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Accurate evaluation of the Birch effect requires continuous CO_2 measurements and relevant controls

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Highlights

- Daily soil organic carbon mineralization rate increased with soil moisture
- A control scenario with water content equal to the average content in dry-wet cycles is suitable
- There is no effect of dry-wet cycles on net CO_2 emission in cultivated Luvisols
- Not accounting the CO_2 emissions in the drying phase may overestimate the dry-wet cycles effect

Abstract

The influence of dry-wet cycles (DWC) on soil organic carbon (SOC) decomposition is still debated given the somehow controversial results observed in the literature. The objective of this study was to evaluate the effects of DWC on SOC mineralization relative to various moisture controls in 7 treatments from two long-term French field experiments presenting contrasted SOC concentrations. A laboratory incubation was conducted for 97 days to quantify CO_2 emissions upon four soil moisture scenarios: continuously wet scenario at pF 1.5 (WET), continuously moderate wet scenario at pF 2.5 (MWET), continuously dry scenario at pF 4.2 (DRY) and dry-wet cycles (DWC) between pF 1.5 and 4.2 . Each cycle contained two phases, 10 days of drying phase, followed by 7 days of moist phase after rewetting. The drying phase consisted of adding silica gel to the incubation jars to absorb water in the soil and then gradually drying the soil We also calculated the SOC mineralization that would correspond to the average water content in DWC (mean_DWC). Our results showed that across all treatments the daily carbon mineralization rate increased with soil moisture (WET>MWET>DRY). In DWC scenario,

mineralization rates fluctuated with the changes in soil moisture. As soils dried, daily mineralization rates decreased and the subsequent soil rewetting, to pF 1.5, caused a rapid mineralization flush or "Birch effect". However, these flushes did not compensate for the low mineralization rates in the drying phase as the cumulative mineralization was not higher in the DWC scenario compared to the mean_DWC which was the scenario with equivalent water content as the DWC. We also observed that not accounting the CO_2 emissions in the drying phase, could lead to an overestimation of the effect of DWC. We recommend to measure continuously the soil respiration during dry-wet experiments and to compare the CO_2 emitted in DWC with a control that has a water content equivalent to the average water content in DWC. In addition, we questioned the importance of the effect of DWC on overall soil respiration.

Keywords: *Dry-wet cycles, carbon mineralization, soil moisture, soil organic carbon.*

Short communication

Soil moisture, in addition to temperature, is a major environmental factor controlling soil organic carbon (SOC) mineralization (Davidson et al., 1998, Davidson et al., 2006, Luo et al., 2016, Sierra et al., 2015). In general, SOC mineralization and the subsequent release of CO_2 from soil respond parabolically to transitional changes in soil moisture (Moyano et al., 2012) as follows: under low soil moisture conditions, SOC mineralization is decreased because of reduction of substrate availability and of microbial activity; under increased moisture conditions, substrate availability and microbial activity are enhanced. However, heterotrophic soil respiration is diminished by reduced oxygen diffusivity under saturated soil moisture conditions. This parabolic relationship between soil organic matter (SOM) decomposition and soil moisture has been well documented and incorporated into a number of models simulating SOM decomposition and the global carbon cycle (Davidson et al., 1998, Davidson et al., 2006, Luo et al., 2016, Sierra et al., 2015).

Climate change has been found to exacerbate terrestrial water evaporation and increase atmospheric moisture content, resulting in frequent droughts and heavy precipitation events (Dai, 2013, Donat et al., 2016). Under climate change, many surface soils are likely to experience more frequent and/or intense rewetting in response to rainfall events that occur following dry conditions (Borken and Matzner, 2009). Fluctuations in soil heterotrophic activity in response to changes in soil moisture are ubiquitous (Manzoni et al., 2012). It is recognized that droughts tend to decrease biological activity, and the subsequent precipitation would lead to "pulses" of CO_2 emissions to the atmosphere (Birch, 1958). These dry-wet events can influence several processes in soils, including soil aggregation (Denef et al., 2001, Cosentino et al., 2006), microbial activity and community structure (Tiemann and Billings, 2011, Evans and Wallenstein, 2012), and SOC mineralization (Birch, 1958, Fierer and Schimel, 2003, Borken and Matzner, 2009, Zhang et al., 2020). The effect of dry-wet cycles on cumulative SOC mineralization remains however controverted in literature (Zhang et al., 2020). Results are very heterogeneous across studies, because of the diversity of drying-rewetting scenario considered, but likely also because of methodological issues. Indeed, there are methodological artefacts, as a number of studies do not measure CO_2 emissions during the drying phase (e.g., Beare et al., 2009, Chow et al., 2006, Kaisermann et al., 2013, 2015), while others do so systematically (e.g., Miller et al., 2005, Mikha et al., 2005, Harrison-Kirk et al., 2013, Shi and Marschner, 2014, 2017, Yemadje et al., 2017). This makes the interpretation of results more complex, because the carbon budget is not complete in such studies. Although flushes of CO_2 emission have been widely reported (Birch effect), it is essential to understand whether the alternance of drying and rewetting conditions leads to a net increase of the CO_2 emitted by soils.

