-CO₂ ha ¹ d⁻¹ and decreased down to 1.3 kg C-CO₂ ha ¹ d⁻¹ before harvesting. For the whole cropping season, high fertilization treatment (368 N) showed slightly greater emissions than 250 N, especially during 2011-2012, when SR averaged 75.8 and 58.2 kg C-CO₂ ha ¹ d⁻¹ in the 20 days after fertilization, respectively (pvalue=0.06, Table S2). Emission peaks also occurred during the postharvest period, observed just one-two days after the harvest operations and lasting for about 20 days. Emissions were particularly high after harvesting in 2012, peaking up to 150 kg C-CO2 ha⁻¹ d⁻¹. In 2013-2014, in spite of lower values compared to the first period, the greater emissions recorded under 368 N after harvesting led to higher averages than 250 N (12.0 vs. 9.3 kg C-CO2 ha⁻¹ d⁻¹, p-value=0.02). Small oscillations still occurred due to changes in soil temperature and water content (Fig. 3).

Daily emissions were always affected by WFPS (Table S2). The effect was positive in the growing season using both dry and fresh manure (Fig. 4). On the other hand, the WFPS effect on the other periods differed based on the fertilizer type. Just after manure application, it was negative using dry manure and positive using fresh manure and vice versa during the bare period, even if at lower extent. Emissions also increased with increasing temperature (p-value <0.001, Table S1).

Average cumulative SR emissions equaled 17175 kg C-CO $_2$ ha⁻¹ in 2011–2012, significantly higher under shallow water table conditions (19128 kg C-CO $_2$ ha⁻¹ in WT vs 13269 kg C-CO $_2$ ha⁻¹ in FD, p-value< 0.05), while no differences were observed between fertilization levels. In 2013–2014, the behavior was reversed, being total emissions (mean of 17893 kg C-CO $_2$ ha⁻¹) mainly driven by the fertilization factor (p < 0.05), 368 N (19321 kg C-CO $_2$ ha⁻¹) > 250 N (kg C-CO $_2$ ha⁻¹) (Fig. 2) rather than water table level. Although WT was not significant, it ranked as follows for both N levels, WT120 > W60 > FD.

Considering heterotrophic respiration (Rh), cumulative values averaged 11590 kg C-CO $_2$ ha $^{-1}$ in 2011–2012 and 11806 kg C-CO $_2$ ha $^{-1}$ in 2013–2014 (Fig. 2). Application of dry manure in FD resulted in significantly lower Rh than WT60 (250 N-FD= 9944 kg C ha $^{-1}$ and 250 N-WT60 =12366 kg C ha $^{-1}$), while the opposite was observed using fresh manure (250 N-FD= 12165 and 250 N-WT60 =10820 kg C ha $^{-1}$) (Fig. 2). Furthermore, during the second time span also N level affected Rh (p-value=0.03), being greater under 368 N than 250 N (12316 vs 11296 kg C ha $^{-1}$).

3.4. Carbon as Methane

Daily methane fluxes were negligible (from -0.12 to $0.97\ kg$ C-CH₄ d $^{-1}$ ha $^{-1}$) under all treatments and did not vary according to fertilization nor water table level in the 2011–2012 period (Table S3). Fluxes were slightly greater in the 2013–2014 period compared to 2011–2012 (Fig. 5) and varied according to WT. Indeed, despite daily values being mainly negative in the initial period, indicating the soil served as a sink of CH₄ rather than a source, sudden peaks recorded in 2014 after manure distribution led to positive cumulative fluxes in the last two years, which averaged $0.69\ kg$ C-CH₄ ha 1 (Fig. 2). No statistical differences were observed for cumulative values (Table S1).

