AN ABSTRACT OF THE THESIS OF

April R. Strid for the degree of Master of Science in Soil Science presented on June 5, 2015

Title: Homogenization of Detrital Leachate in an Old-Growth Coniferous Forest, OR: DOC Fluorescence Signatures in Soils Undergoing Long-Term Litter Manipulations.

Abstract approved: _____

Kate Lajtha

Soil dissolved organic carbon (DOC) is a small but crucial part of the forest carbon cycle. Characterizing the relationship between detrital inputs and soil DOC chemistry is crucial to understanding the ultimate fate of root carbon, fallen wood and needles. Chemical differences in the DOC pool may help to explain whether fractions are sorbed to mineral surfaces and contribute to accumulation of soil organic carbon, respired as CO_2, or exported to nearby catchments. Soil solution DOC was sampled from the detrital input and removal treatment (DIRT) plots located in the H.J. Andrews Experimental Forest, OR to determine whether detrital inputs impart a detectable signal on DOC in mineral soil. Multiple types of fresh litter extracts, along with lysimeter and soil extracts from DIRT treatment plots were characterized using UV-Vis and fluorescence spectroscopy coupled with the Cory and McKnight (2005) parallel factor analysis (PARAFAC) model. Principal component analysis of 13 unique fluorophores distinguished using PARAFAC show that litter and soil extracts (Douglas-fir needles, wood of decomposition Class 1, Class 3 and Class 5, O-horizon, and 0-5 cm A-horizon) each have distinct fluorescence signatures. However, while litter-leached DOC chemistry varies by litter type, neither lysimeter-collected DOC or soil extracts show statistically significant differences in fluorescence signatures among treatments, even after 17 years of litter manipulations. The lack of observed differences among DIRT treatments suggests an exchange hypothesis whereby both abiotic and biotic mechanisms effectively homogenize organic

carbon constituents within the dissolved pool. Change in PARAFAC components and optical indices with soil depth and during a 1-month biodegradation incubation of litter and soil extracts provided insight into the characteristics of PARAFAC components and the application of the Redox Index in soils. Patterns in the Freshness Index and Fluorescence Index as a result of incubation varied widely among replicates, indicating the indices may not be suitable in these soils. The results of this work emphasize the ability of sorption and biodegradation to homogenize soil DOC and demonstrate that fluorescence can be an effective fingerprinting technique for soil DOC composition.

Homogenization of Detrital Leachate in an Old-Growth Coniferous Forest, OR: DOC Fluorescence Signatures in Soils Undergoing Long-Term Litter Manipulations

by
April R. Strid

A THESIS

submitted to

Oregon State University

in partial fulfillment of
the requirements for the
degree of

Master of Science

Presented June 5, 2015
Commencement June, 2016

Master of Science thesis of April R. Strid presented on June 5, 2015

APPROVED:

Major Professor, representing Soil Science

Head of the Department of Crop and Soil Science

Dean of the Graduate School

I understand that my thesis will become part of the permanent collection of Oregon State University libraries. My signature below authorizes release of my thesis to any reader upon request.

April R. Strid, Author

ACKNOWLEDGEMENTS

I would like to thank my advisor Kate Lajtha for the constant support and enthusiasm for my success. I feel lucky to have had the opportunity to work with such a confident and creative researcher to learn about questions in biogeochemistry, and my own independence. My other committee members, Markus Kleber and David Myrold, have been patient and instructive throughout my time at OSU. Thanks to Markus for showing me the significance of an important question in scientific research and emphasizing the value of crystal-clear communication.

This work would not have been possible without the friendship and fluorescence spectroscopy expertise of Baek Soo Lee. Baek's collected composure and thoroughness in everything she touches is unmatched. Both Katie Watkins-Brandt and Baek must be recognized for their extensive knowledge and friendly assistance in Angel White's lab.

Thanks to Kim Townsend for teaching me to navigate the lab with an undying cheerful nature. Field work would have been slow-going and lonely without help from Kim, Ester Gordon, Emerson Helbling and Clint Burdette and I must recognize the H. J. Andrews headquarters crew for keeping everything running smoothly. I would also like to acknowledge my fellow graduate students for their kindness, attentiveness and humor throughout the highs and lows of research and graduate school. Lastly, I am fortunate to have had the opportunity to come to graduate school and thanks to all the family members, friends, and educators who have made this experience possible.

TABLE OF CONTENTS

Page

LIST OF FIGURES

LIST OF TABLES

Homogenization of Detrital Leachate in an Old-Growth Coniferous Forest, OR: DOC Fluorescence Signatures in Soils Undergoing Long-Term Litter Manipulations

Chapter 1:
General Introduction

April R. Strid

Global DOC Cycling

Dissolved organic carbon (DOC) represents the key carbon (C) transport path between the geosphere and hydrosphere and mobilization of DOC from soils is a vital piece of the global C cycle (Deb and Shukla 2011; Kindler et al., 2011; Sanderman et al., 2009). The primary source of soil DOC is the pool of soil organic carbon (SOC) that makes up more than two-thirds of the Earth's C stores (Simson and Simpson 2012). Previously, assessments overlooked C stabilized in deep soils and permafrost, inclusion of which brings the total SOC store to at least 2300 Pg (Tarnocai et al., 2010; Jobbagy and Jackson 2000). The dynamics of this large C pool must be accurately characterized for comprehensive global nutrient models. The flux of CO_2 to the atmosphere from soil respiration is an order of magnitude larger than the annual flux from anthropogenic greenhouse gas emissions and therefore small shifts in SOC cycling would have significant consequences for the global C cycle (Schimel et al., 1996; Reichstein and Beer, 2008). Losses from the SOC pool in the form of DOC leachates can have significant effects on ecosystem C stores (Gielen et al., 2010; Cole et al., 2007).

Export to nearby watersheds removes C from a soil and affects the forest C budget. Catchment-level export of DOC varies both seasonally and hourly (Strohmeier et al., 2013). Leaching from European forests has been estimated to be 3.5 ± 1.3 g C m^{-2} yr^{-1} (Kindler et al., 2011). On a global scale, 0.25 Gt yr^{-1} of DOC is exported through riverine systems to the ocean. Forests comprise 60% of the total with temperate and boreal forests exporting about half the amount from tropical forests (Schlesinger and Melack, 1981). Soluble C should not be ignored in forest soil C accounting.

DOC chemistry is variable within a catchment and differs among soil water, stream water, wetland and riparian groundwater (Inamdar et al., 2011). What is not well known is whether or not the source controls DOC composition and therefore if DOC origin can determine the ultimate fate of the dissolved pool. Thus, the sources of DOC and the role of fresh detritus in the formation of DOC are of particular interest. The relative impact of annual detrital additions such as needles, wood and roots compared to impact of existing SOM on DOC in a soil profile are not well understood. There tend to be high fluxes of leachate from surficial detritus and concentrations tend to drop with soil depth due to sorption (Michalzik et al., 2001). It is crucial that we identify how aboveground and belowground inputs control DOC characteristics because the chemistry has ramifications for long-term stabilization on mineral surfaces and on percolation and storage in mineral soils (Kramer et al., 2012; Fontaine et al., 2007; Rumpel and Kögel-Knabner, 2010). Prior to addressing the impact of forest floor litter on DOC composition, the following sections define DOC in the context of sampling methods and mechanisms involved in belowground DOC cycling. Here, the primary processes of interest include sorption and biodegradation and each of their effects on DOC composition.

Definitions and Sources

DOC is historically defined as the organic materials in soil solution that will pass through a 0.45 μm filter. Soil DOC consists of a complex mixture of organic compounds with various sizes due to the range of contributing forms of organic carbon. Soil colloids are also captured in a solution filtered through a 0.45 μm filter pore size (Zsolnay, 2003).

Sources include organic acids leached from recently-fallen leaf or needle litter, root exudates and microbial breakdown of existing SOC through enzymatic activities (Rosenfeld et al., 2014; Sanderman et al., 2008). A small fraction of DOC consists of low molecular weight organic acids and monomeric carbohydrates that are hydrophilic and therefore readily mobilized into solution (Hongve et al., 2000). The importance of each source and DOC composition depends on the position in a soil profile and sampling season (Asakawa et al., 2006; Inamdar et al., 2011).

Because soil DOC sampling methods vary widely, the interpretations of DOC amount and composition should occur in the context of the DOC sampling method. The heterogeneity of soil structure within a single site and the difficulties in comparing soil structures across sites poses complications to creating definitions of DOC pools that are scalable across landscapes. Sampling tools range from in-situ collection tools such as zero-tension and tension lysimeters to soil extractions with water or solutions of varying ionic strengths (i.e. Qualls, 2000). Methodologies vary in filter pore size, shake time, centrifugation and sample preparations (Chow et al., 2005; Guigue et al., 2014; Zsolnay et al., 1998).

Water-extracted soil DOC and field-collected DOC have been shown to have distinctly different origins (Hagedorn et al., 2004). ^{14}C in field-collected DOC reflected slightly younger ages than bulk SOC while soil solution has the same age as bulk SOC (Sanderman et al., 2008). DOC extraction procedures mobilize all soluble constituents into the dissolved pool that may not be accessible to microorganisms in field conditions due to spatial inaccessibility or sorption processes (von Lützow et al., 2007). In field-collected solutions, tools may be sampling soil solutions from macropore networks that

have particularly active microbial communities ("hot spot" regions) and the sampled solutions have already undergone significant microbial breakdown (Bundt et al., 2001; Sanderman et al., 2008). Accordingly, lysimeter solutions may capture only the residual DOC pool after initial decomposition. It is well documented that collection and soil extraction methods have significant effects on measured DOC character (Zsolnay, 2003; Akagi et al., 2007; Perdrial et al., 2012). Thus, definitions of DOC and fractions therein can vary depending on the context.

Belowground DOC Processing

Within a soil, C can enter the DOC pool and experience a combination of processes including sorption, desorption, flocculation, subsequent microbial processing and reprocessing (Kaiser and Kalbitz 2012; Corvasce et al., 2006). Temporary immobilization and subsequent solubilization occurs as naturally percolating waters and flow-through channel networks suffuse DOC through a soil (Marin-Spiotta et al., 2011). Abiotic and biotic processes alter DOC in such a way that fractions available for rapid microbial uptake or sorption to mineral surfaces are removed from solution, and thus the biochemistry of the remaining DOC pool differs from the biochemistry entering the soil (Kaiser and Kalbitz, 2012). The partitioning can affect what reaches deep soils and the output to the hydrosphere. Ultimately, the measured DOC pool represents a balance between *i.* inputs such as leachates from litter or native SOM or desorption and *ii.* removal of DOC through sorption or mineralization to CO_2 (Kalbitz et al., 2000). While sorption and biodegradation are not the only processes acting upon DOC, the balance

between inputs and outputs and the resulting DOC chemistry can be better understood through examining sorption and biodegradation of DOC.

DOC and Subsoil SOC Accumulation

Sorption removes C compounds from the dissolved pool for long or short time periods and is likely a key mechanism for accumulation of SOC (Kaiser and Guggenberger, 2000). Hydrophobic interactions, ligand exchange, cation bridging, hydrogen bonding, and/or Van der Waals forces can all affect the fate of DOC (i.e. Yano et al., 2004). Sorption is generally a stabilizing force through physical protection by mineral phases with high surface areas and charged surfaces (Kögel-Knabner et al., 2008; Torn et al., 1997). The SOC complex also includes hydrophobic or hydrophilic interactions between sorbed organics and other organic constituents (Kleber et al., 2007). Between 60 and 90% less DOC was mineralized when sorbed to mineral soil or soil minerals compared to DOC in solution during 375 day and 90 day incubations, respectively (Kalbitz et al., 2005; Mikutta et al., 2007). In a deciduous forest in the Oak Ridge Reservation, DOC-derived SOM associated with mineral surfaces was estimated to have mean residence time of 100-200 years while unassociated SOC turned over within 20-30 years (Tipping et al., 2012). Sorption of DOC removes dissolved constituents from solution and adds material to the SOC pool.

Studying DOC through separation into functional pools has begun to illustrate the importance of sorption in determining soil DOC composition. XAD-type resin fractionation shows that two acid fractions, hydrophilic and hydrophobic acids make up

the majority of the DOC pool in many soils and the exchange between these fractions and the solid phase explains changes DOC properties (Hassouna et al., 2012; Yano et al., 2004). Hydrophilic and hydrophobic pools are separated by their different affinities to nonionic resin. Hydrophobic acid fractions are further separated by determining if carboxylic acid groups are protonated at pH 2 and molecules are uncharged (Qualls and Haines, 1991). Hydrophobic acids include microbially-altered plant-derived material high in aromatics while the hydrophilic fraction includes microbially-synthesized compounds with high carboxyl-to-C ratios (Qualls and Haines, 1991). Fractionating DOC into hydrophilic and hydrophobic pools allows researchers to study functional differences in DOC composition through a soil profile.

It is apparent that separate soil horizons have DOC with distinct chemical compositions and these differences can be explained through preferential immobilization of some DOC through sorption. Dominance of hydrophilic fraction in mineral soil DOC has been demonstrated in soils including Ultisols (Qualls and Haines, 1991) and coniferous forests on Inceptisols and Spodosols (Guggenberger et al., 1994). While Oi leachates in an Andisols had a proportion of hydrophilic neutrals which are rich in carbohydrates, the neutral fraction was replaced by hydrophilic and hydrophobic acids in underlying horizons (Crow et al., 2009). Hydrophobic acids were dominant in the O-horizon while hydrophilic acid content increases in 0-10 cm of the mineral soil (Yano et al., 2004). In a laboratory sorption experiment, preferential removal of hydrophobic acids relative to hydrophilic acids suggests hydrophobic interactions are key in promoting DOC sorption (Yano et al., 2004). Ligand exchange between hydroxyls of clays and acidic functional groups could sorb both hydrophilic and hydrophobic acids because both

are high in carboxyls. However, hydrophilic acids have more carboxyl groups per C than hydrophobic acids and therefore would be more readily sorbed if ligand exchange was the dominant sorption mechanism. Hence, hydrophobic interactions help to remove hydrophobic acids from the dissolved phase and hydrophilic acids are retained in solution. Selective sorption could explain trends in $\delta^{13}C$, SUVA and C:N ratios. The decrease in aromatic-rich hydrophobic acids with depth and importance of hydrophobic-driven sorption may expound the corresponding decrease in soil solution SUVA values with depth (Hassouna et al., 2012). Moreover, retention of the ^{13}C-enriched, more microbially processed hydrophilic fraction in solution begins to illuminate the mechanism responsible for observed isotopic trends (Kalbitz et al., 2000). Differences among soil horizons of isotopic, spectroscopic and functional characteristics of DOC can be explained through selective sorption of DOC constituents.

Besides mineral interactions partitioning DOC, research exploring the fate of DOC that reaches subsoils in percolating waters suggests that sorption of DOC may be key in development of subsoil SOC (Kaiser and Kalbitz, 2012; Kramer et al., 2012; Kaiser and Guggenberger, 2000). Although comprising a large portion of the terrestrial C store, processes governing the behavior of deep SOC remain relatively unstudied. The interaction of DOC with mineral surfaces contributes more than commonly assumed to the accumulation of subsoil SOC. Using mineralization rate constants determined from incubations and estimates of DOC retention, Kalbitz and Kaiser (2007) estimate the total SOC derived from DOC sorbed to the Bw horizon in a Norway spruce forest in Germany represents between 19 and 50% of total SOC (Kalbitz et al., 2004). Soil solution C may be a major pathway for C movement and subsoil SOC accumulation.

Biodegradation

Biodegradation is also a key process manipulating the size and composition of the DOC pool. Methods for determining biodegradability include monitoring change in DOC concentration before and after incubations of varying lengths, monitoring production of CO_2 and flow-through bioreactors. Incubations may include added nutrients and can be done on solutions alone or intact litter or soils. While differences in methods can make comparing quantitative results across studies difficult, results have shown variability in total biodegradable DOC and dependence on DOC composition.

Not all DOC is readily decomposed. Incubations demonstrate variable decomposition of fresh litter leachates with total biodegradable DOC ranging from between 63 and 91% and only 14-33% of total measured DOC (Kalbitz, 2003; Qualls and Haines, 1992). While biodegradation studies of bulk DOC report decomposition of less than half the pool after approximately 100 day incubations, turnover of the low molecular weight component of DOC is rapid in forest soils (1-10 h) and suggests that there is high variability in biodegradability in different fractions DOC (van Hees et al., 2005). This estimate of turnover time suggests there may be a small portion of DOC with very rapid turnover times that make it an elusive pool of C to study (McDowell, 2003). Overall, it seems that DOC from unique source materials has different potential biodegradability.

Additionally, total biodegradable DOC varies on a seasonal basis, with soil depth and DOC composition (Yano et al., 2000). Biodegradability tends to decrease with depth and is lower in forest soils compared to agricultural soils (Kalbitz et al., 2003; Boyer and

Groffman, 1996). Leachate from fresher materials can be more readily decomposed relative to older, previously decomposed SOM (Bourbonniere and Creed, 2006). Additionally, hydrophilic and hydrophobic contents of DOC in fresh and decomposed litter leachates were not related to biodegradation (Bourbonniere and Creed, 2006).

DOC is considered by some to be a major substrate for microbial activity and shifts in biodegradability could influence soil respiration rates. Using $\delta^{13}C$ values of SOC and DOC, Bengtson and Bengtsson (2007) related the $\delta^{13}C$ values of respired CO_2 to the flow of C through the system and found that DOC production was the limiting factor decomposition of SOC. However, DOC release was found to have no correlation with CO_2 evolution in a laboratory leaching experiment using C additions to O-horizon soils (Park et al., 2002). The control of DOC production on total measured soil respiration rates could benefit from additional exploration.