In the present study, we compared the SOC mineralization in soil subjected to dry-wet cycles (DWC) and kept at various constant water contents. Soils were sampled from two French long-term field experiments on Luvisols of similar texture situated both in Versailles' area with a mean annual temperature of 11.6 °C and precipitation of 633 mm during the study period, but under contrasting agricultural practices, in order to encompass a diversity of SOC contents. At the "La Cage" field experiment (48°48'N,2°08'E) we considered 3 treatments: conservation agriculture (CA), organic agriculture (ORG) and conventional agriculture (CON-LC), which are presented in detail along with crop rotations, soil management and fertilization in Autret et al. (2016). At the "QualiAgro" experiment (48°52′N, 1°57′E), the soil has been cultivated for 21 years with a conventional wheat-maize rotation and organic waste products (OWPs) are applied every 2 years as presented in detail in Peltre et al. (2012) and Paetsch et al. (2016). On this site we considered three organic treatments: residual municipal solid waste (MSW) composts, biowaste composts (BIOW) and farmyard manure (FYM) and one control treatment without organic inputs (CON-QA). At both sites, four replicate plots were available per treatment. From each plot, 3 sub-samples were taken, thoroughly mixed and combined into one sample. Samples were sieved to 4 mm, homogenized, plant material was removed and stored at 4 °C before incubation experiences.

Based on continuous measurements of soil moisture at the QualiAgro experiment during three consecutive years, we selected -0.015 MPa (pF 1.5) as the upper soil moisture value and -1.6 MPa (pF 4.2) as the lowest soil moisture condition. Indeed, these measurements showed that, at 20 cm depth, the soils never dried out beyond pF 4.2, even in summer (see Fig. S3). The choice of pF 4.2 is therefore reasonable for an extreme drying scenario in these soils. A prior experiment was conducted to determine the time required to dry the soil from a matric potential of -0.015 MPa (pF 1.5) to -1.6 MPa (pF 4.2) with silica gel. Gravimetric soil water content (w) was determined by weight loss at 105 °C for 48 hours. For each soil sample, we made cylinders of soil with soil aggregates < 4 mm at a bulk density of 1.3 $g.cm^{-3}$. Based on the initial water content, these soil cylinders were gradually adjusted to pF 2.5 by adding water with a pasteur pipette. Then, these soil cylinders were mounted in 1L jars containing 15 mL of water at the bottom to stabilize the moisture. The jars were sealed and the whole set was placed in the incubator at 20 °C for a one-week pre-incubation. To evaluate the dry-wet cycles (DWC) effect on SOC mineralization, 4 scenarios were performed and applied at 20 °C: a continuously wet scenario at pF 1.5 (WET); a continuously moderate wet scenario at pF 2.5 (MWET); a continuously dry scenario at pF 4.2 (DRY) and a scenario with five subsequent dry-wet cycles (DWC). To reach pF 1.5 after pre-incubating at pF 2.5, we added deionized water using a pasteur pipette to replicates dedicated to the WET scenario. For replicates dedicated to MWET scenario (pF 2.5), we checked if the soil moisture corresponded to pF 2.5. For the replicates dedicated to the DRY scenario, we introduced 50 g of silica-gel during the pre-incubation phase to dry the soil. As soon as the moisture level corresponding to pF 4.2 was reached, we removed the silica-gel from the jars. For the DWC scenario, five dry–wet cycles were implemented during the experimental period (97 d). Each cycle contained two phases: 10 days of drying phase, followed by 7 days of moist phase after rewetting. The drying phase consisted of adding silica gel to the incubation jars to absorb ambient moisture and hence gradually drying the soil (Fig. 1b). 50 g of silica gel were added to each jar and changed after 6 days when the jars were vented after measuring the CO_2 concentration in the headspace by micro-chromatography. At the end of the drying period, rapid rewetting was performed by adding deionized water (the amount of water needed to reach pF 1.5) with a pasteur pipette (Fig. 1b). The same procedure was repeated for 5 times to simulate the dry-wet cycles events in the different treatments. We measured soil organic carbon (SOC) mineralization over time in both experiments non-destructively using a gas micro-chromatograph (µGC 490; Agilent Technologie; USA).