3.5. Carbon Leaching

Dissolved C loss through percolation water depends on C concentration and amount of percolation water. In the entire period, percolation water was collected and analyzed at 41 events (304 samples in total), averaging 26 mm per percolation event. Total percolation was similar among the WT treatments, with higher values in 250 N-WT120 (1074 mm) and 368 N-WT60 (963 mm). To note, in 2011–2012 no

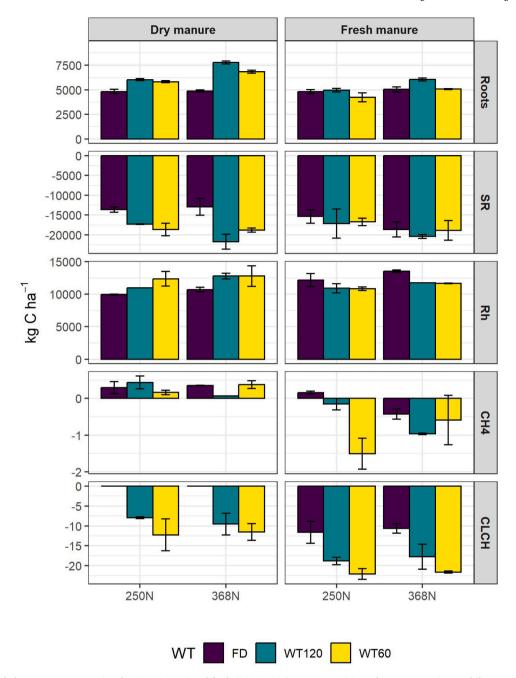


Fig. 2. Cumulative C balance components using dry (2011–2012) and fresh (2013–2014) manure. Positive values represent input while negative represent C losses. SR= total soil respiration, Rh=Heterotrophic respiration, CLCH= Carbon leaching.

percolation was observed in FD treatments, while it was 198 mm (250 N) and 213 mm (368 N) in the 2013-2014.

In 2011–2012, median C concentration in groundwater was 1.8 mg C $\rm L^{-1}$. No differences emerged among fertilization treatments nor WT (Table S3), despite concentration followed the trend WT60 >WT120 (median values were 1.9 and 1.8 mg C $\rm L^{-1}$) (Fig. 6a). Daily amount of C leaching during percolation events varied between WT60 (0.4 kg C ha $^{-1}$ d $^{-1}$) and WT120 (0.2 kg C ha $^{-1}$ d $^{-1}$) (Fig. 6b), which corresponded to cumulative values equal to 11.9 and 8.7 kg C ha $^{-1}$ for WT60 and WT120, respectively.

In 2013–2014, the lower volumes of percolation water under FD (12 mm d^{-1} vs 30 mm d^{-1} as average in WT60 and WT120) resulted in higher dissolved C concentration (median=3.0, 1.7 and 1.5 mg C L⁻¹ for FD, WT60, and WT120, respectively) (Fig. 6a). In 2013–2014, daily loss medians ranged from 0.3 (FD-250 N) to 0.4 kg C ha⁻¹ d^{-1} (WT60–250 N)

(Fig. 6b); peaks > 1.5 kg C ha⁻¹ d⁻¹ were observed during the post-harvest period. Cumulative C_{LCH} losses were lower under FD (5.7 kg C ha⁻¹) than WT120 (9.4 kg C ha⁻¹) and WT60 (11.3 kg C ha⁻¹) (p-value<0.001)(Fig. 2).

3.6. Soil Organic Carbon

SOC concentration varied between 0.47 and 1.09 g 100 g $^{-1}$, according to the profile depths and sampling events (p-value<0.001 for both, Table S4). Across all treatments, highest SOC concentrations were measured in the top 30 cm (0.840 g 100 g $^{-1}$), likely due to incorporation of organic fertilizer, and deeper layers (95–120 cm, 0.78 g 100 g $^{-1}$). SOC decreased in the four years, from 0.79 g 100 g $^{-1}$ in 2011–0.72 g 100 g $^{-1}$ in 2014 or expressed in terms of C stocks (from 137.2 to 124.6 Mg C ha $^{-1}$ considering 0–120 cm depth). Change in soil carbon (Δ C) stock was