Detritus and O-Horizon Leachates

The contributions of dissolved constituents mobilized from any surficial horizon (freshly fallen litter, Oi, Oe, Oa) to underlying horizons is thought to be minimal. While flux through the O-horizon is high, there is growing evidence that DOC at depths greater than a few centimeters into the mineral soil primarily originates from older, more processed SOM. ^{14}C measurements of Oi, Oe and Oa-horizon leachates in a Norway spruce stand in southern Sweden show that cycling of DOM may occur over centimeter spatial scales where DOM in the Oe sublayer originates within the Oe itself (Fröberg et al., 2003). In the same spruce forest, DOC concentrations in leachate from the O-horizon

depended on the composition of source SOM where the Oa horizon contributed the highest amount (50% of total), then Oe and Oi (Fröberg et al., 2005). The total amount of DOC leached from surficial detritus that reaches the mineral soil is relatively small.

The high DOC concentrations observed in litter leachates relative to mineral soils has prompted questions about the impact of fresh litter-derived DOC on mineral soil DOC concentrations and chemistry. Scheibe and Gleixner (2014) found that δ^{13}C-labelled beech and ash litter contributed less than 1% of total DOC from the mineral soil sampled repeatedly for 5 months. Likewise, Fröberg et al. (2007) observed that labelled spruce litter contributed less than 15% of total DOC flux underneath the O-horizon in a Norway spruce forest after 4 months. Research demonstrating that leachate from fresh litter may not contribute significantly to DOC pools in the mineral soil suggests there are mechanisms for rapidly removing fresh plant residue leachate from solution.

There are a variety of trends observed in DOC chemical properties as a function of soil depth that can be explained by sorption and biodegradation. Decreasing DOC concentrations with depth could be attributed to loss through mineralization to CO_2 and sorption to mineral surfaces. However, the available sorption capacity of a soil is not infinite and therefore interaction with mineral surfaces cannot maintain the observed subsoil low DOC concentrations for extended periods of time (Guggenberger and Kaiser, 2003). These two broad categories of processes, abiotic and biotic, may act in tandem because sorption to a biofilm, for example, can be a precursor for rapid turnover. Due to mineralogical and compositional gradients, it is likely that different processes dominate in different depths in a profile.

Detrital Input and Removal Treatment Network (DIRT)

The network of Detrital Input and Removal Treatment (DIRT) sites was established to explore the effects of aboveground and belowground organic matter inputs on soil C and nutrient cycling in forested ecosystems. Both litter and root exclusion treatments at DIRT sites in the University of Wisconsin Arboretum, H.J. Andrews, and Síkfőkút in Hungary have demonstrated changes in SOC, indicating the importance of both aboveground and belowground inputs for mineral soil C accumulation (Fekete et al., 2014; Lajtha et al., 2014a). SOC significantly decreased in the No Root treatments at the Hungary DIRT site after only 4 years of root exclusion (Fekete et al., 2014). Generally, the non-mineral associated fraction of SOC (<1.85 g/cm^3) has been the most responsive to changes to detrital inputs (Lajtha et al., 2014a; Lajtha et al., 2014b). A decrease in SOC has been observed in litter addition treatments at multiple sites, possibly due to fresh inputs providing a source of easily-degradable C necessary to stimulate biotic metabolisms to breakdown existing SOC (Lajtha et al., 2014a; Crow et al., 2009). However, litter manipulation sites in mixed oak forests in Wisconsin and a relatively dry oak forest in northeastern Hungary did not have reduced SOC stores as a result of litter additions (Lajtha et al., 2014a; Fekete et al., 2014). Significant changes in microbial community and enzyme activities have been recorded among H.J. Andrews, OR and Harvard Forest DIRT treatments (Brant et al., 2006; Lajtha et al., 2013). In the Germany site in a Norway spruce forest, Klotzbücher et al. (2012) observed no change in DOC concentrations due to litter exclusion. In the litter addition treatments at the same site, DOC concentrations increased due to changes in decomposition patterns in the Oi, Oe, and Oa layers. DOC release from the Oi layer increased but decreased in the Oe and Oa

as a result of litter additions (Klotzbücher et al., 2012). Adding or removing C in the form of detritus to soils does not result in a simple response from the SOC pool. Evidence exists to suggest that DOC responds in a similarly complex manner due to interactions with minerals, existing SOC, and soil biota (Klotzbücher et al., 2012).

Thesis Objectives

While recent work has answered many problems posed by DOC dynamics, questions remain in the characterization of the ephemeral and complex C pool. We aimed to determine whether long-term litter manipulations have altered the chemistry of DOC extracted from soil or on lysimeter-collected soil solutions from soils that have undergone 17 years of litter manipulations. UV-vis and fluorescence spectroscopy were used to fingerprint DOC chemistry and to assess the power of these tools and previously-correlated indices to characterize soil DOC.

References

Akagi, J., Zsolnay, Á., and Bastida, F. 2007. Quantity and spectroscopic properties of soil dissolved organic matter (DOM) as a function of soil sample treatments: Air-drying and pre-incubation. Chemosphere 69, 1040–6.

Asakawa, D., Mochizuki, H., Yanagi, Y., Suzuki, T., Nagao, S., and Fujitake, N. 2006. Changes in elemental composition, molecular weight and ^{1}H NMR spectra of the water-extractable hydrophobic acid fraction in Cambisol with season and soil depth. Soil Science and Plant Nutrition 52, 361–370.

Bengtson, P. and Bengtsson, G. 2007. Rapid turnover of DOC in temperate forests accounts for increased CO_2 production at elevated temperatures. Ecology Letters 10, 783–790.

Bourbonniere, R.A. and Creed, I.F. 2006. Biodegradability of dissolved organic matter extracted from a chronosequence of forest-floor materials. Journal of Plant Nutrition and Soil Science 169, 101–107.

Boyer, J.N. and Groffman, P.M. 1996. Bioavailability of water extractable organic carbon fractions in forest and agricultural soil profiles. Soil Biology and Biochemistry 28, 783–790.

Brant, J.B., Sulzman, E.W., and Myrold, D.D. 2006. Microbial community utilization of added carbon substrates in response to long-term carbon input manipulation. Soil Biology and Biochemistry 38, 2219–2232.

Bundt, M., Widmer, F., Pesaro, M., Zeyer, J., and Blaser, P. 2001. Preferential flow paths: Biological 'hot spots' in soils. Soil Biology and Biochemistry 33, 729–738.

Chow, A.T., Guo, F., Gao, S., Breuer, R., and Dahlgren, R.A. 2005. Filter pore size selection for characterizing dissolved organic carbon and trihalomethane precursors from soils. Water Research 39, 1255–1264.

Cole, J.J., Prairie, Y.T., Caraco, N.F., McDowell, W.H., Tranvik, L.J., Striegl, R.G., Duarte, C.M., Kortelainen, P., Downing, J.A., Middelburg, J. J., and Melack, J. 2007. Plumbing the global carbon cycle: Integrating inland waters into the terrestrial carbon budget. Ecosystems 10, 172–185.

Corvasce, M., Zsolnay, A., D'Orazio, V., Lopez, R., and Miano, T.M. 2006. Characterization of water extractable organic matter in a deep soil profile. Chemosphere 62, 1583–1590.

Crow, S.E., Lajtha, K., Bowden, R.D., Yano, Y., Brant, J.B., Caldwell, B.A., and Sulzman, E.W. 2009. Increased coniferous needle inputs accelerate decomposition

of soil carbon in an old-growth forest. Forest Ecology and Management 258, 2224–2232.

Deb, S.K., and Shukla, M.K. 2011. A review of dissolved organic matter transport processes affecting soil and environmental quality. Journal of Environmental & Analytical Toxicology 1, 1–11.

Fekete, I., Kotroczó, Z., Varga, C., Nagy, P.T., Várbíró, G., Bowden, R.D., Tóth, J.A., Lajtha, K. 2014. Alterations in forest detritus inputs influence soil carbon concentration and soil respiration in a Central-European deciduous forest. Soil Biology and Biochemistry 74, 106–114.

Fontaine, S., Barot, S., Barré, P., Bdioui, N., Mary, B., and Rumpel, C. 2007. Stability of organic carbon in deep soil layers controlled by fresh carbon supply. Nature 450, 277–80.

Fröberg, M., Kleja, D.B., Bergkvist, B., Tipping, E., and Mulder, J. 2005. Dissolved organic carbon leaching from a coniferous forest floor – a field manipulation experiment. Biogeochemistry 75, 271–287.

Fröberg, M., Kleja, D.B., and Hagedorn, F. 2007. The contribution of fresh litter to dissolved organic carbon leached from a coniferous forest floor. European Journal of Soil Science 58, 108–114.

Gielen, B., Neirynck, J., Luyssaert, S., and Janssens, I.A. 2011. The importance of dissolved organic carbon fluxes for the carbon balance of a temperate Scots pine forest. Agricultural and Forest Meteorology, 151, 270–278.

Guggenberger, G., and Kaiser, K. 2003. Dissolved organic matter in soil: challenging the paradigm of sorptive preservation. Geoderma 113, 293–310.

Guggenberger, G., Zech, W., Shulten, H.-R., 1994. Formation and mobilization pathways of dissolved organic matter: evidence from chemical structural studies of organic matter fractions in acid forest floor solutions. Organic Geochemistry 21, 51–66.

Guigue, J., Mathieu, O., Lévêque, J., Mounier, S., Laffont, R., Maron, P.A., Navarro, N., Chateau, C., Amiotte-Suchet, P., and Lucas, Y. 2014. A comparison of extraction procedures for water-extractable organic matter in soils. European Journal of Soil Science 65, 520–530.

Hagedorn, F., Saurer, M., and Blaser, P. 2004. A ^{13}C tracer study to identify the origin of dissolved organic carbon in forested mineral soils. European Journal of Soil Science 55, 91–100.

Hassouna, M., Théraulaz, F., and Massiani, C. 2012. Production and elimination of water extractable organic matter in a calcareous soil as assessed by UV/Vis absorption and

fluorescence spectroscopy of its fractions isolated on XAD-8/4 resins. Geoderma 189-190, 404–414.

Hongve, D., van Hees, P.A.W., and Lundstrom, U.S. 2000. Dissolved components in precipitation water percolated through forest litter. European Journal of Soil Science 51, 667–667.

Inamdar, S., Finger, N., Singh, S., Mitchell, M., Levia, D., Bais, H., Scott, D., McHale, P. 2011. Dissolved organic matter (DOM) concentration and quality in a forested mid-Atlantic watershed, USA. Biogeochemistry 108, 55–76.

Jobbágy, E.G. and Jackson, R.B. 2000. The vertical distribution of soil organic carbon and its relation to climate and vegetation. Ecological Applications 10, 423-436.

Kaiser, K., and Guggenberger, G. 2000. The role of DOM sorption to mineral surfaces in the preservation of organic matter in soils. Organic Geochemistry 31, 711–725.

Kaiser, K., and Kalbitz, K. 2012. Cycling downwards – dissolved organic matter in soils. Soil Biology and Biochemistry 52, 29–32.

Kalbitz, K. 2003. Changes in properties of soil-derived dissolved organic matter induced by biodegradation. Soil Biology and Biochemistry 35, 1129–1142.

Kalbitz, K., Schmerwitz, J., Schwesig, D., and Matzner, E. 2003. Biodegradation of soil-derived dissolved organic matter as related to its properties. Geoderma 113, 273–291.

Kalbitz, K., and Kaiser, K. 2007. Contribution of dissolved organic matter to carbon storage in forest mineral soils. Journal of Plant Nutrition and Soil Science 171, 52–60.

Kalbitz, K., Schwesig, D., Rethemeyer, J., and Matzner, E. 2005. Stabilization of dissolved organic matter by sorption to the mineral soil. Soil Biology and Biochemistry 37, 1319–1331.

Kalbitz, K., Solinger, S., Park, J.-H., Michalzik, B., and Matzner, E. 2000. Controls on the Dynamics of Dissolved Organic Matter in Soils: A Review. Soil Science 165, 277–304.

Kindler, R., Siemens, J., Kaiser,K., Walmsley, D.C., Bernhofer, C., Buchmann, N., Cellier, P., Eugster, W., Gleixner, G., Grünwald, T., Heim, A., Ibrom, A., Jones, S.K., Jones, M., Klumpp, K., Kutsch, W., Steenberg Larsen, K., Lehuger, S., Loubet, B., McKenzie, R., Moors, E., Osborne, B., Pilegaard, K., Rebmann, C., Saunders, M., Schmidt, M.W.I., Schrumpf, M., Seyfferth, J., Skiba, U., Soussana, J.-F., Sutton, M.A., Tefs, C., Vowinckel, B., Zeeman, M.J., Kaupenjohann, M. 2011.

Dissolved carbon leaching from soil is a crucial component of the net ecosystem carbon balance. Global Change Biology 17, 1167–1185.

Klotzbücher, T., Kaiser, K., Stepper, C., van Loon, E., Gerstberger, P., and Kalbitz, K. 2012. Long-term litter input manipulation effects on production and properties of dissolved organic matter in the forest floor of a Norway spruce stand. Plant and Soil 355, 407–416.

Kögel-Knabner, I., Guggenberger, G., Kleber, M., Kandeler, E., Kalbitz, K., Scheu, S., Eusterhues, K., and Leinweber, P. 2008. Organo-mineral associations in temperate soils: Integrating biology, mineralogy, and organic matter chemistry. Journal of Plant Nutrition and Soil Science 171, 61–82.

Kramer, M.G., Sanderman, J., Chadwick, O.A., Chorover, J., and Vitousek, P.M. 2012. Long-term carbon storage through retention of dissolved aromatic acids by reactive particles in soil. Global Change Biology 18, 2594–2605.

Lajtha, K., Crow, S.E., Yano, Y., Kaushal, S.S., Sulzman, E., Sollins, P., and Spears, J.D.H. 2005. Detrital controls on soil solution N and dissolved organic matter in soils: a field experiment. Biogeochemistry 76, 261–281.

Lajtha, K., Bowden, R.D., and Nadelhoffer, K. 2014b. Litter and Root Manipulations Provide Insights into Soil Organic Matter Dynamics and Stability. Soil Science Society of America Journal.

Lajtha, K., F. Peterson, K. Nadelhoffer, and R. Bowden. 2013. Twenty Years of Litter and Root Manipulations: Insights into Multi-Decadal SOM Dynamics. Soil Science Society of America Journal. *In review*.

Lajtha, K., Townsend, K.L., Kramer, M.G., Swanston, C., Bowden, R.D., and Nadelhoffer, K. 2014a. Changes to particulate versus mineral-associated soil carbon after 50 years of litter manipulation in forest and prairie experimental ecosystems. Biogeochemistry 119, 341-360.

Marin-Spiotta, E., Chadwick, O.A., Kramer, M., and Carbone, M.S. 2011. Carbon delivery to deep mineral horizons in Hawaiian rain forest soils. Journal of Geophysical Research 116, G03011.

Michalzik, B., Kalbitz, K., Park, J., Solinger, S., and Matzner, E. 2001. Fluxes and concentrations of dissolved organic carbon and nitrogen–a synthesis for temperate forests. Biogeochemistry 52, 173–205.

Mikutta, R., Schaumann, G.E., Gildemeister, D., Bonneville, S., Kramer, M.G., Chorover, J., Chadwick, O.A., and Guggenberger, G. 2009. Biogeochemistry of

mineral–organic associations across a long-term mineralogical soil gradient (0.3–4100 kyr), Hawaiian Islands. Geochimica et Cosmochimica Acta 73, 2034–2060.

McDowell, W.H. 2003. Dissolved organic matter in soils — future directions and unanswered questions. Geoderma 113, 179–186.

Park, J.J.-H., Kalbitz, K., and Matzner, E. 2002. Resource control on the production of dissolved organic carbon and nitrogen in a deciduous forest floor. Soil Biology and Biochemistry 34, 813–822.

Perdrial, J.N., Perdrial, N., Harpold, A., Gao, X., Gabor, R., LaSharr, K., and Chorover, J. 2012. Impacts of Sampling Dissolved Organic Matter with Passive Capillary Wicks Versus Aqueous Soil Extraction. Soil Science Society of America Journal 76, 2019.

Qualls, R.G., and Haines, B.L. 1991. Geochemistry of Dissolved Organic Nutrients in Water Percolating through a Forest Ecosystem. Soil Science Society of America Journal 55, 1111-1123.

Reichstein, M., and Beer, C. 2008. Soil respiration across scales: The importance of a model-data integration framework for data interpretation. Journal of Plant Nutrition and Soil Science 171, 344–354.

Rosenfeld, C.E., McCormack, M.L., and Martinez, C.E. 2014. A novel approach to study composition of in situ produced root-derived dissolved organic matter. Soil Biology and Biochemistry 76, 1–4.

Rumpel, C., and Kögel-Knabner, I. 2010. Deep soil organic matter—a key but poorly understood component of terrestrial C cycle. Plant and Soil 338, 143–158.

Sanderman, J., Baldock, J.A., and Amundson, R. 2008. Dissolved organic carbon chemistry and dynamics in contrasting forest and grassland soils. Biogeochemistry 89, 181–198.