The CO_2 measurements on the constant moisture scenarios allowed us to establish a relationship between water content and daily mineralization rate. Based on this relationship, we reconstructed the theoretical value of carbon mineralization corresponding to an average water content measured in the dry-wet cycles during the experiment (Fig. 2b). This theorical value is not often used in dry-wet cycle studies. Thus, we called this theorical value "mean_DWC". Weighing the mass of the cylinders at each CO_2 measurement allowed us to calculate this average water content in the dry-wet cycles (w_{DWC}). To do this, we first calculated the average amount of water lost (m_{lost}) per day when silica gel is introduced during the drying phase after 6 days and last 4 days using this formula:

$$m_{lost\,(g\,H_2O)} = \frac{\left(\frac{(m_{d0}-m_{d6})}{6}+\frac{(m_{d6}-m_{d10})}{4}\right)}{2}$$

(2), with m_{lost} the average mass of water lost per day during the drying phase, m_{d0} represents the mass of the soil cylinder at pF 1.5, m_{d6} the mass of the soil cylinder after 6 days of drying and m_{d10} the soil cylinder mass after the last 4 days of drying. Once this mass of water lost during the drying phase was known, we calculated the water content corresponding to each day via the following formula: $w_{d(\,g\,H_2O.g-1.soil)} = (w_{d-1} - m_{lost})$ (3), with w_d the water content on day d and w_{d-1} the water content on day d-1. The water content in the wetting phase after rewetting being constant, the average water content in the dry-wet cycles was obtained with the following formula: $W_{DWC(g\ H_2O.g-1.soil)} = \frac{w_{d0}+w_{d1}+\cdots+W_{d17}}{17}$ (4). The values of W_{DWC} are shown in Fig. S2 for different treatments. Given this average water content (W_{DWC}), we used the relationship between water content and daily mineralization rate stated above to determine the mineralization rate and cumulative mineralization corresponding to the average moisture value (mean_DWC) for all treatments (Fig. 2b).

All data were tested for normality and homogeneity of variance. Log-transformation was applied, if the transformation improved the normality and variance substantially. A one-way ANOVA and Tukey's test was used to detect the differences in amount of SOC mineralized values among soil moisture within each treatment. All statistical analyses were completed in R (version 4.0.2).

At the end of the incubations, our results showed that across all treatments, the daily carbon mineralization rate increased with soil moisture content: WET>MWET >DRY (Fig. 2a and Fig. S1). In DWC scenario, mineralization rates fluctuated with changes in soil moisture. As soils dried, daily mineralization rates decreased and were lower 10 days after drying started, corresponding to the time when soil had reached a matric potential of pF 4.2 (0.09 to 0.13 $g.H_2O.g^{-1}$ soil depending on the treatments, Fig. S2). Throughout the incubation, the SOC mineralization rate significantly decreased in all moisture scenarios whatever the field treatment (Fig. 2a and Fig. S1). During the drying period, the SOC mineralization rate of DWC scenario significantly decreased, compared with WET and MWET scenarios, but when soil reached pF 4.2 its mineralization rate was similar to the mineralization rate observed on the DRY scenario. These results could be explained by the fact that the diffusive transport of substrates and extracellular enzymes, as well as the active and passive mobility of microorganisms slow down with decreasing water potential and abating thickness and discontinuity of the water film (Broken and Matzner, 2009) which in turns decreases the accessibility of substrates to decomposers (Moyano et al., 2013). At a matric potential of -1.6 MPa (pF 4.2) microbial activity was found to be reduced (Franzluebbers et al., 1994, Pulleman and Tietema, 1999), which could also be due to a reduction in microbial populations (Schimel et al., 1999, Butterly et al., 2009) caused by drying events.

A subsequent rewetting of soils to pF 1.5 in this study caused a rapid mineralization flush (Fig. 2a) or "Birch effect" (Birch, 1958). In the literature, two mechanisms are considered to explain this phenomenon: the first is physiological and is linked to the consumption of intracellular solutes after the death of microbial cells during the drying event; the second is physical and is linked to the exposure of previously protected SOC following the destruction of aggregates (Denef et al., 2001, Fierer and Schimel, 2003, Bailey et al., 2019). Also, the rewetting leads to the re-establishment of hydrological connectivity between substrates and microorganisms (Manzoni et al., 2016). We found in the present study that the mineralization flushes after rewetting did not exceed the mineralization rates in the WET, MWET and mean_DWC scenarios (Fig. 2a and Fig. S1). These results suggest that the soils rewetting did not lead to additional carbon mineralization as expected. In addition, we observed that when soils are exposed to multiple DWC, the CO_2 flush after rewetting generally decreased and stabilized with increasing DWC number (Fig. 2a and Fig. S1). This result is in partial agreement with those of Baumann and Marschner (2013), who observed a decrease and even disappearance of CO_2 flushes with increasing DWC number. This may be due to depletion of available substrates (Shi and Marschner, 2017), to a change in microbial community composition (Fierer and Schimel, 2003), and/or the fact that soil aggregates are no longer disturbed (Denef et al., 2001).

