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Differences in soluble organic carbon chemistry in pore waters sampled from different pore size domains

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ABSTRACT

Spatial isolation of soil organic carbon (SOC) in different sized pores may be a mechanism by which otherwise labile carbon (C) could be protected in soils. When soil water content increases, the hydrologic connectivity of soil pores also increases, allowing greater transport of SOC and other resources from protected locations, to microbially colonized locations more favorable to decomposition. The heterogeneous distribution of specialized decomposers, C, and other resources throughout the soil indicates that the metabolism or persistence of soil C compounds is highly dependent on short-distance transport processes. The objective of this research was to characterize the complexity of C in pore waters held at weak and strong water tensions (effectively soil solution held behind coarse- and fine-pore throats, respectively) and evaluate the microbial decomposability of these pore waters. We saturated intact soil cores and extracted pore waters with increasing suction pressures to sequentially sample pore waters from increasingly fine pore domains. Ultrahigh resolution mass spectrometry of the SOC was used to profile the major biochemical classes (i.e., lipids, proteins, lignin, carbohydrates, and condensed aromatics) of compounds present in the pore waters; some of these samples were then used as substrates for growth of *Cellvibrio japonicus* (DSMZ 16018), *Streptomyces cellulosae* (ATCC® 25439™), and *Trichoderma reseei* (QM6a) in 7 day incubations. The soluble C in finer pores was more complex than the soluble C in coarser pores, and the incubations revealed that the more complex C in these fine pores is not recalcitrant. The decomposition of this complex C led to greater losses of C through respiration than the simpler C from coarser pore waters. Our research suggests that soils that experience repeated cycles of drying and wetting may be accompanied by repeated cycles of increased CO_2 fluxes that are driven by i) the transport of C from protected pools into active, ii) the chemical quality of the potentially soluble C, and iii) the type of microorganisms most likely to metabolize this C.
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1. Introduction

The spatial separation of substrate, microbes, and extracellular activity is an important mechanism of soil organic carbon (SOC) protection in soils. Spatial isolation of SOC in different sized pores may be a mechanism by which otherwise labile carbon (C) could be protected in soils (Jastrow et al., 2007). This mechanism has already been attributed to protecting otherwise labile nitrogen (N) in Arctic tussock soils, where over half of the labile N was calculated to be neither sorbed nor mobile (Darrouzet-Nardi and Weintraub, 2014). Such physical isolation of potentially labile C is intriguing, but the wide spectrum of complex organic C molecules in soil makes it difficult to extrapolate these observations to SOC. Determining if spatial isolation in soil pores results in distinctly different C chemical profiles is a first step to determining the vulnerability of this protected C. When soils are saturated, all pore channels are filled with water and SOC and other nutrient resources can mix with microorganisms through diffusion and transport. As soils dry, these hydrologic connectivities through the soil matrix reduce, and some pores become relatively isolated from these transport processes. Reduced hydrologic connectivity of soil pores can increase the bacterial diversity in soils due to limited competition that

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would normally reduce community diversity (Carson et al., 2010). Bacterial motility and resource diffusion are greatly reduced in soils at low matric potential that are dominated by thin water films, rather than saturated pores (Chowdhury et al., 2011). As soil moisture increases, the size of the labile C pool increases, inferred from fitted first- and zero-order kinetic models of soil respiration data (Bouckaert et al., 2013). Additionally, both the labile C pool size and the slow C pool mineralization rate increase with decreasing soil bulk density, i.e. increased total pore volume. There is evidence for an optimum balance between moisture and oxygen availability (Strong et al., 2004; Moyano et al., 2013) resulting in increased gas fluxes during soil wetting; additionally, the Birch effect of a significant respiration pulse following rewetting (Birch, 1958) may derive from the transport of occluded C to microbially active locations (Moyano et al., 2013). In the Birch effect, increased ionic strength due to drying can i) result in local osmotic stress lysing sensitive microorganisms (Moyano et al., 2013) and ii) changes to the chemical forms of C desorbed from soils under different ionic strengths (Mouvenchery et al., 2016). In the event that new C is mobilized through either process, pulses of soil moisture serve to facilitate the transport of this C from the protected location to microbially active sites, resulting in the Birch pulse of CO_2 (Lawrence et al., 2009; Parker and Schimel, 2011). This suggests that overall soil water distribution is a major driver for where C mineralization reactions occur in soils; ideal conditions may be found in finer pores surrounded by air-filled coarser pores (Bouckaert et al., 2013), or otherwise at the gas-water interface (Strong et al., 2004).