Sanderman, J., Lohse, K. A., Baldock, J. A., and Amundson, R. 2009. Linking soils and streams: Sources and chemistry of dissolved organic matter in a small coastal watershed. Water Resources Research 45, 1–13.

Schimel, D., Alves, D., Enting, I., Heimann, M., Joos, F., Raynaud, D., and Wigley, T. 1996. CO_2 and the carbon cycle, in Houghton, J. T., et al. (eds.): Climate Change 1995: The Science of Climate Change. Contribution of Working Group I to the Second Assessment Report of the IPCC. Cambridge University Press, Cambridge, 65-86.

Simpson, M.J., and Simpson, A.J. 2012. The chemical ecology of soil organic matter molecular constituents. Journal of Chemical Ecology 38, 768–84.

Scheibe, A. and Gleixner, G. 2014. Influence of litter diversity on dissolved organic matter release and soil carbon formation in a mixed beech forest. PloS One 9, e114040.

Schlesinger W.H., Melack J.M. 1981. Transport of organic carbon in the world's rivers. Tellus 33, 172–87.

Strohmeier, S., Knorr, K.-H., Reichert, M., Frei, S., Fleckenstein, J.H., Peiffer, S., and Matzner, E. 2013. Concentrations and fluxes of dissolved organic carbon in runoff from a forested catchment: insights from high frequency measurements. Biogeosciences 10, 905–916.

Tarnocai, C., Canadell, J.G., Schuur, E.A.G., Kuhry, P., Mazhitova, G., and Zimov, S. 2009. Soil organic carbon pools in the northern circumpolar permafrost region. Global Biogeochemical Cycles 23, GB2023.

Tipping, E., Chamberlain, P.M., Fröberg, M., Hanson, P.J., Jardine, P.M. 2012. Simulation of carbon cycling, including dissolved organic carbon transport, in forest soil locally enriched with ^{14}C. Biogeochemistry 108, 97-107.

Torn, M.S., Trumbore, S.E., Chadwick, O.A., Vitousek, P.M., and Hendricks, D.M. 1997. Mineral control of soil organic carbon storage and turnover. Nature 389, 170–173.

van Hees, P.A.W., Jones, D.L., Finlay, R., Godbold, D.L., and Lundström, U.S. 2005. The carbon we do not see—the impact of low molecular weight compounds on carbon dynamics and respiration in forest soils: a review. Soil Biology and Biochemistry 37, 1–13.

von Lützow, M., Kögel-Knabner, I., Ekschmitt, K., Flessa, H., Guggenberger, G., Matzner, E., and Marschner, B. 2007. SOM fractionation methods: Relevance to functional pools and to stabilization mechanisms. Soil Biology and Biochemistry 39, 2183–2207.

Wagai, R., and Sollins, P. 2002. Biodegradation and regeneration of water-soluble carbon in a forest soil: leaching column study. Biology and Fertility of Soils 35, 18–26.

Yano, Y., Lajtha, K., Sollins, P., and Bruce, A.C. 2004. Chemical and seasonal controls on the dynamics of dissolved organic matter in a coniferous old-growth stand in the Pacific Northwest , USA. Biogeochemistry 71, 197–223.

Yano, Y., Mcdowell, W.H., and Aber, J.D. 2000. Biodegradable dissolved organic carbon in forest soil solution and effects of chronic nitrogen deposition 32, 1743–1751.

Zsolnay, Á., Baigar, E., Jimenez, M., Steinweg, B., Saccomandi, F. 1998. Differentiating with Fluorescence spectroscopy the sources of dissolved organic matter in soils subjected to drying. Chemosphere 38, 45–50.

Zsolnay, Á. 2003. Dissolved organic matter: artefacts, definitions, and functions. Geoderma, 113, 187–209.

Chapter 2:
Homogenization of Detrital Leachate in H.J. Andrews, OR: DOC fluorescence signatures in soils undergoing long-term litter manipulations

April R. Strid

Introduction

Soil dissolved organic carbon (DOC) is a vital component of the global carbon (C) cycle (Deb and Shukla 2011; Kindler et al., 2011; Sanderman et al., 2009). Because the retention, movement and formation of soil organic carbon (SOC) are invariably coupled with DOC dynamics, understanding the processes involved in DOC mobilization is essential to modeling the behavior of the large global SOC pool. Losses from SOC in the form of DOC exported to nearby catchments can make up a significant portion of net ecosystem productivity and systematically ignoring these losses can result in overestimation of C stores (Gielen et al., 2011; Cole et al., 2007).

Detrital inputs to soil vary in chemistry. Needles, wood and roots from different species at different stages of decomposition have unique compositions (Thevenot et al., 2010; Berg et al., 1982). Soil DOC originates from both fresh inputs and more processed SOC (Sanderman et al., 2008). While it has been demonstrated that large concentrations of DOC are solubilized from fresh detrital inputs, the contributions of needles, wood and root leachates relative to the influence of indigenous SOC on DOC in a soil profile are not well understood.

If long term changes in detrital input amount and chemistry alter the flux of DOC entering the subsoil, there are implications for the accumulation of SOC. Hydrologic transport moves C downward from surficial layers and subsequent sorption to mineral surfaces may be a major source of subsoil SOC accumulation (Kalbitz et al., 2005; Tipping et al., 2012). It was estimated that between 19 and 50% of SOC in a Bw horizon was derived from sorption of DOC (Kalbitz and Kaiser, 2007).

Previous research has not found a large impact of aboveground inputs on subsoil DOC chemistry. Less than 1 and 15% of mineral soil DOC was derived from fresh aboveground organic materials as assessed by additions of isotopically-labelled European beech and ash litter to a Luvisol and Norway spruce litter added to a Haplic Podzol, respectively (Scheibe and Gleixner, 2014; Fröberg et al., 2007). Belowground C cycling may occur on tight, centimeter-scales as evidenced by distinct DOC compositions within Oi, Oe, and Oa layers (Fröberg et al., 2003, 2005). However, these studies monitored litter inputs for less than 5 months and in the present work we aim to characterize the long-term effects of litter manipulations on DOC chemistry.

The Detrital Input and Removal Treatment (DIRT) plots in the H.J. Andrews Experimental Research Forest have undergone a suite of litter manipulation treatments for 17 years and have shown significant changes in total SOC content (Lajtha et al., 2005). Lysimeters and soil extracts from DIRT treatment plots provide an opportunity to assess the effects of long-term changes in detrital inputs on DOC cycling.

UV-vis and fluorescence spectroscopy have been used extensively to characterize DOC in freshwater, marine and more recently, soil solution. Methods are relatively cheap, can generally be applied to samples with DOC concentrations typical to field conditions, and have produced promising results as a fingerprinting technique for DOC research. For example, methods have aided in identifying DOC source materials, monitoring hydrologic flowpaths, and observing changes as a function of soil depth (Cory and McKnight, 2005; Toosi et al., 2014; Gabor et al., 2013). Separating fluorescence profiles into distinct fluorescing moeities using parallel factor analysis

(PARAFAC) allows for identification of subtle changes in spectral signals during decomposition of DOC or differences among leachate source materials.

The aim of this research is to (1) determine whether litter or roots impart a detectable signal on DOC in mineral soil (2) evaluate UV-vis and fluorescence spectroscopy as a tool to study changes in soil DOC with decomposition and source materials.

We hypothesized that fluorescence signatures would show that extractable DOC from roots, needles, and wood had unique chemistries that reflected the distinct compositions of the source materials (Yano et al., 2005). We expected that the changes in detrital input composition among DIRT treatments have altered the chemistry of DOC in soil extracts and lysimeter-collected soil solutions. However, upon solubilization from organic matter, DOC can be decomposed or incorporated into microbial biomass. DOC also can be sorbed to minerals or organic constituents. As a result of this extensive processing, chemical variation in DOC from different sources of detritus could be eliminated and the signature of DOC would be uniform among DIRT treatments.

Methods

Field site

The Detrital Input and Removal Treatment (DIRT) site at H.J. Andrews Experimental Forest (OR) was established in 1997 approximately 100 m from Lookout Creek (44°15’N, 122°10’W, 531-m elevation). Soils are a combination of Typic Hapludands, Andic Dystrudepts, and Vitrandic Dystrudepts in Douglas-fir, *Pseudotsuga*

menziesii and western hemlock, *Tsuga heterophylla* old-growth stands in H.J. Andrews (Dixon, 2003). The site has a total of 18 plots and six treatments. Each plot is 10 m x 15 m. Three plots were randomly assigned to one of six treatments (n = 3). Treatments are described in Table 1.

DIRT soil sampling and extraction

Soils were sampled with a 1.7 cm corer (0-10 cm) in March 2014. Within each plot, soil cores were taken from areas at least 1 m from plot perimeter with approximately 2-4 m separation between cores. If a selected core location happened to be on a decaying log, the sample was discarded and a new location selected. Three cores were taken from each plot and separated into three different depths (0-10 cm, 10-20 cm, and 20-30 cm). Three cores from each depth from each plot were consolidated to homogenize soil variability within a plot. Visible roots and rocks (>5 mm) were removed in the lab. Moist samples were stored at 2°C for less than 2 weeks until extraction.

Extraction was performed in 0.5 M K_2SO_4 in a 1:10 soil:solvent w/v ratio. Soils were placed on a horizontal shaker table for 2 hours and centrifuged for 45 minutes at 2400 rpm. Extracts were pre-filtered through combusted, GF/F Whatman filters and subsequently through pre-rinsed 0.45 μm cellulose acetate syringe filters and stored in the dark at 2°C prior to analysis. Control solutions of K_2SO_4 were shaken, centrifuged and filtered. All DOC concentrations of soil and litter extracts were measured on a Shimadzu Total Organic Carbon Analyzer (TOC-V_{CSH} SSM-5000A) with duplicates every 10 samples.

Litter sampling and extraction for biodegradation study

Needles, wood of different decomposition classes, A-horizon and O-horizon soils were collected from areas adjacent to DIRT treatment plots for characterization of DOC from different sources of detritus and for the biodegradation incubation (Table 2). Five replicates of all litter and soils were sampled. An off-plot 6 m x 6 m area was selected for O-horizon and A-horizon soil samples to be extracted. From an established grid, five grid-cells were randomly selected for sampling and visible tree roots and decaying logs were avoided. A 10 cm x 10 cm square was cut off of the soil surface and two 1.7 cm soil cores were taken from the 0-5 cm in the A-horizon beneath the collected O-horizon and homogenized. Needles fallen within the last 10 months onto nearby litter manipulation plots with screens were collected from distinct locations on plots. Wood samples include three different decomposition classes identified within the vicinity of the off-plot area and sampled from separate locations. Woodchips stored on-site that are added to Double Wood plots every other year were sampled from varying locations within the storage pile. Decomposition Class 2 refers to recently fallen, relatively undecomposed wood (Sollins, 1982). Sampled wood chips are placed in the Class 2 category due to exclusion of bark and minimal decomposition. Decomposition Class 3 is marked by rotting sapwood and bark that has sloughed off from fallen trees and Class 5 includes logs that are no longer intact and have begun to settle into the forest floor (Sollins, 1982).

Litter and soils were sieved moist, visible roots and animals removed. Moderately decomposed, fibrous wood was broken into pieces less than 2 cm in diameter. Extraction was performed in 0.01 M K_2SO_4 1:100 w:v ratio for needles, Class 2, Class 3, Class 5, and O-horizon and 1:10 for A-horizon samples due to varying C contents of materials and

targeting field-condition DOC concentrations (Kalbitz et al., 2003). Samples were shaken for 10 minutes, left in the refrigerator and stirred 3x in 24 hours to allow for an equilibrium to be established between the solid and dissolved phase (Kalbitz et al., 2003). Solutions were filtered through combusted GF/A and then through pre-rinsed 0.45 μm cellulose acetate syringe filters and analyzed for DOC concentration and spectroscopic properties.

Litter and soil extract biodegradation

In this current study, we define biodegradable DOC (BDOC) as the difference in DOC concentration before and after incubation. Accordingly, BDOC includes *i.* DOC respired as CO_2 and *ii.* DOC incorporated into microbial biomass. It is recognized that only monitoring change in DOC concentrations can overestimate total BDOC due to formation of particulate and colloidal C.

Inoculum was prepared according to methods of Fellman et al., 2008 in which 10 g of each litter and soil type was gently shaken in a 1:5 w:v ratio for 10 minutes with 0.01 M K_2SO_4, diluted 1:1 and filtered. 10 ml of inoculum was added to the corresponding litter or soil extract to ensure similar microbial communities exist in biodegradation experiment as in field conditions. Extracts with inoculum were left in the dark at 25°C for 28 days with loosely placed caps to prevent evaporation. Incubation vessels were gently stirred every three days. DOC concentration measurements and all spectroscopic analyses were performed again after incubation.

Lysimeter collections

In each of the 18 treatment plots, three Prenart Superquartz tension lysimeters were installed at 30 cm depths in 1997. During a period of high rainfall and soil saturation in December 2014, 15 kbar of tension was placed on each apparatus and left in the field to collect soil solution for 24-48 hours. Samples collected, returned to the lab, and refrigerated until analysis.

Analytical measurements

UV-vis and fluorescence spectroscopy were used for C characterization of lysimeter solution DOC and extractable DOC. The methods are rapid, relatively inexpensive and commonly used to analyze aquatic, marine and terrestrially-sourced DOC. Absorbance measurements were made over wavelengths 240-560 nm using a Cary 300 Bio UV-vis spectrophotometer with a 1 cm quartz cuvette rinsed three times with MilliQ water and once with sample prior to measurements. Samples with absorbance greater than 0.2 cm^{-1} were diluted prior to fluorescence measurements to minimize inner filter effects. Specific ultraviolet (UV) absorbance at 254 nm (SUVA) is calculated as the UV absorbance at 254 nm normalized to sample DOC concentration and has been strongly correlated with aromaticity of DOC (Weishaar et al., 2003).

3-D fluorescence scans were run with a Fluorolog® spectrofluorometer (HORIBA Jobin Yvon, Inc.). Emission wavelengths and intensities are recorded during a scan of excitation wavelengths to create 3-D excitation-emission matrices (EEMs). Samples were placed in a 1 cm quartz cuvette and scans were run over an excitation range of 250-400 nm with 10 nm increments and an emission range of 350-550 nm with 2 nm increments. Subtraction of blank MilliQ water or K_2SO_4 solution spectra from sample spectra

removed Raman scatter. Corrections also account for instrument-specific adjustments, first and second order Rayleigh scatter, and inner filter effects.

PARAFAC and fluorescence optical indices

The parallel factor analysis (PARAFAC) model developed by Cory and McKnight (2005) was used to unpack bulk EEMs into different components with unique excitation and emission spectra (Cory and McKnight, 2005). A component may be a single fluorophore or a group of similar fluorophores and therefore the knowledge of the chemical identity of each component varies by constituent (Table 3). The analysis resolves EEMs without making assumptions on the shapes of the spectra and a well-fit model explains greater than 99% of the fluorescence variation and errors should be less than 10%. The Cory and McKnight 13-component PARAFAC model was fit to collected fluorescence data (2005) and explained greater than 99% of DIRT soil DOC fluorescence and errors were less than 10%.

Ecologically relevant information is extracted by calculating a variety of indices including a Redox Index (RI) (Cory and McKnight, 2005), Fluorescence Index (FI) (McKnight et al., 2001, modified by Cory et al., 2010b) and Freshness Index (BIX) (Wilson and Xenopulos, 2009) (Table 4). McKnight et al. (2001) developed FI to gain information about the source of DOC using end members including Antarctic lakes with no nearby terrestrial inputs and catchments in the continental U.S. with abundant terrestrial inputs from partially broken down lignin and other terrestrial biomass. RI was correlated using shifts in PARAFAC component loadings across an oxycline in an Antarctic lake and during ice cover in a subalpine lake in CO. RI is calculated as the ratio

of reduced quinone-like fluorescence to oxidized quinone-like fluorescence (Cory and McKnight 2005). BIX was originally developed by Parlanti et al. (2000) through observations of degradation of macro-algae in marine waters and later updated by Wilson and Xenopulos (2009).

Statistics

General linear mixed models were fit using the function 'lme' in the nlme package (Pinheiro et al. 2015) to DIRT soil extract data with soil depth fit as a repeated measure for each response variable. The best fit model was selected using BIC model selection criteria from four candidate models including a model without a correlation structure, a first-order autoregressive correlation structure, a compound symmetry correlation structure, and a general correlation structure. If appropriate, the assumption of equal variance among treatments was relaxed for model fitting. The most suitable models for each response variables had no correlation structures. After ensuring assumptions of normality and equal variance among depths and treatments were met, *F*-tests were performed on the hypotheses that the interaction coefficient and main effect coefficients did not differ from zero.

A one-way ANOVA was used to test for differences in DOC optical parameters among DIRT treatment lysimeter samples or litter type extracts prior to incubation. Data was log-transformed if appropriate to meet assumptions for statistical tests. Once the main effects were determined to be significant, Tukey's post hoc Honest Significant Difference (HSD) test was used for pairwise comparisons at with a family-wise error rate of $\alpha = 0.05$ for lysimeter, litter and soil extracts used for biodegradation study. Tukey

pairwise comparisons were performed using the package multcomp version 1.4-0 (Hothorn et al., 2008). Paired t-tests and 95% confidence intervals were constructed to test for differences in DOC optical parameters before and after incubation of litter and soil extracts. All statistical analyses were performed in R version 3.0.2 (R Core Team, 2013).