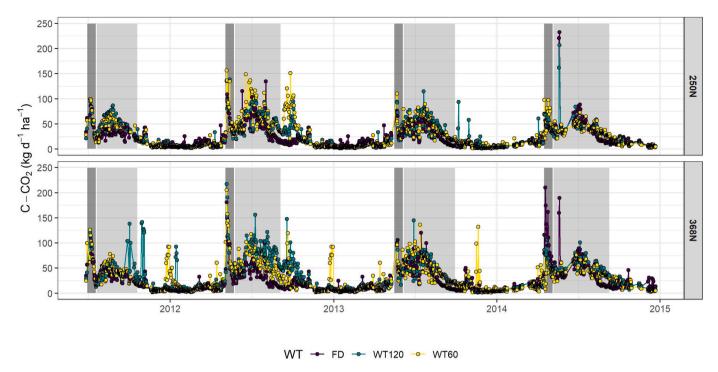


Fig. 3. Daily SR in lysimeters under free drainage (FD), and with water tables set at 120- and 60-cm depths (WT120 and WT60). Dark grey areas represent early crop seasons, light grey areas represent late growing season.

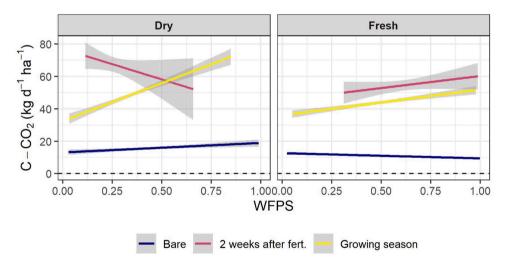


Fig. 4. Water filled pore space (WFPS) effect on daily total soil respiration.

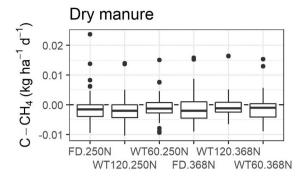
consistently negative for all depths, indicating conditions of depletion in the whole profile (Table 2). Fertilization level influenced ΔC , being depletion greater for 368 N treatments. (e.g., -16.9 vs -5.0 Mg ha⁻¹ at W120 cm for 368 N and 250 N, respectively). Conversely, WT levels did not statistically affect ΔC , despite WT120 and WT60 showed an apparent smaller depletion than FD under 250 N.

3.7. Carbon Balance

The algebraic sum of all C balance components provides an estimate of the change in soil C (ΔC) for the period considered. Positive values suggest C immobilization, while negative values indicate loss of C from the lysimeter profile.

Using dry manure, C contributions from fertilizations overly exceeded root C in the 368 N treatments (9286 vs. 4830 kg C ha $^{-1}$, on average) (Table 3) while inputs were aligned under 250 N as a result of the lower

fertilization (6314 kg C ha⁻¹) and greater root input (6472 kg C ha⁻¹). Manure C in 2013–2014 was lower, equaling 1396 kg C ha⁻¹ (250 N) and 2060 kg C ha⁻¹ (368 N) in the two years. Considering the C output, Rh was the most significant item while both C leaching and methane emissions were negligible. In the first time span all treatments resulted in a considerable increase of C, averaging 1857 kg C ha⁻¹, while the lower C inputs from fertilizers led to the opposite in the second span, whose losses averaged 5333 kg C ha-1 (Table 3). In 2011-2012, C accumulation appeared to increase accordingly with water table depth, ranking FD>WT120 >WT60 for both N levels, with greater accumulation under 368 N, where the greater fertilizer inputs balanced the lower C root. Furthermore, using fresh manure, the large Rh of FD led to a higher C depletion at 368 N (7372 kg C ha⁻¹) while comparable values were observed at 250 N (4563 kg C ha⁻¹). Considering the whole 4-vr period, the average C loss was 3487 kg C ha⁻¹ and lower values were observed under WT120 for both fertilization levels. Soil sampling