Although CO_2 flushes were observed in all treatments after the rewetting phase, these flushes were not enough to compensate the low mineralization rates recorded in the drying phase on a cumulative basis, when considering MWET as a continuously wet control (Table. 1). This result is consistent with those obtained by Mikha et al. (2005) and Zhang et al. (2020): dry-wet cycles do not lead to a net increase in emitted CO_2. However, the effect of dry-wet cycles on cumulative soil carbon mineralization is much debated in the literature. Some studies found a stimulatory effect (Fierer et Schimel, 2002, 2003, Miller et al., 2005, Butterly et al., 2009), while others found an inhibitory effect (Mikha et al., 2005, Guo et al., 2012, Harrison-Kirk et al., 2013, Kaisermann et al., 2013, 2015, Yamadje et al., 2017, Rahman et al., 2018) or similar effect (Jager and Bruins, 1975; Degens and Sparling, 1995, Priemé and Christensen, 2001, Denef et al., 2001, Baumann and Marschner, 2013) of dry-wet cycles on cumulative soil organic carbon mineralization. Zhang et al. (2020) in their meta-analysis showed that, on average, dry-wet-cycles stimulated cumulative carbon mineralization by 72% compared to the dry control and inhibited cumulative carbon mineralization by 25% compared to the wet control, but had a minor effect compared to a medium wet control. The similar cumulative mineralization between the DWC and the medium wet control observed by Zhang et al. (2020), convinced us to reconstitute in our study a control whose water content was equivalent to the average water content in the dry-wet cycles.

When comparing the carbon mineralization in the DWC versus the corresponding mineralization at equivalent water content (mean_DWC), we found that DWC led to a similar cumulated mineralization for some treatments (CON-LC and CA) versus significantly less cumulated mineralization for other treatments (ORG, CON-QA, MSW, FYM and BIOW) (Fig. 3). The average water content in the DWC was generally lower than the water content corresponding to pF 1.5 and pF 2.5 (see Fig. S2). These results indicate that the observed effect of DWC on carbon mineralization depends much on the water content of the selected control scenario as discussed by Guo et al. (2012). It is relevant to have a control with a water content equivalent to the average water content in the dry-wet cycles. The recent study conducted by Zhang et al. (2022) considered a control scenario with a water content equivalent to the average water content in the dry-wet cycles (33.25% WHC) and observed that the flushes of mineralization after rewetting compensated the low mineralization in the drying phase.

Also, the previous study by Guo et al. (2012) using a range of controls with different water field pore space (WFPS) values (30, 45, 60, 75, 90% WFPS), demonstrated that cumulated carbon mineralization in the dry-wet cycles was similar to cumulated carbon mineralization in the control whose water content (60% WFPS) was equivalent to the average water content in the dry-wet cycles (63% WFPS). Next to the selection of an adequate moisture control, the carbon balance can be questioned, as it will depend on whether respiration measurements are performed on both drying and rewetting phases or not.

While a number of studies only account the CO_2 fluxes measured in the rewetting phase to evaluate the effect of dry-wet cycles (e.g., Beare et al., 2009, Chow et al., 2006, Degens and Sparling, 1995; Kaisermann et al., 2013, 2015, Li et al., 2010, 2011, Shah and Gaebler., 2016, Tiemann et al., 2011), the experimental design implemented in our study allowed for continuous CO_2 measurement. We therefore sought to evaluate to which extent measuring evolved CO_2 only during the DWC wet phases versus continuously affected the results. The results in Table. 1 show that when considering continuous measurements, we found a high inhibitory effect (ΔCmin1: -11.5 to -22.4 mg C. g^{-1}. SOC) of DWC relative to MWET on heterotrophic respiration, whereas when measuring evolved CO_2 discontinuously, i.e., only in the rewetting phase, the inhibitory effect of DWC relative to MWET was less important (ΔCmin2: -1.26 to -5.82 mg C. g^{-1}. SOC). These results suggest a possible overestimation of the effect of dry-wet cycles on SOC mineralization if the measurements are only carried out in the rewetting phase. Indeed, during the drying phase, a substantial amount of carbon is mineralized, but this amount is not accounted for in the global evaluation.