Because EEM PARAFAC components can be complex to visualize and detect variation, principal component analysis (PCA) was applied to the correlation matrices of components to reduced dimensions that account for as much of the variation in the data as possible. Standardized data was shifted from original coordinate system to alternative orthogonal axes and values of principal component scores are projected onto these axes. The contribution to total fluorescence of components SQ3, SQ1 and C10 was less than 3% and therefore these three components were removed prior to analysis. PCA was applied to PARAFAC components of shallow soil and litter extracts before and after biodegradation and lysimeter and soil extracts from DIRT treatments.

Results

Off-plot litter and soil extract optical properties

HQ and Q2 tended to make up the largest proportion of total fluorescence in lysimeters and litter and soil extracts (Figure 1). The percent of total fluorescence from HQ of all samples was between 10-28% and the range of Q2 is 10-26%. SQ3, SQ1 and C10 generally contributed less than 3% of fluorescence. The small proportion of SQ1 and SQ3 that represent reduced quinone-like fluorescence could be indicative of relatively

oxidized DOC. The average percent contribution of SQ2 was 4% across all samples, further suggestive of oxidized extractable DOC.

Needles and A-horizon extracts had significantly lower aromaticity (SUVA) than O-horizon and wood extracts (Table 5). Class 5 wood extracts had lower SUVA than O-horizon, Class 3 and Class 2 wood extracts. A-horizon and needle extracts had higher FI than O-horizon and wood extracts (Table 5). Needle extracts had the highest RI indices, suggesting DOC in these extracts has the most oxidized-like fluorescence. The proportion of reduced quinone-like fluorescence increased from Class 2, Class 3, Class 5, O-horizon and A-horizon extracts. Needle extracts had the highest proportion of protein-like fluorescence.

In the PCA of litter and soil extracts, PC1 explained 43.7% of variance and PC2 explained a further 24.3% of variance (Figure 2). The dominant variables in determining PC1 scores included Q1, C6 and C3 and variables largely determining PC2 scores were Q3, C1, Tyrosine, and Tryptophan. Extracts of the most decomposed and the least decomposed materials, A-horizon and needles, had more negative scores on the PC1 axis than all other groups. Extracts from the A-horizon are further distinguished from needle extracts by more positive PC2 scores, suggesting more influence of Q3, C1 and aromatic amino acid-like fluorescence in A-horizon DOC relative to needle-derived DOC. Class 2 wood extractable DOC was distinguished from all other groups with more negative PC2 scores.

DIRT treatment lysimeter and soil extract DOC chemistry

There were no significant differences in concentrations or optical parameters of DOC among DIRT treatments from lysimeter solutions or soil extracts. DOC concentrations in lysimeters ranged from 1.46 to 13.78 mg C L^{-1} with a mean of 5.34 mg C L^{-1} but significant differences did not exist among treatments ($P = 0.80$) (Table 6). While DOC concentrations in Control and Double Litter treatment lysimeter solutions are slightly higher than in exclusion treatments, the difference is not statistically significant. In soil extracts, DIRT treatments had no impact on the values of SUVA, RI, BIX, and protein-like fluorescence (Table 7).

PCA of lysimeter solution PARAFAC components showed that PC1 explained 68.2% of variance and PC2 explained 20.0% of variance (Figure 3). Tryptophan, Q1, Q3, and C6 had the highest influence on scores on PC1. SQ2, C1, and C3 had the highest loadings for PC2. Lysimeter DOC scores did not show any clustering by treatment on the reduced-dimension PC axes.

Because soil extracts may capture a different pool of DOC than lysimeter-collected DOC, we expected that the soil extract PCA may uncover underlying DOC chemistry patterns that were not visible in the lysimeter PCA. PC1 explained 43.3% of variance and PC2 explained 20.2% of additional variance in the analysis of soil extracts from DIRT treatment soils from all depths (Figure 3). C6, Q2 and C1 had the highest loadings in PC1 and Tryptophan, C3, HQ and Q3 were highest in PC2. Similar to lysimeter analysis, PCA scores for soil extracts did not appear to vary with DIRT treatments. Overall, PARAFAC components of lysimeter DOC and extractable DOC collected from DIRT treatment plots showed that fluorescence signatures did not differ among DIRT treatments.

DOC content, SUVA and RI did not vary among DIRT treatments but there were significant differences observed with soil depth (Figure 4). DOC content decreased rapidly from 0-10 cm and subsequently did not change significantly from 10-30 cm. SUVA in all treatments showed a distinct decrease in aromaticity from 10-20 cm relative to shallower and deeper soils and RI increased with soil depth, indicative of more reduced material in deep soils. The proportion of HQ was significantly larger in 20-30 cm soils compared to the overlying 10-20 cm and 0-10 cm soils. The proportion of Q2 had the opposite pattern with soil depth as HQ. Q2 had significantly lower proportions at 20-30 cm than overlying 10-20 cm and 0-10 cm soils. BIX, Q3, C6, and the proportion of protein-like fluorescence had no statistically significant differences with soil depth.

FI was the only optical index to have a significant interaction between litter manipulation treatment and soil depth, indicating that the difference in FI among soil depths varied among DIRT treatment ($P = 0.04$). FI tended to increase with soil depth in No Root, No Input and Control treatments (Figure 4). Double Wood and No Litter treatments had slight increases in FI from 0-10 cm to 10-20 cm and then decreased in the 20-30 cm interval. However, treatment replicates had different patterns with soil depth and therefore the FI may not be particularly meaningful for soil DOC characterization.

DOC amount and composition differed between lysimeter DOC and soil extract DOC. SUVA values of extractable DOC had between 4.5% and 9.0% lower aromaticity than lysimeter solution DOC as calculated by the Weishaar et al. (2003) equation ($P = 0.005$). Percent aromaticity ranged from 9-27% in lysimeters and 7-23% in soil extracts (Weishaar et al., 2003). Lysimeter FI ranged between 1.36 and 1.66 which is significantly lower than the FI of soil extracts that had a range of 1.49-1.74 ($P < 0.005$). The RI of

DOC from lysimeters and extractable DOC was relatively oxidized. However, a larger RI in lysimeters indicates slightly more reduced quinone-like fluorescing DOC than in soil extracts ($P = 0.01$). The mean proportion of protein-like fluorescence in lysimeters is significantly larger than in soil extracts ($P < 0.005$). BIX did not differ significantly between lysimeter and soil extracts ($P = 0.49$).

Litter and soil extract DOC biodegradation

DOC content decreased in all extracts after biodegradation (Table 5). BDOC in all soil and litter extracts ranged from 4-49% of initial DOC (Figure 5). Class 2 wood had the lowest percentage of BDOC with a range of 4-11%. Biodegradability did not seem to be related to initial extractable DOC optical properties. The highest correlation found between BDOC and indices or components was with Q2 ($R^2 = -0.62$).

All confidence intervals on the difference in means pre and post incubation include a difference of zero for RI and FI (Table 5). Although not statistically significant, the estimated mean change in SUVA suggested a slight enrichment in aromaticity during degradation of all wood decomposition classes (Figure 6). O-horizon extracts had the only DOC to show a significant increase in SUVA ($P = 0.04$). Protein-like fluorescence decreased during biodegradation in A-horizon, needles, and O-horizon extracts, but there was no statistically significant change in protein-like fluorescence in wood extracts of any decomposition class (Table 5).

PCA on PARAFAC components of soil and litter extracts before and after incubation showed biodegradation resulted in movement on the PC axes (Figure 2). Biodegradation of needles, A-horizon and O-horizon extracts increased the PC1 scores

and slightly increased the scores on PC2. Decomposition of wood extracts increased the PC2 scores of Class 3, but replicates of Class 2 and Class 5 extracts shifted in various directions on the PC axes as a result of biodegradation.

Discussion

Changes in DOC chemistry in DIRT treatments

Total SOC in H.J. Andrews DIRT treatments has changed as a result of litter manipulations (Lajtha et al., 2005). Because decomposition products of existing SOC contribute to the dissolved pool, we expected to find changes in the bulk SOC content reflected in the chemistry of DOC (Sanderman et al., 2008). We also expected to observe differences in DOC composition among DIRT treatments because fluorescence signatures of extractable DOC from different sources of detritus showed that extract chemistry varies by source. However, even after 17 years of litter additions and exclusions in DIRT treatments, DOC composition has not changed in either soil extracts or lysimeter solutions as assessed by UV-vis and fluorescence spectroscopic fingerprinting techniques.

Other research using litter manipulations over shorter durations has demonstrated similar results. Yano et al. (2005) used resin fractionation of O-horizon leachate from detrital addition treatments (Double Litter, Double Wood) to show that there were minimal changes in DOC composition 3-4 years after DIRT treatment establishment. Efforts to use UV-vis and fluorescence indices to measure differences in DOC composition with changes to fresh litter inputs have also seen no see changes in DOC

chemistry. Kalbitz et al. (2007) identified no differences in fluorescence indices in O-horizon DOC as a result of 6 years of litter manipulations in sandy to loamy Haplic Cambisols. In other research, aboveground litter additions resulted in an increase in DOC concentration in leachate from the O-horizon but resulted in no change in UV absorbance (Fröberg et al., 2005). Altering litter inputs does not have major effects on soil DOC composition.

Biotic and abiotic homogenization of detrital leachates

Given the lack of differences in DOC chemistry among DIRT treatments, we posit that both biotic and abiotic processes are responsible for regulating the chemistry of incoming DOC from fresh litter. Briefly summarized, leachate from aboveground litter or root biomass or decomposition products from microbial processing enter the dissolved pool and undergo biodegradation during hydrologic transport (Bengtson and Bengtsson, 2007; van Hees et al., 2005). Concurrently, inorganic-organic and organic-organic interactions can remove C from the dissolved pool either temporarily or to be retained in a long-term SOC store (Qualls, 2000). Breakdown of SOC within the profile continues to mobilize additional DOC that undergoes a similar combination of abiotic and biotic dynamic exchange reactions moving C between the solid, liquid and gaseous phases (Sanderman et al., 2008). DOC loss occurs through both respiration and export. A conceptual model similar to that proposed by Sanderman et al. (2008) coincides with the patterns observed in the soils studied here.

There is ample evidence that fresh litter leachate makes only minor contributions to the DOC pool in fine-textured soils, leading to further support of the abiotic and biotic

regulation hypothesis. δ^{13}C-labelled beech and ash litter was found to make up less than 1% of total DOC in a mineral soil (Scheibe and Gleixner, 2014). Similarly, labelled spruce litter contributed less than 15% of total DOC flux underneath the O-horizon in a Norway spruce forest (Fröberg et al., 2007). While litter influence on DOC chemistry has been monitored over periods less than 6 months, data presented here confirm these results are not an artifact of short-term observations. Decomposition, incorporation and transformations of DOC by soil biota and interaction of DOC with mineral surfaces generates a pool in the mineral soil with a signature unlike that of DOC initially mobilized from fresh litter.

Evidence of DOC cycling within a horizon or small area of soil supports the model of continuous C exchange. In this study, fluorescence signatures show that the chemical composition of bulk SOC in the O-horizon is different from that of the mineral soil below. Significantly higher SUVA in O-horizon extracts relative to A-horizon extracts could indicate that DOC source materials in the O-horizon have a higher proportion of potentially soluble aromatics. In addition, the higher sorptive capacity in the mineral A-horizon could preferentially remove the hydrophobic aromatic acids from solution and result in lower measured aromaticity in A-horizon DOC (Kramer et al., 2012). The spatially tight cycling of DOC has been demonstrated in other soils. In a podzolized soil in Sweden, DOM in an Oe layer had ^{14}C signatures showing that the dissolved pool was produced within the Oe and that incoming Oi-derived DOM could either be sorbed within the Oe or lost through rapid biodegradation (Froberg et al., 2003). Ongoing exchange occurs between potentially mobile SOC and the dissolved pool.

Biodegradation

Understanding the relative impacts of microbial processing and sorption in determining DOC chemistry are critical for models of belowground DOC cycling. The incubation experiment determined whether microbial biodegradation of extracts from different aboveground OM sources and O-horizon transformed the fluorescence signatures to be more similar to the DOC from the A-horizon.

The BDOC between 4-49% observed during the 28 day incubation is similar to that observed in other studies (Qualls and Haines, 1992). Kalbitz et al. (2003) found a higher percentage of BDOC in extracts from fresher plant litter but in our data, there was no statistically significant difference in BDOC among soil horizons, needle, Class 3 and Class 5 extracts. There were changes in optical parameters of DOC from litter and soil extracts as a result of biodegradation, but the changes did not transform the chemistry to a signature closer to that observed in the O-horizon or A-horizon extracts. If biodegradation is not responsible for homogenization of the incoming DOC, abiotic processes dominate. The principal processes controlling the chemistry of DOC in each soil horizon likely differ between the O- and A-horizons given the differences in relative abundances of mineral versus organic materials. Furthermore, the dominance of biotic transformations relative to selective sorption mechanisms is likely to vary with soil mineralogy (Scott and Rothstein, 2014).

DOC and DIRT treatments

Besides biotic and abiotic processes regulating DOC chemistry, alternate explanations for the lack of differences in DOC chemistry among DIRT treatments

include (i) shifts in DIRT soil microbial community composition as a result of changes to OM inputs and (ii) changes in composition occurring in non-fluorescing fractions of DOC.

Adaptations of soil biotic communities to OM input chemistry may explain why there are no changes in DOC composition in Double Wood treatments. A higher fungal:bacterial ratio observed in Double Wood soils associated with the breakdown of phenols could control the incoming DOC chemistry and result in no change in fluorescence signatures between Double Wood and Control soils (Brant et al., 2006). While changes in microbial community were demonstrated in Double Wood soils, Yarwood et al. (2013) found no differences in microbial biomass or community between Control and No Input treatments so shifts in community composition varies by DIRT treatment. Future research should address the capability of community composition to shape DOC chemistry.

Root exclusion treatments may have altered DOC chemistry in ways that are not detectable using fluorescence spectroscopy. Root-derived C inputs seem to be important for DOC production. Yano et al. (2005) found that extracts of recently harvested roots had the highest DOC content relative to wood and needle extracts. The major role of root litter in producing DOC has also been suggested in a tree girdling experiment in a Scots pine forest on Entic Haplocryods. Soil extractable DOC concentrations were reduced by 40% by diminishing the flow of recent photosynthate to tree roots and associated mycorrhizal fungi (Giesler et al., 2007). Because major decreases were seen in compounds such as citrate, trehalose and monosaccharides that may not be captured in fluorescence techniques employed here, changes in chemistry as a result of root exclusion

in No Root and No Input DIRT treatments may not be detectable with UV and fluorescence spectroscopy (Giesler et al., 2007).

Evidence of a "priming effect" has been documented in plots experiencing litter additions at multiple DIRT treatment sites (Lajtha et al., 2014a; Crow et al., 2009). Sulzman et al. (2005) found that the respiration in Double Litter treatments exceeded the expected increase given the amount of added OM, suggesting that added litter was stimulating microbial communities to breakdown older, in-situ SOC. Data has not been collected to confirm that priming has persisted in the Double Litter treatment since the original respiration measurements that occurred 6 years after treatment establishment (Crow et al., 2009). If priming has continued to occur in the Double Litter plots, we are not observing a simultaneous increase in DOC production as assessed by lysimeter-collected solutions and soil extracts. This result is contrary to other research that has found that adding easily-degradable material was associated with an increase in DOC production (Kalbitz et al., 2007; Park and Matzner, 2003).

Capability of spectroscopic analyses for soil DOC characterization

Besides monitoring the influence of litter inputs on DOC chemistry in DIRT treatments, another goal of this research was to determine whether optical parameters derived from UV-vis and fluorescence spectroscopy were able to provide meaningful information about DOC chemistry. Fluorescence coupled with PARAFAC was capable of differentiating unique signatures of DOC derived from different source materials.

Optical indices reflected differences among litter and soil extract chemistries that coincide with existing knowledge of source material chemistry. Needle extracts had a

higher proportion of protein-like fluorescence than soil and wood extracts although the difference was only statistically significantly larger relative to O-horizon, Class 5, and Class 2. Wood extracts had a high proportion of aromatics likely due to the high input of mobilized lignin decomposition products. The lack of mineral interaction with hydrophobic aromatics in the O-horizon may explain the high proportion of aromatics in O-horizon extracts relative to the A-horizon. While needle extracts were expected to release the least decomposed and most reduced DOC, RI of needle extracts showed the opposite. The A-horizon soil that likely contains the most processed SOC reflected the most reduced RI values.

Optical indices also showed differences among soil depths. In general, SUVA and $\delta^{13}C$ tend to decrease with soil depth with $\delta^{13}C$ decreasing due to additions of processed DOC or because constituents already in solution are becoming increasingly altered (Sanderman et al., 2008). A distinctive pattern was observed in DIRT treatment soils in which SUVA decreased from 10-20 cm relative to the soil above and the soil below. The high SUVA values in 0-10 cm could be capturing DOC leaving the O-horizon that is particularly high in aromatics. Alternatively, the decrease in aromaticity from 10-20 cm is indicating a change in mineralogy hence, a soil sorptivity shift that involves removal of aromatics from solution due to hydrophobic interactions. As expected, RI increased with soil depth, reflecting the more reducing conditions with increased soil depth. There were no distinct patterns of FI with soil depth and because the values of FI in the biodegradation experiment were also inconsistent, further adjustment of the FI calculation may be necessary before it can reliably be used in soil DOC characterization. Gabor et al. (2013) showed that a lower FI in 0-70 cm soils with higher plant inputs

corresponded to a higher, more reduced RI in soil extracts from the Colorado Front Range. Similar to our observations of a greater change in optical indices with soil depth than among litter manipulation treatments, Gabor et al. (2013) found that microbial communities and fluorescence indices varied more with soil depth than across landscape positions.