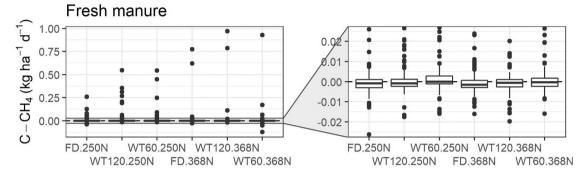


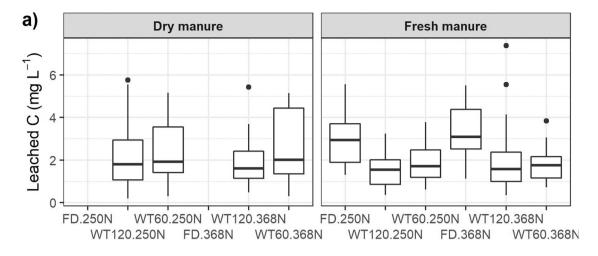
Fig. 5. Daily CH₄ fluxes in lysimeters under free drainage (FD), and with water tables set at 120- and 60-cm depths (WT120 and WT60).

analysis in the 0–60 cm layer also found high C losses showing a general agreement ($R^2=0.46$) with the balance method (Table 3). Both methods indicated higher depletion in 368 N than 250 N over the 4-yr period. However, some discrepancies were observed between the two methods, as the balance method overestimated C depletion at 250 N-WT120 and underestimated it at 368 N WT120 and WT60 with respect to soil sampling.

4. Discussion

Components of C balance in the soil system are driven by various chemical, biological, and environmental factors such as CO2 fertilization, land use change, and N addition (Tian et al., 2011). In our study a crucial role was played by the water table depth, which differently affected the final ΔC as a result of complex interactions between fertilization, WFPS and crop growth. Total soil respiration (Sr, autotrophic + heterotrophic) augmented under WT60 and WT120, which could be attributed to the response of the rewetting conditions (Barnard et al., 2020) caused by the upward fluxes, ranging from 269 to 670 mm h⁻¹ (Longo et al., 2021b). Shallow water table creates indeed heterogenous conditions in the soil environment with three distinct zones, saturated, semi-saturated (i.e., capillary fringe due to capillary rise) and non-saturated (i.e., vadose zone) (Baird and Low, 2022). In particular, capillary rise is associated with redox oscillations and acts as hot-spots and hot-moments, being of great importance to chemical cycling (Zhang and Furman, 2021). Rezanezhad et al. (2014) demonstrated that water table fluctuations directly increased CO2 emissions because of the consociated changes in O2 availability and water saturation. Recently, also Jeanneau et al. (2020) and Pronk et al. (2020) found that water table dynamics positively affected CO₂ fluxes and organic C oscillations. The former study explained the effect as due to the destabilization of organic matter caused by WFPS fluctuations, suggesting that limiting large variations in WFPS, and in turn wet-dry cycles, by a throughout management of the water table level, could help reducing CO2 emissions. Other studies have also shown that management practices such as controlled drainage may have significant effects on soil respiration. Similar to our findings, Nangia et al. (2013) observed greater CO₂ fluxes under controlled tile drainage compared to freely drained fields. Despite Crézé and Madramootoo (2019) did not find any significant correlation to seasonal changes in soil WFPS, CO₂ fluxes under shallow WT (75 cm) were 21% greater than under FD after mineral fertilization. Similarly, Kruse et al. (2004) found a significant moisture effect on C mineralization, and previous studies by Doran et al. (1990) and Linn and Doran (1984) have reported a close relationship between respiration and water content or WFPS. In our study, we observed a 20-days CO2 bursts after manure incorporation (both using dry and fresh manure) which can be related to the combined effect of the input of C (Lai et al., 2017) and the tillage operation, which possibly increased aeration and exposed previously protected organic matter by aggregates (Six et al., 2004). Interestingly, we observed a contrasting effect of soil WFPS on CO2 fluxes, depending on the type of fertilizer used. Specifically, the WFPS effect was negative for dry manure and positive for fresh manure. We speculate that this contrasting effect may be due to differences in the amount of recalcitrant C in the two types of manure. This is substantiated by the study of Piccoli et al. (2022), which also observed a contrasting interaction between the type of organic fertilizer and the WT conditions. The use of fresh beef manure in shallow WT conditions with WFPS > 80% was associated to higher CO2 fluxes in the 2 weeks following application. On the contrary, the distribution of solid digestate originated higher CO2 emission in free drainage conditions with WFPS levels averaging at 55%. Those differences were explained by the greater content of labile C forms in fresh manure than solid digestate, that promoted C degradation under anoxic conditions. Lai et al. (2017) confirmed the effect of the fertilizer source on soil respiration rate, which was related to the proportion of mineral to total N. The authors cited studies (Abalos et al., 2013; Kaur et al., 2008) indicating that farmyard manure and slurry applications impact soil organic matter pools and result in a high concentration of soil dissolved organic C, which are strongly associated with soil microbial and enzymatic activity (Kiikkilä et al., 2014).