We did not measure any net effect of dry wet cycles on net CO_2 mineralization for cultivated Luvisols with contrasted SOC contents (Table 1). Several studies have reported that a longer drying phase resulted in a larger CO_2 flush after rewetting (Birch, 1960, Meisner et al., 2013), likely because longer dry periods lead to more accumulation of carbon substrates available for respiration after rewetting (Chowdhury et al., 2011). It is hence possible that in the present study, the drying duration and intensity was not sufficient to cause a large accumulation of the substrate. However, the duration and intensity of drying used in this study is close to the scenarios used in several other studies. Some studies dried their soils more than we did here (e.g., Beare et al., 2009, Butterly et al., 2009, Degens and Sparling, 1995, Kaisermann et al., 2013, 2015), while some did not (e.g., Fierer and Schimel, 2002, Chow et al., 2006).

Based on these results, we strongly recommend to measure continuously the soil respiration during dry-wet experiments and to compare the CO_2 emitted in dry-wet cycles with a control that has a water content equivalent to the average water content in dry-wet cycles. In addition, we question the importance of the effect of dry-wet cycles on overall CO_2 emissions, while acknowledging that, even though the carbon balance might be similar, the fate of nitrogen might be very contrasted under dry wet cycles, carbon peaks being associated with nitrogen peaks, which are easily leached into the soil by rain.

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Table. 1: Soil organic carbon contents, cumulative SOC mineralization calculated from continuous measurements (MWET, DWC) or with partial measurements only in the wet phase (pMWET and pDWC). ΔCmin1 and ΔCmin2 represent the delta SOC mineralized. If the value of ΔCmin1 or ΔCmin2 is negative, it means that DWC has an inhibitory effect, while, if the value of ΔCmin1 or ΔCmin2 is positive, it means that DWC has a stimulatory effect. MWET represent a scenario continuously at pF 2.5 and DWC: Dry-Wet-Cycles scenario.

LTEs	**TREATMENTS**	**SOC**	**SOC mineralization (mg C g^{-1}. SOC)**					
		$g.kg^{-1}$	MWET	DWC	pMWET	pDWC	ΔC_{min1} = DWC-MWET	ΔC_{min2} = pDWC-pMWET
La Cage	CON-LC	9.82 ± 0.48	44.3 ± 7.4	32.7 ± 4.7	20.76 ± 3.62	18.15 ± 4.17	-11.6 ± 8.7	-2.61 ± 5.52
	ORG	10.39 ± 0.42	55.7 ± 3.1	33.3 ± 4.3	25.98 ± 1.69	20.16 ± 1.67	-22.4 ± 5.3	-5.82 ± 2.37
	CA	13.30 ± 1.05	46.7 ± 2.6	32.8 ± 6.7	24.02 ± 1.44	18.75 ± 2.53	-13.9 ± 7.1	-5.27 ± 2.91
QualiAgro	CON-QA	10.11 ± 1.12	49.2 ± 2.2	32.2 ± 2.6	22.88 ± 2.50	21.62 ± 2.09	-17.0 ± 3.4	-1.26 ± 3.26
	MSW	13.38 ± 0.45	38.6 ± 3.3	25.5 ± 0.8	17.81 ± 1.67	15.77 ± 1.77	-13.1 ± 3.4	-2.04 ± 2.43
	FYM	14.31 ± 0.63	39.9 ± 2.4	26.5 ± 1.1	18.50 ± 1.18	16.88 ± 1.42	-13.4 ± 2.6	-1.62 ± 1.84
	BIOW	15.81 ± 1.23	35.8 ± 1.8	24.3 ± 1.9	16.60 ± 1.03	14.85 ± 1.42	-11.5 ± 2.6	-1.75 ± 1.75

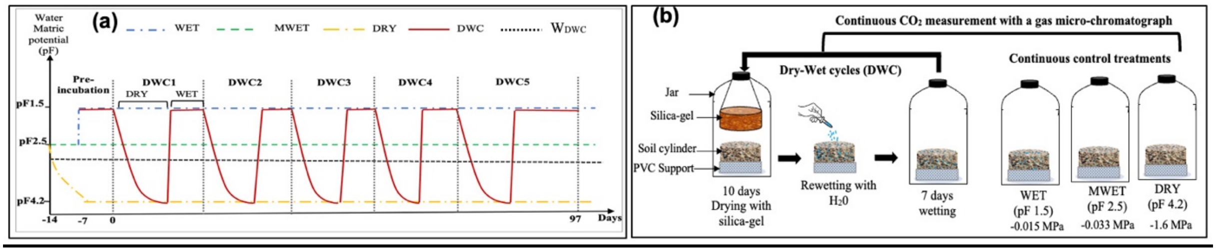

Fig. 1: (a) A schematic diagram of the experimental design showing constant moisture scenario (WET, MWET, DRY), average water content in dry-wet cycles (W_{DWC}) and dry-wet cycles scenario (DWC), (b) diagram of incubation procedures. WET represents a continuously wet scenario at pF 1.5; MWET represents a continuously moderate wet scenario at pF 2.5 and DRY represents a continuously dry scenario at pF 4.2