When patterns of each PARAFAC component with soil depth are observed individually, a few components show consistent trends but others have unsystematic variation such as tryptophan and tyrosine components. HQ, a component that corresponds to hydroquinone-like fluorescence, was relatively constant from 0-20 cm and a subsequent increase in the deeper 20-30 cm could signify a shift towards more reduced conditions. An oxidized quinone-like component, Q2, decreased from 20-30 cm and reflects the increasing degree of decomposition with soil depth. The increase in Q2 during incubation in all extracts except Class 2 and Class 3 suggests that the fluorophore may be a decomposition product from microbial decomposition. Q3 is thought to be representative of relatively oxidized quinone-like fluorescence and was found to be abundant at all soil depths with no statistically significant difference among depths. Cory and McKnight (2005) noted a relationship between the percent aliphatic C and proportion of Q3. The expected loss of relatively easily-degraded aliphatic C during biodegradation was confirmed by a decrease in Q3 during the incubation.

Evidence of the relatively oxidized conditions in H.J. Andrews DIRT soils were apparent in the components with reduced-like fluorescence contributing less than 5% of total fluorescence in most samples. HQ is a component thought to have fluorescence similar to quinones in intermediate redox states (Cory and McKnight, 2005). The slight

increase in HQ at 20-30 cm shows that fluorescence spectroscopy may be a powerful tool for observing soil DOC redox conditions.

Conclusions

C movement and accumulation in soils is unequivocally linked to the chemistry and processes controlling DOC. The DIRT network provides a unique opportunity to study DOC response to long term changes in needles, wood and root inputs. Although there have been changes in SOC among the H.J. Andrews Forest DIRT site (Lajtha et al., 2005), DOC concentrations and chemistry have not responded to 17 years of litter manipulations. A biotic and abiotic driven homogenization model explains the lack of differences among treatments. Confirming that the chemistry of DOC was strongly dependent on the precursor material, PCA on PARAFAC components of 3-D EEMs successfully distinguished among the chemistry of extractable DOC from O-horizon, A-horizon, Douglas-fir needles, and wood of multiple decomposition classes. Even with long-term change to aboveground and belowground detrital inputs to soils, fluorescence signatures of soil extract and lysimeter DOC did not vary among DIRT treatments. Biodegradation of soil and litter extracts resulted in a loss in DOC and a change in optical signatures, but did not result in signatures more similar to the mineral soil, signifying the importance of abiotic processing in DOC fractionation. Extractable DOC in the O-horizon had a fluorescence signature that was distinct from that of the 0-5 cm A-horizon, suggesting spatially-tight cycling of DOC. Results from this work support a model of DOC dynamics in which litter leachate is only a small contributor and complex

interactions between existing SOC and the dissolved phase determine the composition of DOC.

This work also demonstrates the ability of UV-vis and fluorescence spectroscopy to characterize soil DOC and changes in composition during incubation. SUVA and RI varied significantly with soil depth with an increase in RI with depth showing the increasingly reduced character of DOC. Optical indices including BIX and FI did not change as a result of incubation or with soil depth, suggesting that additional adjustments may be needed prior to application of these indices to soils. Lastly, observing trends in PARAFAC components with soil depth and with the biodegradation incubation provided some insight into the biochemical identity of various components.

Future Research

DIRT treatment DOC characterization should take into account shifts in microbial communities induced by changes to detrital inputs and seasonal variation. Significant emphasis should be placed on accounting for the high variability in H.J. Andrews soils (Dixon, 2003). With increased replication and separating soils into depth increments smaller than the 10 cm used in this work, research is likely to illuminate further trends in chemistry with soil depth that are measurable with optical spectroscopy. Also, the No Input and No Root treatments could provide a unique opportunity to study the effect of long-term exclusion of root biomass and rhizosphere impacts on DOC chemistry.

The relative impact of microbial community and inorganic-organic and organic-organic interactions in DOC homogenization in the first few centimeters of a soil profile are not well constrained. Research has explored DOC chemistry within the O-horizon of

other soils (i.e. Fröberg et al., 2003; Kalbitz et al., 2007), but further separation of the shallow mineral soil into small increments is necessary to characterize the interactions between detritus, O-horizon and the mineral soil.

We have not yet fully explored the capability of UV-vis and fluorescence spectroscopy as a fingerprinting technique for soil DOC. While SUVA and RI have provided meaningful information about DOC composition, care should be taken when applying previously-correlated optical indices into diverse ecological systems. A number of recent studies have shown the application of fluorescence methods in soils. For example, Banaitis et al. (2006) effectively used PARAFAC to characterize DOC fractionation as a result of sorption. Future research should monitor the response of Cory and McKnight (2005) PARAFAC components in soils experiencing redox extremes, changes in fluorescence over significantly longer incubations, and fractionation due to sorption. PARAFAC models of fluorescence EEMs could also benefit from a greater understanding of the biochemistry of particular components.

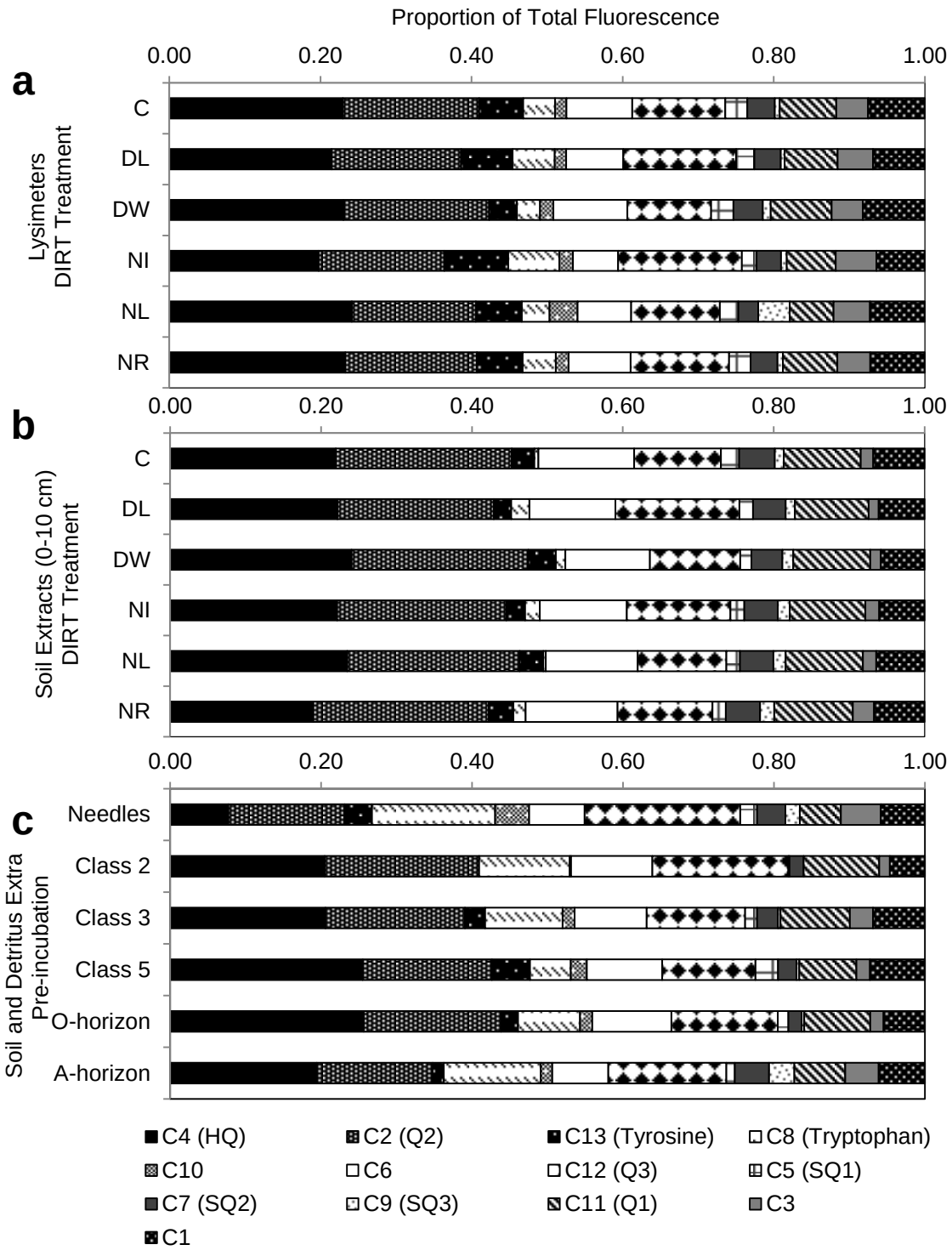

Figure 1. PARAFAC components as a proportion of total fluorescence. (a) Lysimeter-collected (30 cm) soil solution DOC from DIRT treatments, (b) DOC extracted from soils in DIRT treatments sampled from 0-10 cm, (c) DOC extracted from off-plot soils and detritus.

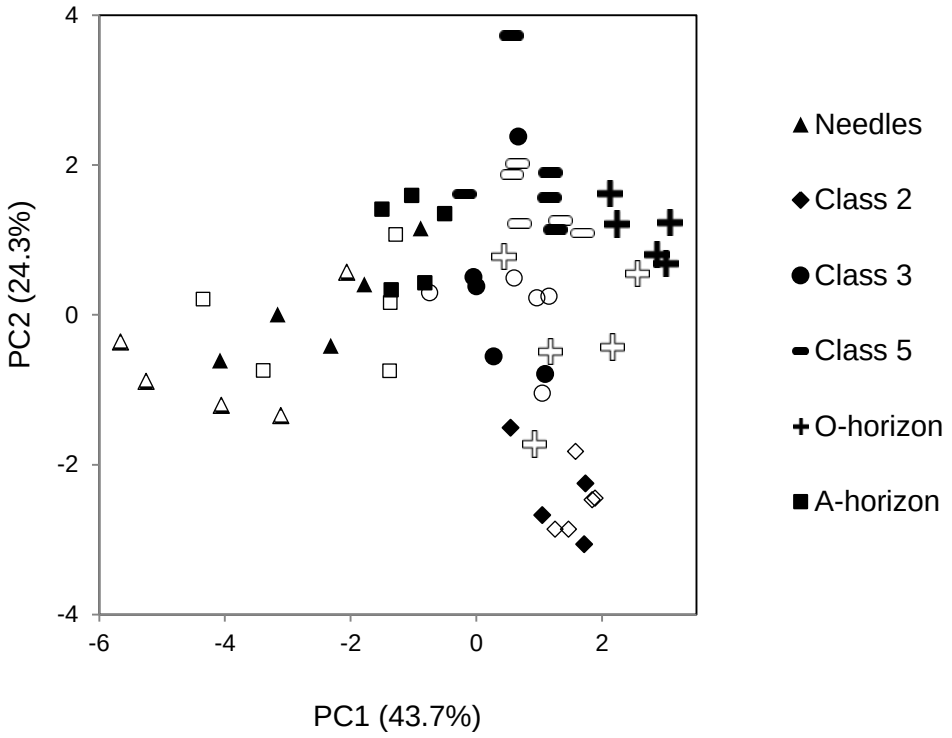

Figure 2. Litter and soil extract PCA. Scores from pre and post biodegradation PARAFAC components. Pre biodegradation scores are shown with hollow shapes. Class 2 is least decomposed, Class 5 is most decomposed wood, and A-horizon is 0-5 cm mineral soil.

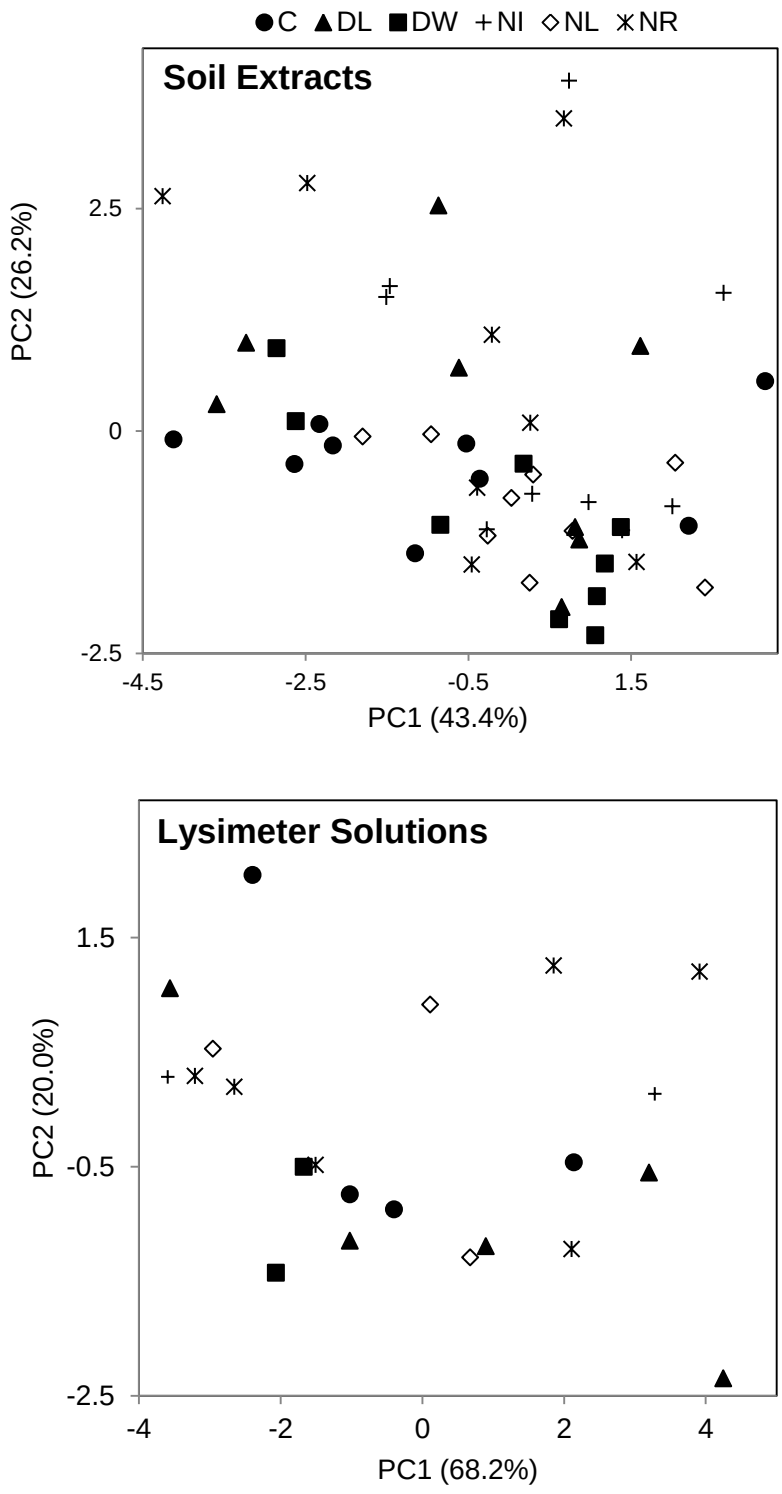

Figure 3. DIRT soil extracts and lysimeter solution PCA. PCA on PARAFAC components from soil extracts from all soil depths (top) and lysimeter solutions (bottom). C = Control, DL = Double Litter, DW = Double Wood, NI = No Inputs, NL = No Litter, NR = No Roots.