In studies of C decomposition at different matric potentials, the fastest decomposition of freshly added litter occurred in soils that were dominated by pore throats between 15 and 60 μm in diameter, and most slowly in soils with an abundance of pores with throat diameters <4 μm (Strong et al., 2004). However, this decomposition focused on freshly added C (Strong et al., 2004). Evaluating the C chemistry in native soils is intriguing because evidence suggests that soil C occurs as heterogeneously distributed "hotspots" throughout the soil, and that clear patterns of distribution do not exist for organic C chemical classes, other than the association of aliphatic and carboxylic compounds with clay mineral surfaces (Lehmann et al., 2007). Additionally, no consistent relationships between organic chemical class with soil residence time has been reported (Schmidt et al., 2011), suggesting that chemical composition doesn't limit decomposability. Thus, while "aged" or "weathered" soil C is often considered to be stable, under different moisture and temperature conditions this "stable" C will mobilize (Stutter et al., 2007). As C moves from protected locations, diffusion and decomposition processes make it difficult to predict the final spatial disposition of the solubilized C.

The objective of this research was to characterize the chemical composition of C in pore waters held at weak and strong water tensions (effectively soil solution held behind coarse- and fine-pore throats, respectively) and evaluate the microbial decomposability of the C dissolved in these pore waters. We hypothesized that the soluble C in these two pore domains would differ significantly from one another, with more condensed, aromatic SOC located in the finer pore domains because partially saturated conditions would isolate fine pores from the diverse microbial communities capable of decomposing these substrates. We also hypothesized that the chemical nature of the SOC in the pore waters would control its decomposition potential, with a fungal inoculant better able to deplete complex compounds (such as lignin and tannins) compared to the bacterial inoculants due to the differences in the metabolic potential of each microorganism. We tested these hypotheses in soils sampled across three locations within a hydrologic gradient at DWP, and at three soil depths. We studied intact soil cores collected from the Disney Wilderness Preserve (DWP), FL, USA. Water dynamics in this system, particularly water table rise and fall, influence the inputs and disposition of organic C at the surface and also at depth, making the DWP soils a good test case for examining how soil matrix accessibility, hydrologic connectivity, and decomposition potential are related.

2. Materials and methods

2.1. Soils

Intact soil cores (10 cm diameter, 30 cm height) were sampled from three locations across the Disney Wilderness Preserve (Orlando, FL) in June 2013. Soils at DWP are dominated by sandy textures, and depending on local topographic position show moderate to high levels of organic matter accumulation at the surface. The three locations were a drier pine flatwood stand where the soil was an Immokalee Series, sandy, siliceous, hyperthermic Arenic Alaquod (28.104641°, −81.419027°); an intermittent marsh composed of a Basinger fine sand, depressional (28.105535, −81.418896); and a wet "supermarsh" (28.099252, −81.417913) where the soil was a Floridana mucky fine sand, depressional. Soil cores were sampled from 0 to 30 cm, 30–60 cm, and 60–90 cm depth intervals from the same borehole. Four replicates of each set of sequential cores were sampled from each of the three locations, randomly located and separated by 2–5 m. Physical and chemical properties of the soils used in this study are included as Supplemental Table 1. Soils were placed on blue ice, and transported back to the University of Central Florida (Orlando, FL, USA), where they were placed in a freezer at −20 for 48 h in order to comply with USDA-APHIS regulations controlling transport of soil from a fire ant quarantine area, before being shipped to Pacific Northwest National Lab (Richland, WA, USA). Once received at PNNL, soils were stored at 4 °C prior to conditioning and saturation.

2.2. Pore water sampling

Soils were conditioned in the lab at room temperature (21 °C), and allowed to freely imbibe water from the bottom through a saturated porous ceramic plate (Soil Moisture Equipment Corp., Goleta, CA, USA). Cores were incubated under these conditions for 72 h. Pore waters were sampled by transferring each core onto individual 100 kPa Tempe Pressure Cell units fit with a high flow ceramic plate (Soil Moisture Equipment Corp. Goleta, CA, USA) to sequentially collect pore waters with suctions of −1.5 kPa, then −15 kPa, and finally, −50 kPa using a pump with a PCD Dual Valve pressure controller (Alicat Scientific, Tuscon, AZ). The ceramic pore plates have a pore size diameter of 2.5 μm, effectively removing particulates and some of the bacteria from the pore water. Pore water was collected into borosilicate vials for 24 h, or until flow ceased at a given suction, at each suction setting and stored at −20 °C until analysis. In this manuscript, pore water fractions will be identified by their sampling suctions. Preliminary tests indicated that the duration of suction had no effect on SOC composition.

The Kelvin equation for perfectly wettable soils can be used to estimate the largest water-filled pore diameter at a given water potential (Marshall and Holmes, 1996). It can be reduced to:

$$\text{largest water filled pore diameter } (\mu m) = \frac{300}{\textit{water potential } (kPa)}$$

Using the Kelvin equation (Marshall and Holmes, 1996; Carson

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