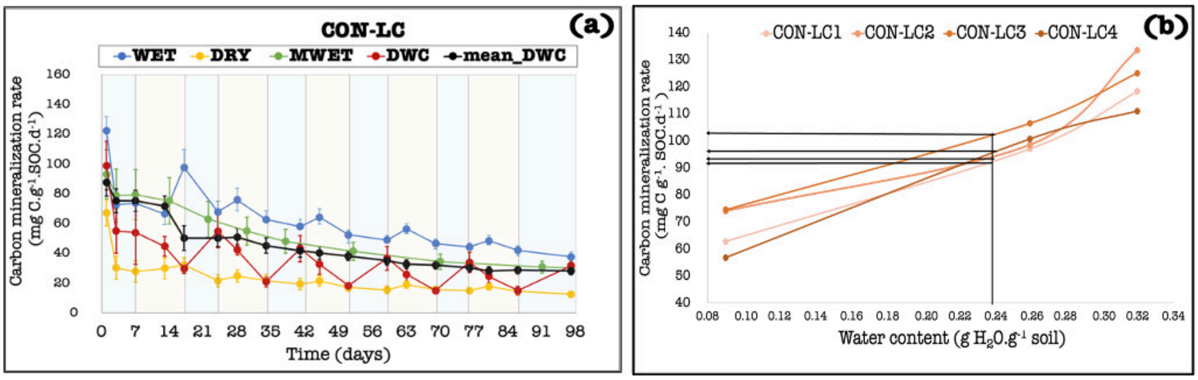

Fig. 2: (a) Carbon mineralization rates under constant soil moisture and dry-wet-cycles scenario for soil from plot under conventional agriculture at La Cage experiment (CON-LC). Error bars indicate standard deviation of 4 replicates per treatment. (b) Relationship between water content and daily mineralization rate on the CON-LC plot to reconstruct the mineralization rate corresponding to an average moisture in DWC scenario.

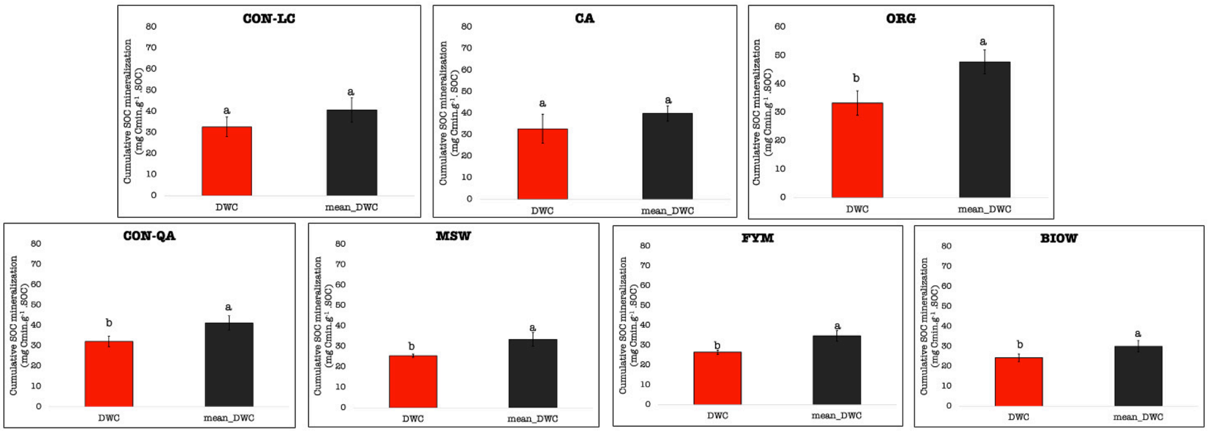

Fig. 3 Cumulative SOC mineralization upon dry-wet-cycles and mean_DWC under conservation agriculture (CA), organic agriculture (ORG), conventional agriculture at La Cage (CON-LC); Conventional agriculture without organic inputs at QualiAgro (CON-QA); residual municipal solid waste compost (MSW); Farmyard Manure (FYM) and Biowaste Compost (BIOW). Error bars indicate standard deviation of 4 replicates per treatment.