Effect of fertilizer was also observed on denitrification dynamics, concurrently monitored at the same time of this experiment, with 368 N having greater cumulative N_2O losses when applying fresh manure but not when applying dry manure (Cocco et al., 2018). There is no clear



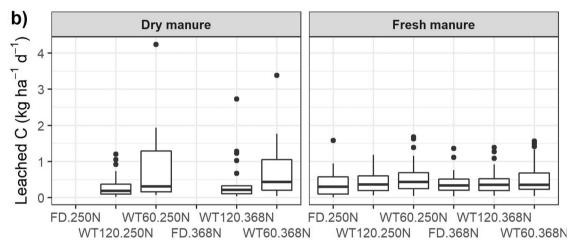


Fig. 6. C concentration in percolation water (a) and daily C losses during leaching periods (b) in lysimeters under free drainage (FD), and with water tables set at 120- and 60-cm depths (WT120 and WT60).

Table 2 SOC stocks in the 0– $120\,\mathrm{cm}$ profiles at the beginning and the end of the experiment.

		SOC Mg C ha ⁻¹		ΔC Mg C ha ⁻¹	
Fertilization	WT	2011	2014		
250 N	FD	121.7	106.5	-15.2	
	WT120	141.9	136.9	-5.0	
	WT60	141.0	134.4	-6.6	
368 N	FD	140.7	126.6	-14.1	
	WT120	143.8	126.9	-16.9	
	WT60	134.5	116.2	-18.3	

evidence about the role of N input on soil respiration. Previous studies have suggested that emissions may decrease as a result of fertilization (Foereid et al., 2004; Giardina et al., 2004) due to the potential for N addition to suppress the decomposition of native SOC (Al-Kaisi et al., 2008).

Plant growth may alter C dynamics being the percentage transfer of recently assimilated C from shoot into the root-soil system based on the developmental stages and greater in the young plants (Hoffmann et al., 2018; Remus and Augustin, 2016). We hypothesize that fertilization may have stimulated plant growth, leading to an increase in root-translocated C, which could have been used as exudates for microbial respiration or root respiration, as suggested by Kou et al. (2007). In support of this, Yan et al. (2021) found that root respiration

contributed more to the total increase in CO_2 after N application than microbial respiration, being related to the increase of leaf area index. This is also in accordance with Amos and Walters (2006) who identified the primary maize root growth between 20 and 70 days after emergence.

The substantial increase in soil respiration observed under 368 N fertilization significantly contributed to the final C balance, with the highest C losses observed under high N input. Daily N_2O emissions measured at this site shown to be promoted by high N doses and high WFPS, which could also have fostered CO_2 emission (Cocco et al., 2018). These findings are also consistent with previous studies by Poffenbarger et al. (2017) and Singh et al. (2018), which demonstrated that excessive N fertilization leads to depletion of SOC. Ding et al. (2010) also supported these results, showing that N fertilization can increase the decomposition of native SOC by up to 6.5%. Even high-C input organic fertilization may result in a significantly negative soil C balance in Mediterranean regions, as shown by Lai et al. (2017).