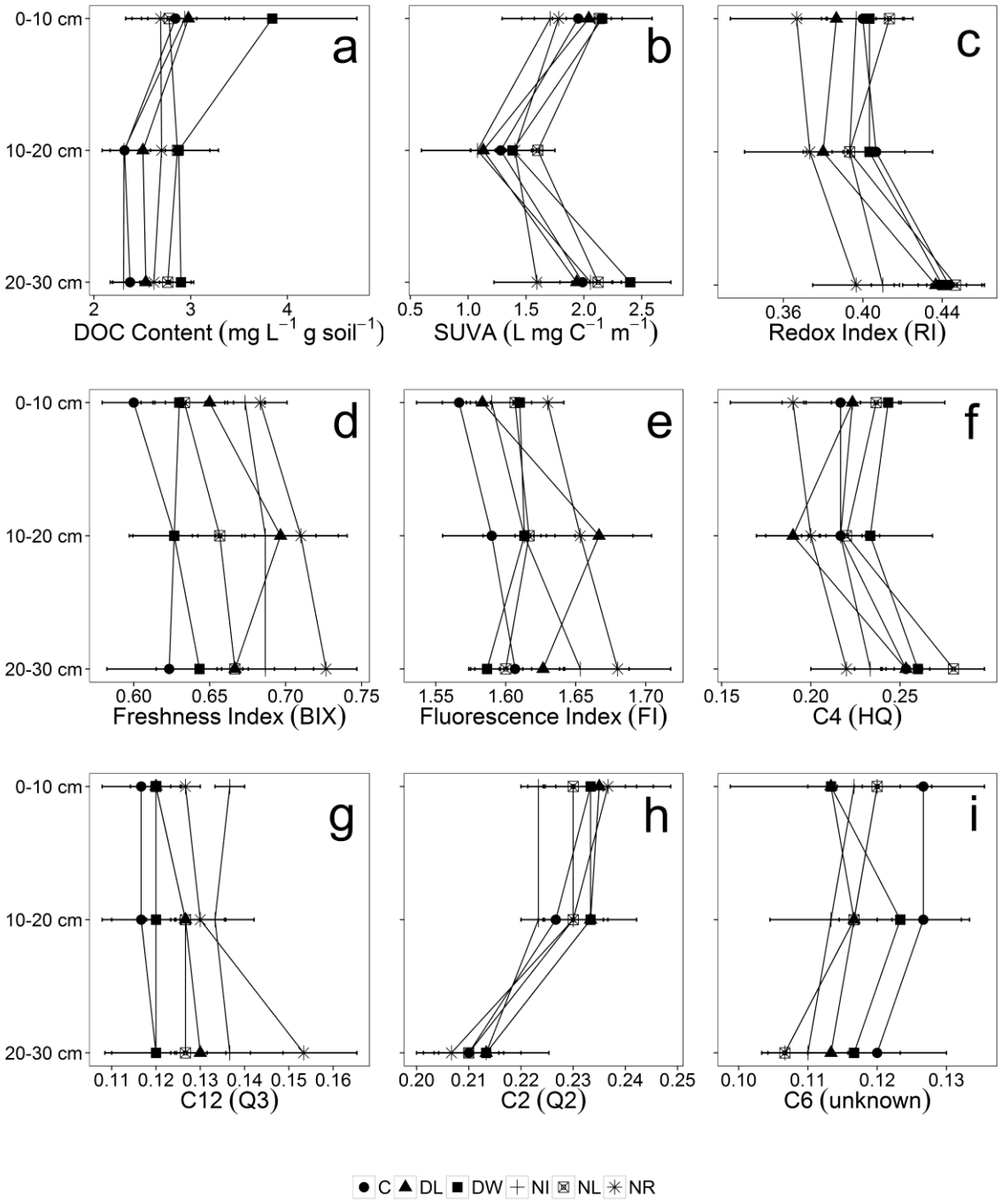

Figure 4. Differences in optical parameters of DIRT soil extracts with soil depth. Soils were sampled in 10 cm increments. Error bars represent SE. Soils were sampled in March 2014. (a) DOC content, (b) Specific UV absorbance at 254 nm (SUVA) (c) Redox Index (RI), (d) Freshness Index (BIX), and (e) Fluorescence Index (FI). The four most abundant PARAFAC components include: (f) C4 (HQ), (g) C12 (Q3), (h) C2 (Q2), (i) C6 (unknown). See Table 3 for descriptions of PARAFAC components.

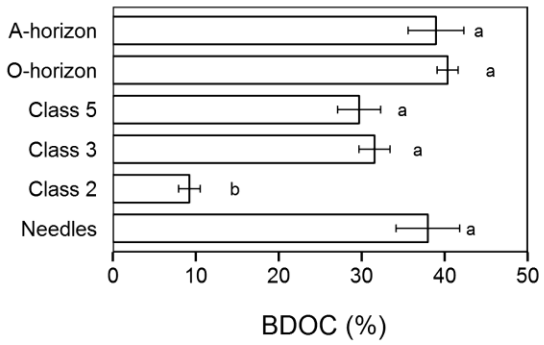

Figure 5. BDOC during 28-day incubation of litter and soil extracts. BDOC did not differ significantly among bars with the same letter (Tukey's HSD).

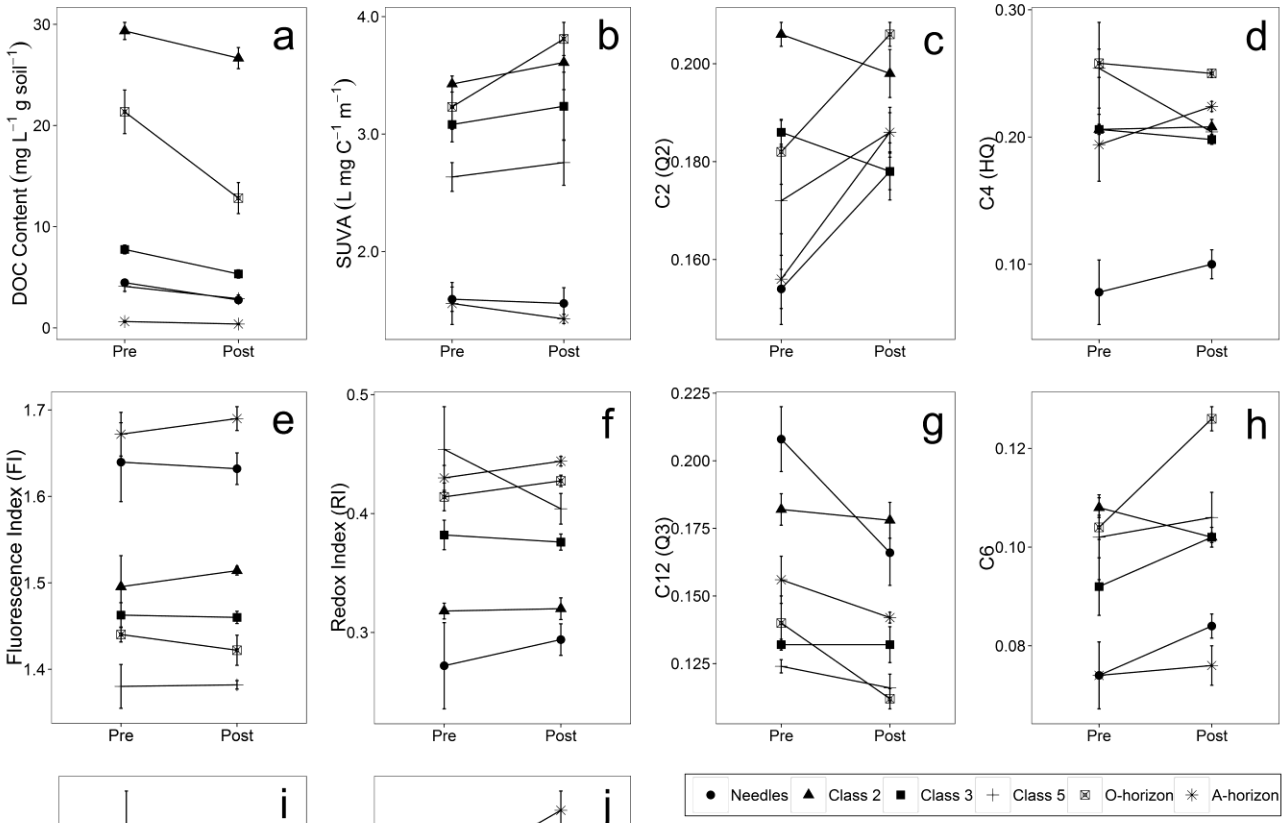

Figure 6. Incubation change in optical parameters. Changes in DOC content, optical parameter and four most abundant PARAFAC components of detritus and soil extract DOC as a result of biodegradation. Error bars represent SE. (a) DOC content, (b) Specific UV absorbance at 254 nm (SUVA) (c) Fluorescence Index (FI), (d) Redox Index (RI), (e) Proportion protein-like fluorescence, and (f) Freshness Index (BIX). The four most abundant PARAFAC components include: (g) C2 (Q2), (h) C4 (HQ), (i) C12 (Q3), (j) C6 (unknown

Table 1. DIRT treatment descriptions of plots established in H.J. Andrews in 1997. Three plots were randomly assigned to each of the six treatments.

DIRT Treatment	Description
C	Control; No change to aboveground or belowground detrital inputs
DN	Double Needle; Fallen needles onto exclusion plots are added annually to DN plots
DW	Double Wood; Woodchips are added biannually to approximate the amount of C added to DN plots
NL	No Litter; Litterfall consisting primarily of Doug-fir needles are excluded through screening
NR	No Roots; Roots are excluded through living plant removal and lateral ingrowth is prevented through trenching
NI	No Inputs; Combination of NL and NR treatments

Table 2. Description of litter and soils collected from areas nearby DIRT treatments for extraction and biodegradation experiment.

Litter Type	Description
Needles	Needles; Fallen within 10 months of collection; primarily Doug-fir
Class 2	Woodchips that have undergone minimal decomposition; no bark
Class 3	Moderately decomposed wood; sloughed bark, structure maintained
Class 5	Highly decomposed; structure not intact; heartwood and sapwood decomposed
O-horizon	O-horizon soils
A-horizon	A-horizon soils (0-5 cm)

Table 3. 13 Cory and McKnight (2005) PARAFAC components with associated excitation maxima, minima and interpretations from various sources. Component identities from Cory and McKnight (2005) unless otherwise noted. Q represents quinone-like fluorescence, SQ is semiquinone-like fluorescence, and HQ is hydroquinone-like fluorescence. C11 is component 11 in Cory and McKnight (2005) PARAFAC model.

Cory & McKnight (2005) Component	Excitation minima (nm)	Emission maxima (nm)	Description
Q1 (C11)	260	540	Oxidized quinone-like, terrestrial humic-like; photo-refractory (Nishimura et al., 2012)
Q2 (C2)	250	510	Oxidized quinone-like, terrestrial humic-like; photo-refractory (Stedmon et al., 2007)
Q3 (C12)	250	390	Oxidized; Correlated with percent aliphatic C; Higher aliphatic fraction has been correlated with microbial precursor material (Cory and McKnight 2005); Alternatively, relationship between aliphatic and Q3 could be due to quenching by nonaliphatic C
SQ1 (C5)	290	515	Terrestrial humic-like, humic acid-type; very sensitive to microbial and photochemical degradation (Nishimura et al., 2012); related to aromatic DOC; reduced semiquinone
SQ2 (C7)	270 (380)	460	SQ2 has blue-shifted spectra relative to SQ1, suggesting SQ2 has a more conjugated system relative to SQ1; reduced semiquinone
SQ3 (C9)	345 (265)	410	Reduced semiquinone
HQ (C4)	265	550	Correlated with percent ketone/aldehyde C; hydroquinone
Tryptophan-like (C8)	270	360	Indicative of amino acid tryptophan; proteins or peptides that could be free amino acids or bound; could be less-degraded peptide material (Fellman et al., 2010)
Tyrosine-like (C13)	280	350	Indicative of amino acid tyrosine; proteins or peptides that could be free amino acids or bound; could be more-degraded peptide material (Fellman et al., 2010)
C3	315	385	Indicates DOC of microbial or planktonic origin; listed as unknown component type in Cory & McKnight (2005)

Table 3. *Continued*

C6	265-270	430	Listed as unknown component type in Cory & McKnight (2005)
C10	305	425	Terrestrial humic-like, fulvic acid-type (Nishimura et al. 2012); listed as unknown component type in Cory & McKnight (2005)
C1	335-340	450	May be a quinone derivative (a ketal) formed by reaction of a quinone with an alcohol; Correlated with percent anomeric, acetal, ketal C; Ubiquitous humic-like, associated with increased FI (Nishimura et al., 2012); Listed as unknown component type in Cory & McKnight (2005)

Table 4. Absorbance and fluorescence DOC optical parameter calculations and interpretation.

DOC Optical Parameter	Reference	Interpretation
Specific UV Absorbance (SUVA) (L mg C^{-1} m^{-1})	Weishaar et al. (2003)	UV absorbance of light at 254 nm normalized to sample DOC concentration. Values are strongly correlated with aromaticity of DOC.
Fluorescence Index (FI)	McKnight et al. (2001); Cory et al. (2010)	Ratio of emission intensities of 470 nm / 520 nm at excitation 370 nm.
Freshness Index (BIX)	Parlanti et al. (2000); Wilson & Xenopulos (2009)	Ratio of emission intensities of 380 nm / max between 420-435 nm at excitation 310 nm. Indicates ratio of recently produced DOC to older, more decomposed DOC.
Redox Index (RI)	Cory & McKnight (2005); Miller et al. (2006)	$\sum$ loadings reduced quinones (SQ1 + SQ2 + SQ3 + HQ) / $\sum$ loadings oxidized quinones (Q11 + Q2 + Q3). Index is an indicator of oxidation state of DOC.
Proportion protein-like fluorescence	Cory & McKnight (2005); Fellman et al. (2008); Fellman et al. (2009)	$\sum$ Tyrosine (C13) + Tryptophan (C8) | Amino acids, suggestive of protein-like fluorophores in DOM.

Table 5. Mean and (SE) of DOC content and optical parameters of litter and soil extract DOC from fluorescence measurements. Pre and post columns represent measurements before and after 28-day incubation. Means with the same letter within each column are not significantly different (Tukey's honest significant difference). Asterisk after letter significance in post data columns represents significant difference between pre- and post-incubation data. One asterisk (*) indicates $P \leq 0.05$, two asterisks (**) indicate $P \leq 0.005$. No asterisk indicates an insignificant difference between pre- and post-incubation ($P > 0.05$). n = 5 for all values.

OM Extracted	DOC Content (mg C L^{-1} g $soil^{-1}$)		SUVA (L mg C^{-1} m^{-1})		Redox Index (RI)	
	Pre	*Post*	*Pre*	*Post*	*Pre*	*Post*
Needles	4.47 (0.20)^{c}	2.75 (0.14)^{d,**}	1.59 (0.10)^{c}	1.56 (0.13)^{c}	0.27 (0.04)^{c}	0.29 (0.01)^{d}
Class 2	29.34 (0.85)^{a}	26.66 (1.04)^{a,**}	3.43 (0.07)^{a}	3.61 (0.23)^{a}	0.32 (0.01)^{b}	0.35 (0.02)^{c}
Class 3	7.75 (0.44)^{b}	5.33 (0.41)^{c,**}	3.08 (0.13)^{ab}	3.24 (0.29)^{ab}	0.38 (0.01)^{a}	0.37 (0.01)^{b}
Class 5	4.10 (0.49)^{c}	2.88 (0.38)^{d,**}	2.63 (0.12)^{b}	2.76 (0.19)^{b}	0.42 (0.01)^{a}	0.38 (0.02)^{b}
O-horizon	21.35 (2.16)^{a}	12.83 (1.53)^{b,**}	3.23 (0.19)^{ab}	3.81 (0.14)^{a,*}	0.41 (0.01)^{a}	0.42 (0.004)^{a}
A-horizon	0.62 (0.09)^{d}	0.38 (0.05)^{e,**}	1.56 (0.18)^{c}	1.43 (0.04)^{c}	0.43 (0.01)^{a}	0.45 (0.004)^{a}

Table 5. *Continued*

OM Extracted	Protein		Freshness Index (BIX)		Fluorescence Index (FI)	
	Pre	*Post*	*Pre*	*Post*	*Pre*	*Post*
Needles	0.20 (0.02)^{a}	0.16 (0.01)^{a,*}	0.76 (0.04)^{ab}	0.71 (0.03)^{b,*}	1.64 (0.05)^{a}	1.63 (0.02)^{b}
Class 2	0.12 (0.01)^{b}	0.12 (0.01)^{ab}	0.67 (0.01)^{bc}	0.68 (0.01)^{bc}	1.50 (0.04)^{b}	1.51 (0.004)^{c}
Class 3	0.13 (0.002)^{ab}	0.11 (0.01)^{b}	0.62 (0.01)^{cd}	0.65 (0.01)^{cd,*}	1.46 (0.01)^{b}	1.46 (0.01)^{cd}
Class 5	0.10 (0.02)^{b}	0.10 (0.01)^{c}	0.55 (0.01)^{d}	0.61 (0.01)^{d,*}	1.38 (0.03)^{b}	1.38 (0.004)^{e}
O-horizon	0.10 (0.01)^{b}	0.07 (0.003)^{c,*}	0.58 (0.02)^{d}	0.55 (0.01)^{e}	1.44 (0.01)^{b}	1.42 (0.02)^{de}
A-horizon	0.14 (0.03)^{ab}	0.07 (0.01)^{c,*}	0.78 (0.03)^{a}	0.85 (0.02)^{a,*}	1.67 (0.02)^{a}	1.69 (0.01)^{a}

Table 6. Mean DOC optical parameters of lysimeter-collected soil solutions in December 2014 from DIRT treatments and (SE). Lysimeters are installed at 30 cm depth. C = Control, DL = Double Litter, DW = Double Wood, NI = No Inputs, NL = No Litter, NR = No Roots. Analyses of variance demonstrated no significant differences in response variables among treatments.

Treatment	Count	DOC (mg C L^{-1})	SUVA (L mg C^{-1} m^{-1})	Redox Index (RI)	Protein	Freshness Index (BIX)	Fluorescence Index (FI)
C	4	6.55 (2.11)	1.95 (0.36)	0.44 (0.01)	0.10 (0.02)	0.67 (0.05)	1.44 (0.03)
DL	6	6.31 (1.72)	1.95 (0.30)	0.41 (0.02)	0.12 (0.02)	0.71 (0.05)	1.50 (0.03)
DW	2	6.79 (1.38)	2.82 (0.23)	0.45 (0.02)	0.07 (0.01)	0.62 (0.005)	1.47 (0.02)
NI	3	3.99 (3.38)	2.23 (0.72)	0.39 (0.05)	0.15 (0.06)	0.64 (0.07)	1.49 (0.06)
NL	4	4.09 (1.46)	2.44 (0.56)	0.50 (0.06)	0.10 (0.01)	0.61 (0.03)	1.50 (0.06)
NR	6	4.60 (1.24)	1.95 (0.43)	0.45 (0.01)	0.10 (0.02)	0.74 (0.08)	1.48 (0.03)

Table 7. Mean DOC optical parameters of soil extracts within DIRT treatments and (SE). Given an insignificant interaction between treatment and depth, the mean of each treatment represents the response variable averaged across three sampled soil depths. C = Control, DL = Double Litter, DW = Double Wood, NI = No Inputs, NL = No Litter, NR = No Roots.