Supplementary materials

Accurate evaluation of the Birch effect requires continuous CO_2 measurements and relevant controls

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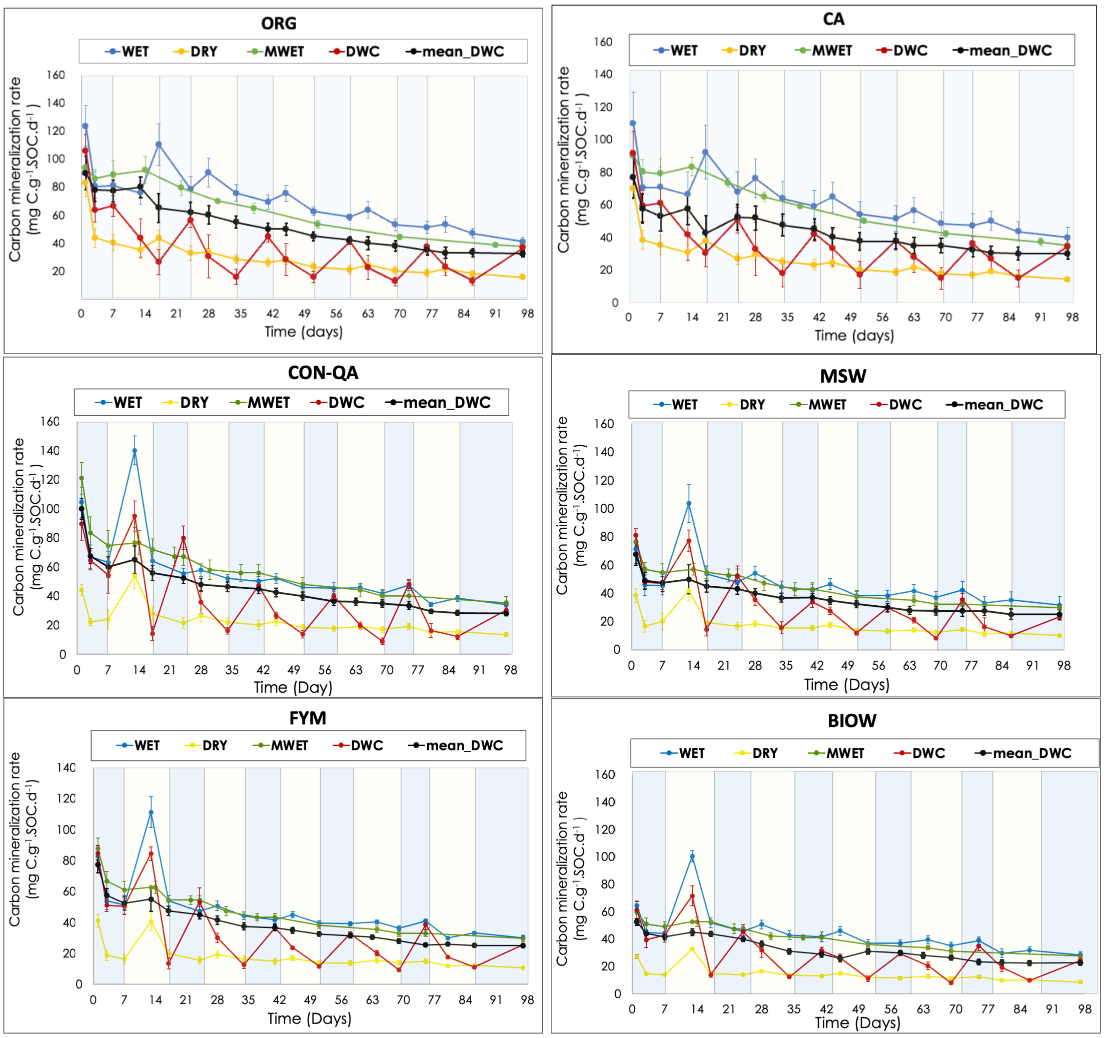

Fig. S1: Carbon mineralization rates under constant soil moisture and dry-wet-cycles scenario for soil from plots under conservation agriculture (CA), organic agriculture (ORG), CON-QA: control treatment without organic inputs; MSW: residual municipal solid waste compost; FYM: Farmyard Manure and BIOW: biowaste compost. Error bars indicate standard deviation of 4 replicates per treatment.