C leaching and CH₄ emissions were two minor items of C balance. Jeanneau et al. (2020) reported that water table dynamics drove C leaching (C_{LCH}), which some authors consider a crucial component of the C balance (Kindler et al., 2011; Nakhavali et al., 2021). However, in our experiment, C_{LCH} was negligible due to low C concentration in groundwater, which was 50 times lower than that reported by Kindler et al. (2011). Similarly, C loss as CH₄ was a minor component of the final ΔC , with soil acting as a sink rather than a source. Median daily CH₄ values were consistent with those reported by Franco-Luesma et al. (2020) and simulated by Longo et al. (2021), except for positive

Table 3Carbon balance components and C changes measured by flux differences and soil sampling.

Fertilizer	N level	WT	Manure	Roots	Heterotrophic	Leaching	CH ₄	ΔC^a	ΔC^{b}
			kg C ha ⁻¹						
Dry manure	250 N	FD	6314	6231	-9944	0.0	0.3	2602	Ü
(2011–2012)	(2011–2012)	WT120	6314	6718	-10962	-4.1	0.4	2067	
368 N		WT60	6314	6469	-12366	-6.3	0.2	411	
	368 N	FD	9286	3971	-10702	0.0	0.4	2555	
		WT120	9286	5597	-12779	-4.9	0.1	2099	
		WT60	9286	4922	-12788	-5.9	0.4	1414	
Fresh manure	250 N	FD	1396	6212	-12165	-6.0	0.2	-4563	
(2013–2014) 368 N		WT120	1396	5516	-10904	-9.7	-0.2	-4002	
		WT60	1396	4703	-10820	-11.4	-1.5	-4734	
	368 N	FD	2060	4102	-13529	-5.5	-0.4	-7373	
		WT120	2060	4346	-11749	-9.1	-1.0	-5352	
		WT60	2060	3642	-11669	-11.2	-0.6	-5979	
Dry + fresh manure $$250\ N$$ $$368\ N$$	250 N	FD	7710	12443	-22109	-11.6	0.4	-1968	-2394
		WT120	7710	12234	-21867	-26.7	0.3	-1949	-318
		WT60	7710	11173	-23186	-34.3	-1.1	-4339	-3298
	368 N	FD	11346	8073	-24230	-10.7	-0.1	-4823	-6268
		WT120	11346	9943	-24528	-27.3	-0.9	-3267	-7986
		WT60	11346	8564	-24458	-33.1	-0.2	-4581	-8264

^a C differences estimated according to the C balance method

emissions observed in 2014, likely due to volatilization of CH₄ from fresh manure, which had been stored previously (Dalby et al., 2021).

In their meta-analysis, Mathew et al. (2020) identified maize as one of the crops with the greatest allocation to the soil, and roots were found to play a significant role in the C balance in our study, especially under the low fertilization treatment where C-root constituted the primary input. This result is consistent with the idea proposed by Ordonez et al. (2021) that plants may allocate more C belowground to support shoot growth when resources are more limited. However, excessive soil water content can inhibit root growth and reduce the R/S ratio (Ren et al., 2016). Nichols et al. (2019) also found that the depth to the water table is a strong predictor of maize root investment in the top 30 cm of soil. Additionally, plant allocation between above and belowground organs is influenced by nutrient availability (Farrar and Jones, 2000). Research by Pausch and Kuzyakov (2018) showed that root biomass is negatively correlated with soil mineral N, which is consistent with the resource optimization hypothesis that increased nutrient availability reduces the carbon costs for nutrient acquisition (Ågren and Franklin, 2003; Farrar and Jones, 2000). This principle can also explain the low R/S ratio observed under optimal soil moisture conditions in our study.

Lower root C input under high N input (368 N), associated to the higher Rh, explains the higher C depletion estimated by both C balance and SOC stocks methods.