Treatment	Count	DOC (mg C L^{-1} g^{-1} soil)	SUVA (L mg C^{-1} m^{-1})	Redox Index (RI)	Protein	Freshness Index (BIX)	Fluorescence Index (FI)
C	9	2.51 (0.19)	1.74 (0.16)	0.41 (0.01)	0.04 (0.002)	0.62 (0.02)	1.59 (0.01)
DL	9	2.67 (0.16)	1.70 (0.18)	0.40 (0.01)	0.04 (0.003)	0.67 (0.02)	1.63 (0.02)
DW	9	3.21 (0.32)	1.98 (0.25)	0.42 (0.01)	0.04 (0.01)	0.63 (0.01)	1.60 (0.01)
NI	9	2.52 (0.22)	1.62 (0.25)	0.40 (0.01)	0.04 (0.003)	0.68 (0.02)	1.62 (0.01)
NL	9	2.80 (0.15)	1.95 (0.10)	0.42 (0.01)	0.04 (0.003)	0.65 (0.01)	1.61 (0.02)
NR	9	2.67 (0.10)	1.59 (0.16)	0.38 (0.02)	0.05 (0.003)	0.71 (0.01)	1.65 (0.02)

References

Banaitis, M.R., Waldrip-Dail, H., Diehl, M.S., Holmes, B.C., Hunt, J.F., Lynch, R.P., and Ohno, T. 2006. Investigating sorption-driven dissolved organic matter fractionation by multidimensional fluorescence spectroscopy and PARAFAC. Journal of Colloid and Interface Science 304, 271–6.

Bengtson, P., and Bengtsson, G. 2007. Rapid turnover of DOC in temperate forests accounts for increased CO_2 production at elevated temperatures. Ecology Letters 10, 783–790.

Berg, B., Hannus, K., Popoff, T., and Theander, O. 1982. Changes in organic chemical components of needle litter during decomposition. Long-term decomposition in a Scots pine forest. I. Canadian Journal of Botany 60, 1310–1319.

Brant, J.B., Sulzman, E.W., and Myrold, D.D. 2006. Microbial community utilization of added carbon substrates in response to long-term carbon input manipulation. Soil Biology and Biochemistry 38, 2219–2232.

Cole, J.J., Prairie, Y.T., Caraco, N.F., McDowell, W.H., Tranvik, L.J., Striegl, R.G., Duarte, C.M., Kortelainen, P., Downing, J.A., Middelburg, J. J., and Melack, J. 2007. Plumbing the global carbon cycle: Integrating inland waters into the terrestrial carbon budget. Ecosystems 10, 172–185.

Cory, R.M., and Mcknight, D.M. 2005. Fluorescence Spectroscopy Reveals Ubiquitous Presence of Oxidized and Reduced Quinones in Dissolved Organic Matter. Environmental Science and Technology *39*, 8142–8149.

Cory, R.M., Miller, M.P., Mcknight, D.M., Guerard, J.J., and Miller, P.L. 2010. Effect of instrument-specific response on the analysis of fulvic acid fluorescence spectra. Limnology and Oceanography: Methods 67–78.

Crow, S.E., Lajtha, K., Bowden, R.D., Yano, Y., Brant, J.B., Caldwell, B.A., and Sulzman, E.W. 2009. Increased coniferous needle inputs accelerate decomposition of soil carbon in an old-growth forest. Forest Ecology and Management 258, 2224–2232.

Deb, S.K., and Shukla, M.K. 2011. A review of dissolved organic matter transport processes affecting soil and environmental quality. Journal of Environmental & Analytical Toxicology 1, 1–11.

Dixon, J. 2003. Applying GIS to soil-geomorphic landscape mapping in the Lookout Creek Valley, Western Cascades, Oregon. M.S. Thesis. Oregon State University, Corvallis, OR.

Fellman, J.B., D'Amore, D.V., Hood, E., and Boone, R.D. 2008. Fluorescence characteristics and biodegradability of dissolved organic matter in forest and wetland soils from coastal temperate watersheds in southeast Alaska. Biogeochemistry 88, 169–184.

Fellman, J.B., Hood, E., and Spencer, G M. 2010. Fluorescence spectroscopy opens new windows into dissolved organic matter dynamics in freshwater ecosystems: A review. Limnology and Oceanography 55, 2452–2462.

Fröberg, M., Berggren, D., Bergkvist, B., Bryant, C., and Knicker, H. 2003. Contributions of Oi, Oe and Oa horizons to dissolved organic matter in forest floor leachates. Geoderma 113, 311–322.

Fröberg, M., Kleja, D.B., Bergkvist, B., Tipping, E., and Mulder, J. 2005. Dissolved organic carbon leaching from a coniferous forest floor – a field manipulation experiment. Biogeochemistry 75, 271–287.

Fröberg, M., Kleja, D.B., and Hagedorn, F. 2007. The contribution of fresh litter to dissolved organic carbon leached from a coniferous forest floor. European Journal of Soil Science 58, 108–114.

Gabor, R.S., Eilers, K., McKnight, D.M., Fierer, N., and Anderson, S.P. 2013. From the litter layer to the saprolite: Chemical changes in water-soluble soil organic matter and their correlation to microbial community composition. Soil Biology and Biochemistry October, 1–11.

Gielen, B., Neirynck, J., Luyssaert, S., and Janssens, I.A. 2011. The importance of dissolved organic carbon fluxes for the carbon balance of a temperate Scots pine forest. Agricultural and Forest Meteorology, 151, 270–278.

Giesler, R., Högberg, M.N., Strobel, B.W., Richter, A., Nordgren, A., and Högberg, P. 2007. Production of dissolved organic carbon and low-molecular weight organic acids in soil solution driven by recent tree photosynthate. Biogeochemistry 84, 1–12.

Hothorn, T., Bretz, F., and Westfall, P. 2008. Simultaneous inference in general parametric models. Biometrical Journal 50, 346--363.

Kalbitz, K., and Kaiser, K. 2007. Contribution of dissolved organic matter to carbon storage in forest mineral soils. Journal of Plant Nutrition and Soil Science 171, 52–60.

Kalbitz, K., Meyer, A., Yang, R., and Gerstberger, P. 2007. Response of dissolved organic matter in the forest floor to long-term manipulation of litter and throughfall inputs. Biogeochemistry 86, 301–318.

Kalbitz, K., Schmerwitz, J., Schwesig, D., and Matzner, E. 2003. Biodegradation of soil-derived dissolved organic matter as related to its properties. Geoderma 113, 273–291.

Kalbitz, K., Schwesig, D., Rethemeyer, J., and Matzner, E. 2005. Stabilization of dissolved organic matter by sorption to the mineral soil. Soil Biology and Biochemistry 37, 1319–1331.

Kindler, R., Siemens, J., Kaiser,K., Walmsley, D.C., Bernhofer, C., Buchmann, N., Cellier, P., Eugster, W., Gleixner, G., Grünwald, T., Heim, A., Ibrom, A., Jones, S.K., Jones, M., Klumpp, K., Kutsch, W., Steenberg Larsen, K., Lehuger, S., Loubet, B., McKenzie, R., Moors, E., Osborne, B., Pilegaard, K., Rebmann, C., Saunders, M., Schmidt, M.W.I., Schrumpf, M., Seyfferth, J., Skiba, U., Soussana, J.-F., Sutton, M.A., Tefs, C., Vowinckel, B., Zeeman, M.J., Kaupenjohann, M. 2011. Dissolved carbon leaching from soil is a crucial component of the net ecosystem carbon balance. Global Change Biology 17, 1167–1185.

Kramer, M.G., Sanderman, J., Chadwick, O.A., Chorover, J., and Vitousek, P.M. 2012. Long-term carbon storage through retention of dissolved aromatic acids by reactive particles in soil. Global Change Biology 18, 2594–2605.

Lajtha, K., Crow, S.E., Yano, Y., Kaushal, S.S., Sulzman, E., Sollins, P., and Spears, J.D.H. 2005. Detrital controls on soil solution N and dissolved organic matter in soils: a field experiment. Biogeochemistry 76, 261–281.

Lajtha, K., Townsend, K.L., Kramer, M.G., Swanston, C., Bowden, R.D., and Nadelhoffer, K. 2014a. Changes to particulate versus mineral-associated soil carbon after 50 years of litter manipulation in forest and prairie experimental ecosystems. Biogeochemistry 119, 341-360.

McKnight, D.M., Boyer, E.W., Westerhoff, P.K., Doran, P.T., Kulbe, T., and Andersen, D.T. 2001. Spectrofluorometric characterization of dissolved organic matter for indication of precursor organic material and aromaticity. Limnology and Oceanography 46, 38–48.

Nishimura, S., Maie, N., Baba, M., Sudo, T., Sugiura, T., and Shima, E. 2012. Changes in the quality of chromophoric dissolved organic matter leached from senescent leaf litter during the early decomposition. Journal of Environmental Quality 41, 823–33.

Park, J.H. and Matzner, E. 2003. Controls on the release of dissolved organic carbon and nitrogen from a deciduous forest floor investigated by manipulations of aboveground litter inputs and water flux. Biogeochemistry 66, 265–286.

Parlanti, E., Wo, K., Geo, L., and Lamotte, M. 2000. Dissolved organic matter fluorescence spectroscopy as a tool to estimate biological activity in a coastal zone submitted to anthropogenic inputs. *31*, 1765–1781.

Qualls, R.G. 2000. Comparison of the behavior of soluble organic and inorganic nutrients in forest soils. Forest Ecology and Management 138, 29–50.

Qualls, R.G. and Haines, B. L. 1991. Geochemistry of dissolved organic nutrients in water percolating through a forest ecosystem. Soil Science Society of America Journal 55, 1111–1123.

R Core Team. 2013. R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria. URL http://www.R-project.org/

Sanderman, J., Baldock, J.A., and Amundson, R. 2008. Dissolved organic carbon chemistry and dynamics in contrasting forest and grassland soils. Biogeochemistry 89, 181–198.

Sanderman, J., Lohse, K.A., Baldock, J.A., and Amundson, R. 2009. Linking soils and streams: Sources and chemistry of dissolved organic matter in a small coastal watershed. Water Resources Research 45, 1–13.

Scheibe, A. and Gleixner, G. 2014. Influence of litter diversity on dissolved organic matter release and soil carbon formation in a mixed beech forest. PloS One 9, e114040.

Scott, E.E. and Rothstein, D.E. 2014. The dynamic exchange of dissolved organic matter percolating through six diverse soils. Soil Biology and Biochemistry 69, 83–92.

Sollins, P. 1982. Input and decay of coarse woody debris in coniferous stands in western Oregon and Washington. Canadian Journal of Forest Research 12, 18–28.

Sulzman, E.W., Brant, J.B., Bowden, R.D., and Lajtha, K. 2005. Contribution of aboveground litter, belowground litter, and rhizosphere respiration to total soil CO2 efflux in an old growth coniferous forest. Biogeochemistry 73, 231–256.

Thevenot, M., Dignac, M.-F., and Rumpel, C. 2010. Fate of lignins in soils: A review. Soil Biology and Biochemistry 42, 1200–1211.

Tipping, E., Chamberlain, P.M., Fröberg, M., Hanson, P.J., Jardine, P.M. 2012. Simulation of carbon cycling, including dissolved organic carbon transport, in forest soil locally enriched with ^{14}C. Biogeochemistry 108, 97-107.

Toosi, E.R., Schmidt, J.P., and Castellano, M.J. 2014. Land use and hydrologic flowpaths interact to affect dissolved organic matter and nitrate dynamics. Biogeochemistry 120, 89–104.

van Hees, P.A.W., Jones, D.L., Finlay, R., Godbold, D.L., and Lundström, U.S. 2005. The carbon we do not see—the impact of low molecular weight compounds on carbon dynamics and respiration in forest soils: a review. Soil Biology and Biochemistry 37, 1–13.

Weishaar, J.L., Aiken, G.R., Bergamaschi, B.A., Fram, M.S., Fujii, R., and Mopper, K. 2003. Evaluation of specific ultraviolet absorbance as an indicator of the chemical composition and reactivity of dissolved organic carbon. Environmental Science and Technology 37, 4702–4708.

Wilson, H.F., Xenopoulos, M.A. 2009. Effects of agricultural land use on the composition of fluvial dissolved organic matter. Nature Geoscience 2, 37-41.

Yano, Y., Lajtha, K., Sollins, P., and Caldwell, B.A. 2005. Chemistry and dynamics of dissolved organic matter in a temperate coniferous forest on Andic soils: effects of litter quality. Ecosystems 8, 286–300.

Yarwood, S., Brewer, E., Yarwood, R., Lajtha, K., and Myrold, D. 2013. Soil microbe active community composition and capability of responding to litter addition after 12 years of no inputs. Applied and Environmental Microbiology 79, 1385–92.

Bibliography

Akagi, J., Zsolnay, Á., and Bastida, F. 2007. Quantity and spectroscopic properties of soil dissolved organic matter (DOM) as a function of soil sample treatments: Air-drying and pre-incubation. Chemosphere 69, 1040–6.

Asakawa, D., Mochizuki, H., Yanagi, Y., Suzuki, T., Nagao, S., and Fujitake, N. 2006. Changes in elemental composition, molecular weight and ^{1}H NMR spectra of the water-extractable hydrophobic acid fraction in Cambisol with season and soil depth. Soil Science and Plant Nutrition 52, 361–370.

Banaitis, M.R., Waldrip-Dail, H., Diehl, M.S., Holmes, B.C., Hunt, J.F., Lynch, R.P., and Ohno, T. 2006. Investigating sorption-driven dissolved organic matter fractionation by multidimensional fluorescence spectroscopy and PARAFAC. Journal of Colloid and Interface Science 304, 271–6.

Bengtson, P., and Bengtsson, G. 2007. Rapid turnover of DOC in temperate forests accounts for increased CO_2 production at elevated temperatures. Ecology Letters 10, 783–790.

Berg, B., Hannus, K., Popoff, T., and Theander, O. 1982. Changes in organic chemical components of needle litter during decomposition. Long-term decomposition in a Scots pine forest. I. Canadian Journal of Botany 60, 1310–1319.

Bourbonniere, R.A. and Creed, I.F. 2006. Biodegradability of dissolved organic matter extracted from a chronosequence of forest-floor materials. Journal of Plant Nutrition and Soil Science 169, 101–107.

Boyer, J.N. and Groffman, P.M. 1996. Bioavailability of water extractable organic carbon fractions in forest and agricultural soil profiles. Soil Biology and Biochemistry 28, 783–790.

Brant, J.B., Sulzman, E.W., and Myrold, D.D. 2006. Microbial community utilization of added carbon substrates in response to long-term carbon input manipulation. Soil Biology and Biochemistry 38, 2219–2232.

Bundt, M., Widmer, F., Pesaro, M., Zeyer, J., and Blaser, P. 2001. Preferential flow paths: Biological ‘hot spots’ in soils. Soil Biology and Biochemistry 33, 729–738.

Chow, A.T., Guo, F., Gao, S., Breuer, R., and Dahlgren, R.A. 2005. Filter pore size selection for characterizing dissolved organic carbon and trihalomethane precursors from soils. Water Research 39, 1255–1264.

Cole, J.J., Prairie, Y.T., Caraco, N.F., McDowell, W.H., Tranvik, L.J., Striegl, R.G., Duarte, C.M., Kortelainen, P., Downing, J.A., Middelburg, J. J., and Melack, J. 2007. Plumbing the global carbon cycle: Integrating inland waters into the terrestrial carbon budget. Ecosystems 10, 172–185.

Corvasce, M., Zsolnay, A., D'Orazio, V., Lopez, R., and Miano, T.M. 2006. Characterization of water extractable organic matter in a deep soil profile. Chemosphere 62, 1583–1590.

Cory, R.M., and Mcknight, D.M. 2005. Fluorescence Spectroscopy Reveals Ubiquitous Presence of Oxidized and Reduced Quinones in Dissolved Organic Matter. Environmental Science and Technology *39*, 8142–8149.

Cory, R.M., Miller, M.P., Mcknight, D.M., Guerard, J.J., and Miller, P.L. 2010. Effect of instrument-specific response on the analysis of fulvic acid fluorescence spectra. Limnology and Oceanography: Methods 67–78.

Crow, S.E., Lajtha, K., Bowden, R.D., Yano, Y., Brant, J.B., Caldwell, B.A., and Sulzman, E.W. 2009. Increased coniferous needle inputs accelerate decomposition of soil carbon in an old-growth forest. Forest Ecology and Management 258, 2224–2232.

Deb, S.K., and Shukla, M.K. 2011. A review of dissolved organic matter transport processes affecting soil and environmental quality. Journal of Environmental & Analytical Toxicology 1, 1–11.

Dixon, J. 2003. Applying GIS to soil-geomorphic landscape mapping in the Lookout Creek Valley, Western Cascades, Oregon. M.S. Thesis. Oregon State University, Corvallis, OR.

Fekete, I., Kotroczó, Z., Varga, C., Nagy, P.T., Várbíró, G., Bowden, R.D., Tóth, J.A., Lajtha, K. 2014. Alterations in forest detritus inputs influence soil carbon concentration and soil respiration in a Central-European deciduous forest. Soil Biology and Biochemistry 74, 106–114.

Fontaine, S., Barot, S., Barré, P., Bdioui, N., Mary, B., and Rumpel, C. 2007. Stability of organic carbon in deep soil layers controlled by fresh carbon supply. Nature 450, 277–80.