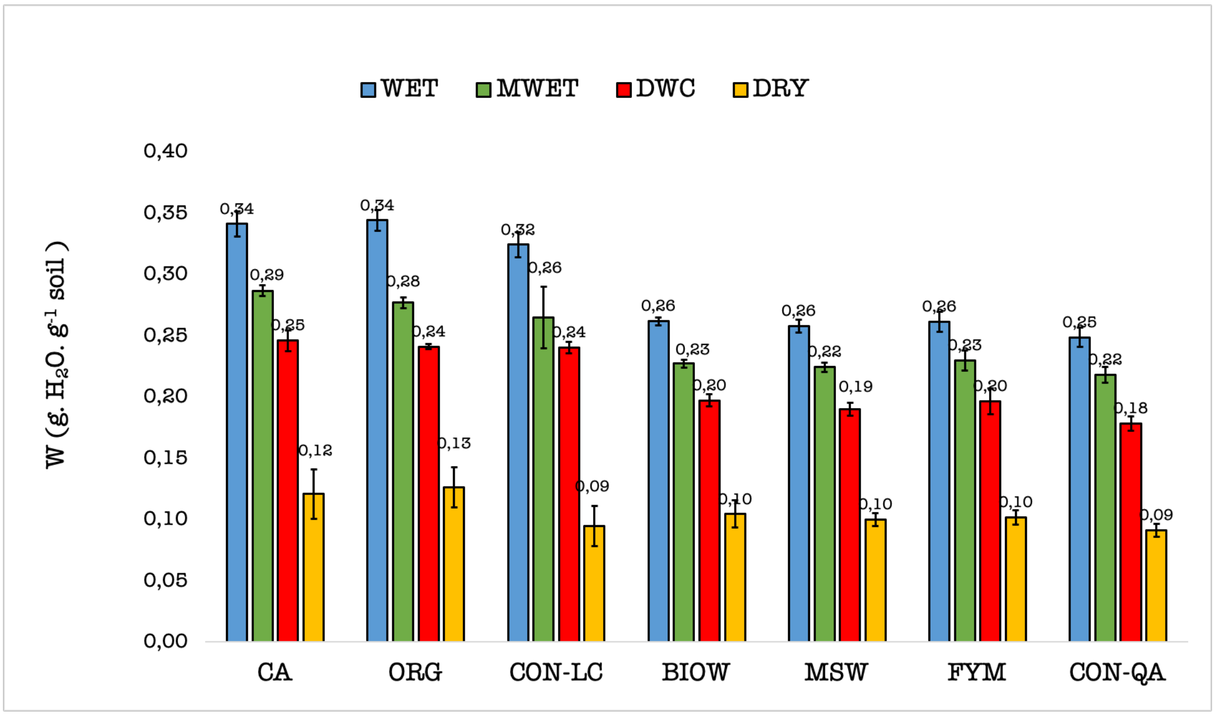

Fig. S2: Soil water content of the different moisture treatments tested. In the case of the DWC treatment it corresponds to an average soil moisture during drying and rewetting phases (W_{DWC}). Error bars indicate standard deviation of 4 replicates per treatment.

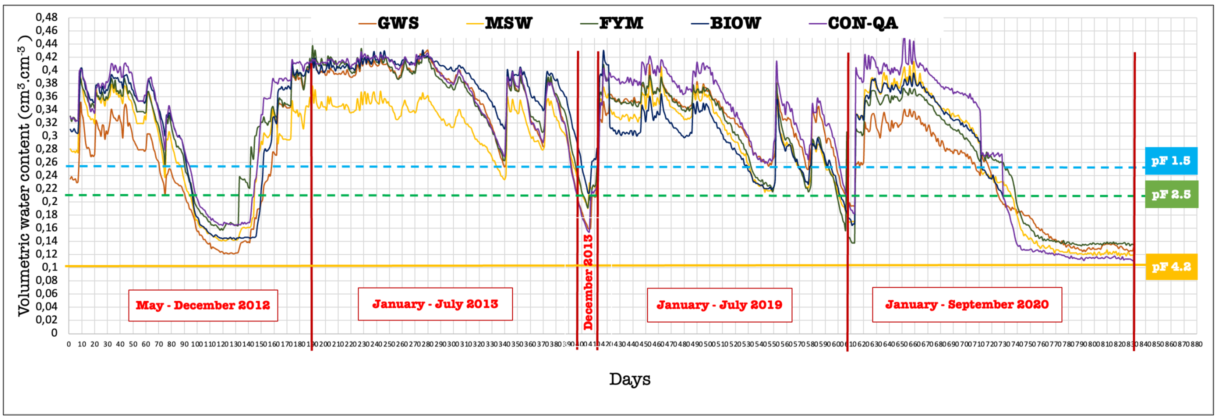

Fig. S3: Continuous measurement of volumetric water content at a depth of 20 cm in a Luvisol at QualiAgro. CON-QA: control treatment without organic inputs, BIOW: biowaste compost, MSW: municipal solid waste compost, GWS: green waste and sewage sludge compost and FYM: farmyard manure amended soils.