The crucial role of roots in the carbon balance of agricultural soils has been well documented in the past (Hirte et al., 2018; Keel et al., 2017; Wilts et al., 2004), with estimates suggesting that they can account for 30–90% of total organic carbon inputs. However, the role played by roots in the long-term soil storage has been recently reconsidered (Wang et al., 2023), which helps explaining the substantial SOC reduction observed in our experiment even considerable amount of C was returned to the soil.

As a final remark, it was evident that fresh manure played a fundamental role in the overall C loss measured over the 4-year period. The increase in SOC decomposition following fresh organic matter addition is often reported (Fontaine et al., 2003) and practices which enhance organic matter quality such as composting, biochar addition or anaerobic digestion have shown to limit C depletion with agronomic performances comparable to those of mineral fertilizers (Kok et al., 2022; Lerch et al., 2019; Piccoli et al., 2022, 2023). Moreover, fresh manure effect was already observed on long-term experiments conducted in the same soils. Indeed, a SOC depletion was measured after replacing farmyard manure with slurry (Morari et al., 2006), which was attributed by the authors as also the result of the combined addition of easily

mineralizable N and organic matter. On the other hand, mature farmyard manure increased SOC stocks (Dal Ferro et al., 2020) and showed humification coefficient roughly double that of slurry (Berti et al., 2016), producing high-quality humus with a higher degree of polycondensation and resistance to microbial attack (Nardi et al., 2004), which is essential for building stable SOC stocks, and substantiating our findings about the positive effect of dry manure. This stresses the importance of a careful choice of management practices for limiting SOC losses.

5. Conclusions

A four-year lysimeter experiment was set-up in order to evaluate the effect of shallow water table and its interaction with fertilization practices on GHGs emission and C balance.

Pivotal roles on C balance were played by root biomass, organic fertilizer and heterotrophic respiration while C leaching and CH₄ emission were less noticeable. A C depletion was measured under all treatments when heterotrophic respiration was not compensated by the consistent C input of organic fertilizer. Furthermore, high fertilization doses with fresh liquid manure and urea increased SOC mineralization and, in turn, decreased the C balance. These results confirm the critical conditions of the soils of Veneto Region whose SOC level has been decreasing in the last decades. As already demonstrated by previous studies conducted in the same region (e.g., Dal Ferro et al., 2020; Morari et al., 2006), the use of the solid manure (e.g. farmyard manure), even at high doses, is one or the most effective way to increase the SOC in the soils. By promoting or integrating incorporation of solid digestate produced by anaerobic digestion in soils could be a viable alternative, providing new insight into a circular economy model. This experiment confirmed also the importance of the interaction between shallow water table and type of organic carbon that is crucial for C balance in mineral soils, affecting both the root C input and heterotrophic respiration. It is important to note that this lysimeter study allowed us to strictly control experimental factors and monitor their effects on the C balance. However, the results are limited in validity to one type of soil and climate regime. Additionally, the study investigates the biogeochemical processes and their interactions with farming practices at the mesocosm scale. Therefore, further studies are needed to confirm the effects of a shallow water table in mineral soils under different pedo-climatic conditions, while considering real farming management practices at the field scale. To conclude, our initial hypothesis that C losses would increase at increasing WFPS and high N input was partially accepted,

^b C differences estimated according to the soil samplings in the 0-60 cm depth

being C losses ranking FD>WT60 >WT120. On the other hand, results suggest that maintaining water table level around 120-cm depth could limit SOC depletion.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Data Availability

Data will be made available on request.

Acknowledgments

The authors are grateful to Elisa Cocco, Riccardo Polese, and all the staff of the Experimental Farm "L. Toniolo" for their precious support during the investigation operations. This research was co-funded by the Rural Development Programme for Veneto 2007–2013.

Appendix A. Supporting information

Supplementary data associated with this article can be found in the online version at doi:10.1016/j.agwat.2023.108508.

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