Fröberg, M., Berggren, D., Bergkvist, B., Bryant, C., and Knicker, H. 2003. Contributions of Oi, Oe and Oa horizons to dissolved organic matter in forest floor leachates. Geoderma 113, 311–322.

Fröberg, M., Kleja, D.B., Bergkvist, B., Tipping, E., and Mulder, J. 2005. Dissolved organic carbon leaching from a coniferous forest floor – a field manipulation experiment. Biogeochemistry 75, 271–287.

Fröberg, M., Kleja, D.B., and Hagedorn, F. 2007. The contribution of fresh litter to dissolved organic carbon leached from a coniferous forest floor. European Journal of Soil Science 58, 108–114.

Fellman, J.B., D'Amore, D.V., Hood, E., and Boone, R.D. 2008. Fluorescence characteristics and biodegradability of dissolved organic matter in forest and wetland soils from coastal temperate watersheds in southeast Alaska. Biogeochemistry 88, 169–184.

Fellman, J.B., Hood, E., and Spencer, G M. 2010. Fluorescence spectroscopy opens new windows into dissolved organic matter dynamics in freshwater ecosystems: A review. Limnology and Oceanography 55, 2452–2462.

Gabor, R.S., Eilers, K., McKnight, D.M., Fierer, N., and Anderson, S.P. 2013. From the litter layer to the saprolite: Chemical changes in water-soluble soil organic matter and their correlation to microbial community composition. Soil Biology and Biochemistry October, 1–11.

Gielen, B., Neirynck, J., Luyssaert, S., and Janssens, I.A. 2011. The importance of dissolved organic carbon fluxes for the carbon balance of a temperate Scots pine forest. Agricultural and Forest Meteorology, 151, 270–278.

Giesler, R., Högberg, M.N., Strobel, B.W., Richter, A., Nordgren, A., and Högberg, P. 2007. Production of dissolved organic carbon and low-molecular weight organic acids in soil solution driven by recent tree photosynthate. Biogeochemistry 84, 1–12.

Guggenberger, G., and Kaiser, K. 2003. Dissolved organic matter in soil: challenging the paradigm of sorptive preservation. Geoderma 113, 293–310.

Guggenberger, G., Zech, W., Shulten, H.-R., 1994. Formation and mobilization pathways of dissolved organic matter: evidence from chemical structural studies of organic matter fractions in acid forest floor solutions. Organic Geochemistry 21, 51–66.

Guigue, J., Mathieu, O., Lévêque, J., Mounier, S., Laffont, R., Maron, P.A., Navarro, N., Chateau, C., Amiotte-Suchet, P., and Lucas, Y. 2014. A comparison of extraction procedures for water-extractable organic matter in soils. European Journal of Soil Science 65, 520–530.

Hagedorn, F., Saurer, M., and Blaser, P. 2004. A ^{13}C tracer study to identify the origin of dissolved organic carbon in forested mineral soils. European Journal of Soil Science 55, 91–100.

Hassouna, M., Théraulaz, F., and Massiani, C. 2012. Production and elimination of water extractable organic matter in a calcareous soil as assessed by UV/Vis absorption and fluorescence spectroscopy of its fractions isolated on XAD-8/4 resins. Geoderma 189-190, 404–414.

Hongve, D., van Hees, P.A.W., and Lundstrom, U.S. 2000. Dissolved components in precipitation water percolated through forest litter. European Journal of Soil Science 51, 667–667.

Hothorn, T., Bretz, F., and Westfall, P. 2008. Simultaneous inference in general parametric models. Biometrical Journal 50, 346--363.

Inamdar, S., Finger, N., Singh, S., Mitchell, M., Levia, D., Bais, H., Scott, D., McHale, P. 2011. Dissolved organic matter (DOM) concentration and quality in a forested mid-Atlantic watershed, USA. Biogeochemistry 108, 55–76.

Jobbágy, E.G. and Jackson, R.B. 2000. The vertical distribution of soil organic carbon and its relation to climate and vegetation. Ecological Applications 10, 423-436.

Kaiser, K., and Guggenberger, G. 2000. The role of DOM sorption to mineral surfaces in the preservation of organic matter in soils. Organic Geochemistry 31, 711–725.

Kaiser, K., and Kalbitz, K. 2012. Cycling downwards – dissolved organic matter in soils. Soil Biology and Biochemistry 52, 29–32.

Kalbitz, K. 2003. Changes in properties of soil-derived dissolved organic matter induced by biodegradation. Soil Biology and Biochemistry 35, 1129–1142.

Kalbitz, K., and Kaiser, K. 2007. Contribution of dissolved organic matter to carbon storage in forest mineral soils. Journal of Plant Nutrition and Soil Science 171, 52–60.

Kalbitz, K., Meyer, A., Yang, R., and Gerstberger, P. 2007. Response of dissolved organic matter in the forest floor to long-term manipulation of litter and throughfall inputs. Biogeochemistry 86, 301–318.

Kalbitz, K., Schmerwitz, J., Schwesig, D., and Matzner, E. 2003. Biodegradation of soil-derived dissolved organic matter as related to its properties. Geoderma 113, 273–291.

Kalbitz, K., Schwesig, D., Rethemeyer, J., and Matzner, E. 2005. Stabilization of dissolved organic matter by sorption to the mineral soil. Soil Biology and Biochemistry 37, 1319–1331.

Kalbitz, K., Solinger, S., Park, J.-H., Michalzik, B., and Matzner, E. 2000. Controls on the dynamics of dissolved organic matter in soils: A review. Soil Science 165, 277–304.

Kindler, R., Siemens, J., Kaiser,K., Walmsley, D.C., Bernhofer, C., Buchmann, N., Cellier, P., Eugster, W., Gleixner, G., Grünwald, T., Heim, A., Ibrom, A., Jones, S.K., Jones, M., Klumpp, K., Kutsch, W., Steenberg Larsen, K., Lehuger, S., Loubet, B., McKenzie, R., Moors, E., Osborne, B., Pilegaard, K., Rebmann, C., Saunders, M., Schmidt, M.W.I., Schrumpf, M., Seyfferth, J., Skiba, U., Soussana, J.-F., Sutton, M.A., Tefs, C., Vowinckel, B., Zeeman, M.J., Kaupenjohann, M. 2011. Dissolved carbon leaching from soil is a crucial component of the net ecosystem carbon balance. Global Change Biology 17, 1167–1185.

Klotzbücher, T., Kaiser, K., Stepper, C., van Loon, E., Gerstberger, P., and Kalbitz, K. 2012. Long-term litter input manipulation effects on production and properties of dissolved organic matter in the forest floor of a Norway spruce stand. Plant and Soil 355, 407–416.

Kögel-Knabner, I., Guggenberger, G., Kleber, M., Kandeler, E., Kalbitz, K., Scheu, S., Eusterhues, K., and Leinweber, P. 2008. Organo-mineral associations in temperate soils: Integrating biology, mineralogy, and organic matter chemistry. Journal of Plant Nutrition and Soil Science 171, 61–82.

Kramer, M.G., Sanderman, J., Chadwick, O.A., Chorover, J., and Vitousek, P.M. 2012. Long-term carbon storage through retention of dissolved aromatic acids by reactive particles in soil. Global Change Biology 18, 2594–2605.

Lajtha, K., Crow, S.E., Yano, Y., Kaushal, S.S., Sulzman, E., Sollins, P., and Spears, J.D.H. 2005. Detrital controls on soil solution N and dissolved organic matter in soils: a field experiment. Biogeochemistry 76, 261–281.

Lajtha, K., Bowden, R.D., and Nadelhoffer, K. 2014b. Litter and Root Manipulations Provide Insights into Soil Organic Matter Dynamics and Stability. Soil Science Society of America Journal.

Lajtha, K., F. Peterson, K. Nadelhoffer, and R. Bowden. 2013. Twenty Years of Litter and Root Manipulations: Insights into Multi-Decadal SOM Dynamics. Soil Science Society of America Journal. *In review*.

Lajtha, K., Townsend, K.L., Kramer, M.G., Swanston, C., Bowden, R.D., and Nadelhoffer, K. 2014a. Changes to particulate versus mineral-associated soil carbon after 50 years of litter manipulation in forest and prairie experimental ecosystems. Biogeochemistry 119, 341-360.

Marin-Spiotta, E., Chadwick, O.A., Kramer, M., and Carbone, M.S. 2011. Carbon delivery to deep mineral horizons in Hawaiian rain forest soils. Journal of Geophysical Research 116, G03011.

Michalzik, B., Kalbitz, K., Park, J., Solinger, S., and Matzner, E. 2001. Fluxes and concentrations of dissolved organic carbon and nitrogen–a synthesis for temperate forests. Biogeochemistry 52, 173–205.

Mikutta, R., Schaumann, G.E., Gildemeister, D., Bonneville, S., Kramer, M.G., Chorover, J., Chadwick, O.A., and Guggenberger, G. 2009. Biogeochemistry of mineral–organic associations across a long-term mineralogical soil gradient (0.3–4100 kyr), Hawaiian Islands. Geochimica et Cosmochimica Acta 73, 2034–2060.

McDowell, W.H. 2003. Dissolved organic matter in soils — future directions and unanswered questions. Geoderma 113, 179–186.

McKnight, D.M., Boyer, E.W., Westerhoff, P.K., Doran, P.T., Kulbe, T., and Andersen, D.T. 2001. Spectrofluorometric characterization of dissolved organic matter for indication of precursor organic material and aromaticity. Limnology and Oceanography 46, 38–48.

Nishimura, S., Maie, N., Baba, M., Sudo, T., Sugiura, T., and Shima, E. 2012. Changes in the quality of chromophoric dissolved organic matter leached from senescent leaf litter during the early decomposition. Journal of Environmental Quality 41, 823–33.

Park, J.H-H. and Matzner, E. 2003. Controls on the release of dissolved organic carbon and nitrogen from a deciduous forest floor investigated by manipulations of aboveground litter inputs and water flux. Biogeochemistry 66, 265–286.

Park, J.J.-H., Kalbitz, K., and Matzner, E. 2002. Resource control on the production of dissolved organic carbon and nitrogen in a deciduous forest floor. Soil Biology and Biochemistry 34, 813–822.

Parlanti, E., Wo, K., Geo, L., and Lamotte, M. 2000. Dissolved organic matter fluorescence spectroscopy as a tool to estimate biological activity in a coastal zone submitted to anthropogenic inputs. *31*, 1765–1781.

Perdrial, J.N., Perdrial, N., Harpold, A., Gao, X., Gabor, R., LaSharr, K., and Chorover, J. 2012. Impacts of Sampling Dissolved Organic Matter with Passive Capillary

Wicks Versus Aqueous Soil Extraction. Soil Science Society of America Journal 76, 2019.

Qualls, R. G. 2000. Comparison of the behavior of soluble organic and inorganic nutrients in forest soils. Forest Ecology and Management 138, 29–50.

Qualls, R.G., and Haines, B.L. 1991. Geochemistry of Dissolved Organic Nutrients in Water Percolating through a Forest Ecosystem. Soil Science Society of America Journal 55, 1111-1123.

R Core Team. 2013. R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria. URL http://www.R-project.org/

Reichstein, M., and Beer, C. 2008. Soil respiration across scales: The importance of a model-data integration framework for data interpretation. Journal of Plant Nutrition and Soil Science 171, 344–354.

Rosenfeld, C.E., McCormack, M.L., and Martinez, C.E. 2014. A novel approach to study composition of in situ produced root-derived dissolved organic matter. Soil Biology and Biochemistry 76, 1–4.

Rumpel, C., and Kögel-Knabner, I. 2010. Deep soil organic matter—a key but poorly understood component of terrestrial C cycle. Plant and Soil 338, 143–158.

Sanderman, J., Baldock, J.A., and Amundson, R. 2008. Dissolved organic carbon chemistry and dynamics in contrasting forest and grassland soils. Biogeochemistry 89, 181–198.

Sanderman, J., Lohse, K. A., Baldock, J. A., and Amundson, R. 2009. Linking soils and streams: Sources and chemistry of dissolved organic matter in a small coastal watershed. Water Resources Research 45, 1–13.

Scheibe, A. and Gleixner, G. 2014. Influence of litter diversity on dissolved organic matter release and soil carbon formation in a mixed beech forest. PloS One 9, e114040.

Schimel, D., Alves, D., Enting, I., Heimann, M., Joos, F., Raynaud, D., and Wigley, T. 1996. CO_2 and the carbon cycle, in Houghton, J. T., et al. (eds.): Climate Change 1995: The Science of Climate Change. Contribution of Working Group I to the Second Assessment Report of the IPCC. Cambridge University Press, Cambridge, 65-86.

Schlesinger W.H., Melack J.M. 1981. Transport of organic carbon in the world's rivers. Tellus 33, 172–87.

Scott, E.E. and Rothstein, D.E. 2014. The dynamic exchange of dissolved organic matter percolating through six diverse soils. Soil Biology and Biochemistry 69, 83–92.

Simpson, M.J., and Simpson, A.J. 2012. The chemical ecology of soil organic matter molecular constituents. Journal of Chemical Ecology 38, 768–84.

Sollins, P. 1982. Input and decay of coarse woody debris in coniferous stands in western Oregon and Washington. Canadian Journal of Forest Research 12, 18–28.

Strohmeier, S., Knorr, K.-H., Reichert, M., Frei, S., Fleckenstein, J.H., Peiffer, S., and Matzner, E. 2013. Concentrations and fluxes of dissolved organic carbon in runoff from a forested catchment: insights from high frequency measurements. Biogeosciences 10, 905–916.

Sulzman, E.W., Brant, J.B., Bowden, R.D., and Lajtha, K. 2005. Contribution of aboveground litter, belowground litter, and rhizosphere respiration to total soil CO2 efflux in an old growth coniferous forest. Biogeochemistry 73, 231–256.

Tarnocai, C., Canadell, J.G., Schuur, E.A.G., Kuhry, P., Mazhitova, G., and Zimov, S. 2009. Soil organic carbon pools in the northern circumpolar permafrost region. Global Biogeochemical Cycles 23, GB2023.

Thevenot, M., Dignac, M.-F., and Rumpel, C. 2010. Fate of lignins in soils: A review. Soil Biology and Biochemistry 42, 1200–1211.

Tipping, E., Chamberlain, P.M., Fröberg, M., Hanson, P.J., Jardine, P.M. 2012. Simulation of carbon cycling, including dissolved organic carbon transport, in forest soil locally enriched with ^{14}C. Biogeochemistry 108, 97-107.

Toosi, E.R., Schmidt, J.P., and Castellano, M.J. 2014. Land use and hydrologic flowpaths interact to affect dissolved organic matter and nitrate dynamics. Biogeochemistry 120, 89–104.

Torn, M.S., Trumbore, S.E., Chadwick, O.A., Vitousek, P.M., and Hendricks, D.M. 1997. Mineral control of soil organic carbon storage and turnover. Nature 389, 170–173.

van Hees, P.A.W., Jones, D.L., Finlay, R., Godbold, D.L., and Lundström, U.S. 2005. The carbon we do not see—the impact of low molecular weight compounds on carbon dynamics and respiration in forest soils: a review. Soil Biology and Biochemistry 37, 1–13.

von Lützow, M., Kögel-Knabner, I., Ekschmitt, K., Flessa, H., Guggenberger, G., Matzner, E., and Marschner, B. 2007. SOM fractionation methods: Relevance to

functional pools and to stabilization mechanisms. Soil Biology and Biochemistry 39, 2183–2207.

Wagai, R., and Sollins, P. 2002. Biodegradation and regeneration of water-soluble carbon in a forest soil: leaching column study. Biology and Fertility of Soils 35, 18–26.

Weishaar, J.L., Aiken, G.R., Bergamaschi, B.A., Fram, M.S., Fujii, R., and Mopper, K. 2003. Evaluation of specific ultraviolet absorbance as an indicator of the chemical composition and reactivity of dissolved organic carbon. Environmental Science and Technology 37, 4702–4708.

Wilson, H.F., Xenopoulos, M.A. 2009. Effects of agricultural land use on the composition of fluvial dissolved organic matter. Nature Geoscience 2, 37-41.

Yano, Y., Lajtha, K., Sollins, P., and Caldwell, B.A. 2004. Chemical and seasonal controls on the dynamics of dissolved organic matter in a coniferous old-growth stand in the Pacific Northwest , USA. Biogeochemistry 71, 197–223.

Yano, Y., Lajtha, K., Sollins, P., and Caldwell, B.A. 2005. Chemistry and dynamics of dissolved organic matter in a temperate coniferous forest on Andic soils: effects of litter quality. Ecosystems 8, 286–300.

Yano, Y., Mcdowell, W.H., and Aber, J.D. 2000. Biodegradable dissolved organic carbon in forest soil solution and effects of chronic nitrogen deposition 32, 1743–1751.

Yarwood, S., Brewer, E., Yarwood, R., Lajtha, K., and Myrold, D. 2013. Soil microbe active community composition and capability of responding to litter addition after 12 years of no inputs. Applied and Environmental Microbiology 79, 1385–92.

Zsolnay, Á., Baigar, E., Jimenez, M., Steinweg, B., Saccomandi, F. 1998. Differentiating with Fluorescence spectroscopy the sources of dissolved organic matter in soils subjected to drying. Chemosphere 38, 45–50.

Zsolnay, Á. 2003. Dissolved organic matter: artefacts, definitions, and functions. Geoderma, 113, 187–209.